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# Diversity and assembly processes of guyanese freshwater fish assemblages

Kévin Cilleros

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# THÈSE

En vue de l'obtention du

## DOCTORAT DE L'UNIVERSITÉ DE TOULOUSE

Délivré par : l'Université Toulouse 3 Paul Sabatier (UT3 Paul Sabatier)

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Présentée et soutenue le 04/12/2017 par :

**Kévin CILLEROS**

**Diversité et règles d'assemblage des communautés de poissons d'eau douce de Guyane**

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PhD thesis

# **Diversity and assembly processes of Guyanese freshwater fish assemblages**

Kévin CILLEROS

## Jury composition

Núria BONADA	Associate Professor	Referee
Catherine GRAHAM	Professor	Referee
Christopher BARALOTO	Professor	Jury member
Gaël GRENOUILLET	Senior lecturer	Jury member
Thierry OBERDORFF	Researcher	Jury member
Sébastien BROSSE	Professor	PhD supervisor

**Defended on December 4, 2017**



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# Chapter I: Introduction

## I.1 Biodiversity: from pattern to process

### I.1.1 The historical roots of biological diversity

Diversity, the range of variation in a set of features, has long been described for biological organisms. The first known written facts date back to Aristotle in his *History of Animals* (350 BCE). He recognized that some animals have the same characteristics, whereas others are different, both in their morphology (or in their “parts”, Aristotle, 1910) and in their behavior (“in their modes of subsistence, in their actions, in their habits”, Aristotle, 1910). This pioneering work of zoology has ruled, without improvement, until the xvi<sup>th</sup> century (Barthélemy-Saint-Hilaire, 1883), when two French zoologists, Pierre Belon and Guillaume Rondelet, pursued Aristotle’s work and travelled around the Mediterranean Basin to describe plants and animals (especially aquatic animals; Belon, 1555; Rondelet, 1558). The next progresses occurred during the xviii<sup>th</sup> century with the works of Carl Linnaeus, widely known for its biological classification of organisms, which principles are still used in taxonomy. Georges L. Leclerc, Comte de Buffon, then linked observations and description of species with the history of Earth to explain some of the patterns he observed that set first principle in biogeography (Buffon’s Law that states that geographically isolated areas with similar environment have different species, Buffon, 1761). A tipping point occurred during the xix<sup>th</sup> century, with the works of Charles Darwin, where the classical natural observational approach faded to the benefit of a more inductive or ‘scientific’ approach. From here, biological sciences benefited from the emergence of new fields (evolution, genetic, ecology) and the fortification of previous theories in biogeography (e.g. Alexander von Humboldt’s or Alfred R. Wallace’s works). However it was not until the second half of the xx<sup>th</sup> century that the term of “biological diversity” emerged from the field of conservation biology. The term biological diversity was first used by Thomas Lovejoy (1980) to refer to species diversity in tropical forests, and its contraction into “**biodiversity**” appeared soon after, for the “National Forum on BioDiversity” in 1986 organized by Walter G. Rosen and the publication of the proceedings of this forum under the name “BioDiversity” (Wilson & Peter, 1988), then started its popularity among scientists, politics and people.

### I.1.2 The current definition of biodiversity

Biodiversity can be defined as “the variety of life, at all levels of organization, classified both by evolutionary (phylogenetic) and ecological (functional) criteria” (Colwell, 2012). Colwell’s definition of biodiversity can be decomposed into three parts. The first part, “the variety of life” is synonymous to biological diversity and does not inform much on what is understood when speaking about “life”. It’s the second element of the definition that explicit it: the expression “levels of organization” refers to the nested organization

of life from genes to ecosystems. From this, “life” will have a different meaning at each of these levels. For example, the diversity of species at the **community** or **assemblage** level is the most known descriptor of biodiversity. Biodiversity can also be described at the gene level using the diversity of alleles or genes (genetic polymorphism), or at the population level with the diversity in individual genotypes and phenotypes. To determine those of biodiversity descriptors, Colwell’s definition lies on evolutionary and ecological characteristics. These characteristics have been commonly used to delineate species, but they can also apply to other levels of organization (genetic lineage, individuals, functional groups or landscapes).

### 1.1.3 Biodiversity at the community level

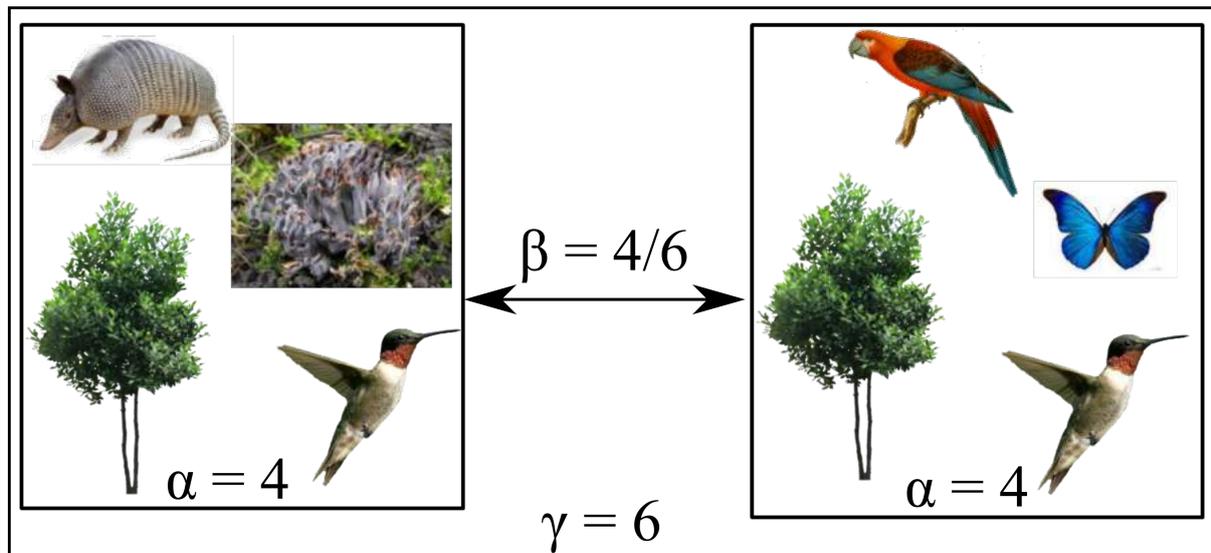
Using Colwell’s definition, species can be used as the unit of diversity at the community level. If we consider a set of communities, a first and simple diversity descriptor will be the list or number of species, also called species richness, in each community (Figure I.1). We can also look if all species have the same abundance in each community or if some species dominate the community. In this case, the equitability or evenness of the community is used as a descriptor of diversity. Together, species richness and equitability reflect community composition and represent community diversity.

These measures of community diversity apply to each community and represent within-community diversity, or  **$\alpha$ -diversity**. Summarizing communities using their species richness and evenness allows comparisons across communities but does not detect potential differences in the species compositions between communities. For instance, in Figure I.1, the two communities have each 4 species ( $\alpha$ -diversity = 4) but they do not share all their species (only two are shared). One way to deal with this is to compare communities and look at between-community diversity, or  **$\beta$ -diversity**. A classical measure of  $\beta$ -diversity is the percentage of species that are shared (or not shared) between communities (e.g. Jaccard, 1912 or Sørensen, 1948 indices). In the Figure I.1, the two communities have 2 species in common and 4 species are only found in one of the two communities. So, the diversity between the two communities is  $\frac{4}{6} \approx 0.67$ . This value means that the two communities share about 33% of the total number of species found in these communities. This type of metrics can be extended to several communities and some metrics were developed to take into account species abundances [Bray-Curtis (Bray & Curtis, 1957) or Manhattan measures (Michener & Sokal, 1957)].

Finally, we can look at the total diversity across all the communities or  **$\gamma$ -diversity**. It reflects information given by both  $\alpha$ - and  $\beta$ -diversity measures. For example, if communities share a lot of species, the total diversity tends to be close to the maximal value of  $\alpha$ -diversity. In contrast,  $\gamma$ -diversity will be high if each community has a unique set of species.

These terms of  $\alpha$ -,  $\beta$ - and  $\gamma$ -diversity were firstly introduced by Whittaker (1960) and several definitions and metrics have followed (Whittaker, 1972; Anderson *et al.*, 2011) but all converge towards the idea that within-, between- and total diversity

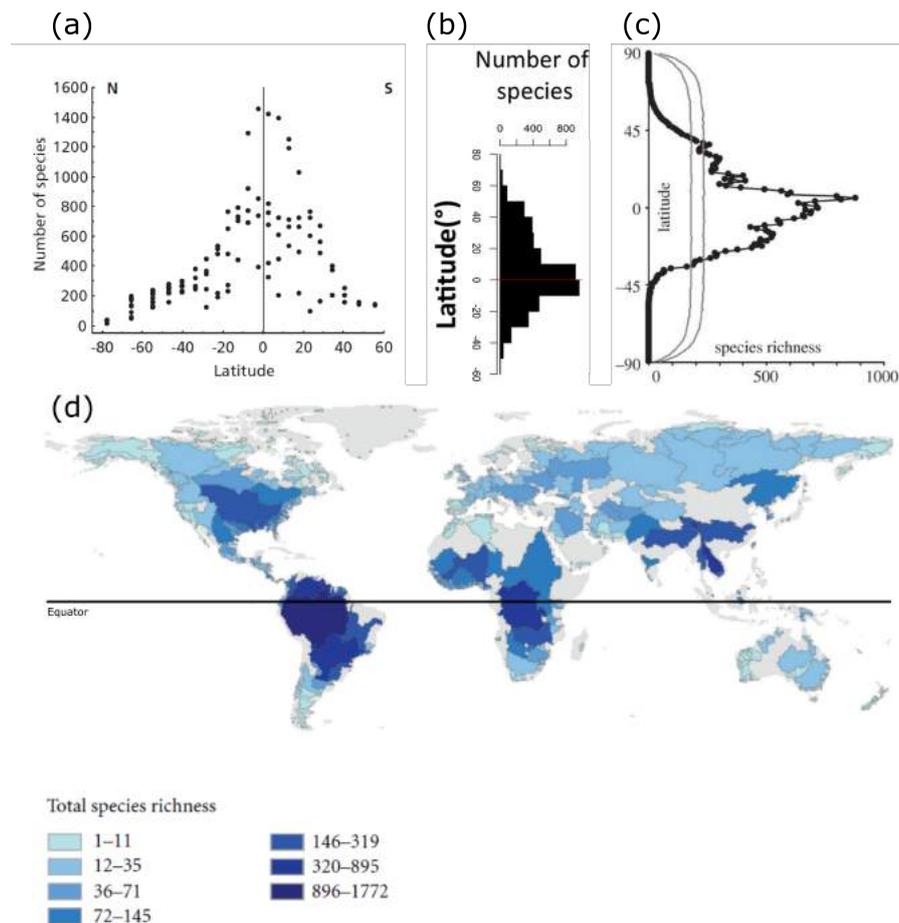
represent different facets of biodiversity, that provide complementary information to describe patterns of diversity.



**Figure I.1:** Biodiversity can be spatially decomposed in 3 levels: the diversity of a local assemblage or habitat ( $\alpha$ -diversity), the difference in diversity between two or more assemblages or habitats ( $\beta$ -diversity), and the total diversity of assemblages or habitats. In the example above, each community is composed of 4 different species (thus  $\alpha$ -diversity = 4 for each). If we compare the 2 communities, we see that they share two species and each community has 2 unique species. The 2 communities thus share 2 species in common over 6 species, that is  $\frac{2}{6} \approx 33\%$ , and the difference in diversity between the two communities ( $\beta$ -diversity) is  $1 - \frac{2}{6} = \frac{4}{6} \approx 67\%$ . In total, the diversity in these communities is equal to 6 species ( $\gamma$ -diversity)

#### I.1.4 The patterns of biodiversity

Diversity is not evenly distributed across the globe and thus exhibits spatial variability, from large to local spatial scales. At the global scale, the best studied biodiversity pattern is the latitudinal diversity gradient. Several authors have reported that a high number of species occur around the equator, and that their number decrease toward the poles, causing a hump shaped relationships between species richness and latitude (Figure I.2). This pattern is shared by most organisms (woody plants, birds, freshwater fish) despite some variability in the position of the peak of richness and the rate of richness decrease with increasing or decreasing latitude (Hillebrand, 2004). Diversity also differs between continents and regions for a same latitudinal range. For example, the number of freshwater fish species in South America is 1.3 times higher to that of Africa (Lévêque *et al.*, 2007). But the most striking difference between these regions is the difference in the identity of species. Such difference in species composition between regions is among the most striking biogeographical patterns, and was used to define the biogeographic realms in early biogeographic studies from the XIX<sup>th</sup> century (Wallace, 1876; Sclater, 1858; Engler, 1879). Within a region, local diversity is also unequally distributed between the different ecosystems and habitat features, with patterns and gradients highly variable. Forest ecosystems do not have the same communities (group of directly or indirectly interacting species, co-occurring



**Figure 1.2:** The latitudinal diversity gradient is described in several taxa, from New World birds (a), mammals (b), amphibians (c) and freshwater fishes (d). Figures from Gaston & Blackburn (2008), Rolland *et al.* (2014), Pyron *et al.* (2015) and Oberdorff *et al.* (2011) respectively

in space and time) and the same species richness as grassland ecosystems. In freshwater ecosystems, within a drainage basin, species richness increases from small upstream sites to large rivers and freshwater communities change along this gradient, with species associated to headwater streams and others endemic of large and deep channels (Huet's zonation (1959) and River Continuum Concept from Vannote *et al.*, 1980). Understanding what shape these patterns remains an outstanding key question in ecology and requires combining several research fields (phylogeography, niche and spatial modelling, evolutionary biology). The improvement of modeling techniques and the development of new methods and tools of have accompanied the rapid and vast gathering of biodiversity data (via automatic recording devices, high-throughput DNA sequencing, citizen science; Bush *et al.*, 2017) and their availability through online databases [GBIF (<http://gbif.org>), GenBank (Benson *et al.*, 2017), The Ocean Biogeographic Information System (Grassle, 2000)] allow to tackle this question.

### 1.1.5 A diversity of processes

The processes that shape the spatial patterns of biodiversity are scale dependent, although often not exclusive to a given spatial (or temporal) scale. A process that

explains the global variation of species richness will therefore have a low influence on local diversity variation. For instance, the latitude is highly influential at the global scale, but it hardly affects local biodiversity variation. The reverse for more local processes also holds (e.g. interactions between species occur at a local scale and hardly affect diversity gradients measured at higher spatial scales). We can thus sort these processes according to the spatial scale considered. This led to formalize these processes as hierarchical filters (Tonn, 1990; Poff, 1997) acting at global, regional or local scales on a set of candidate species (or **pool of species**). Those filters therefore select the species occurring at a given scale from a set of potential species co-occurring at higher scales (Srivastava, 1999).

### **Global scale processes**

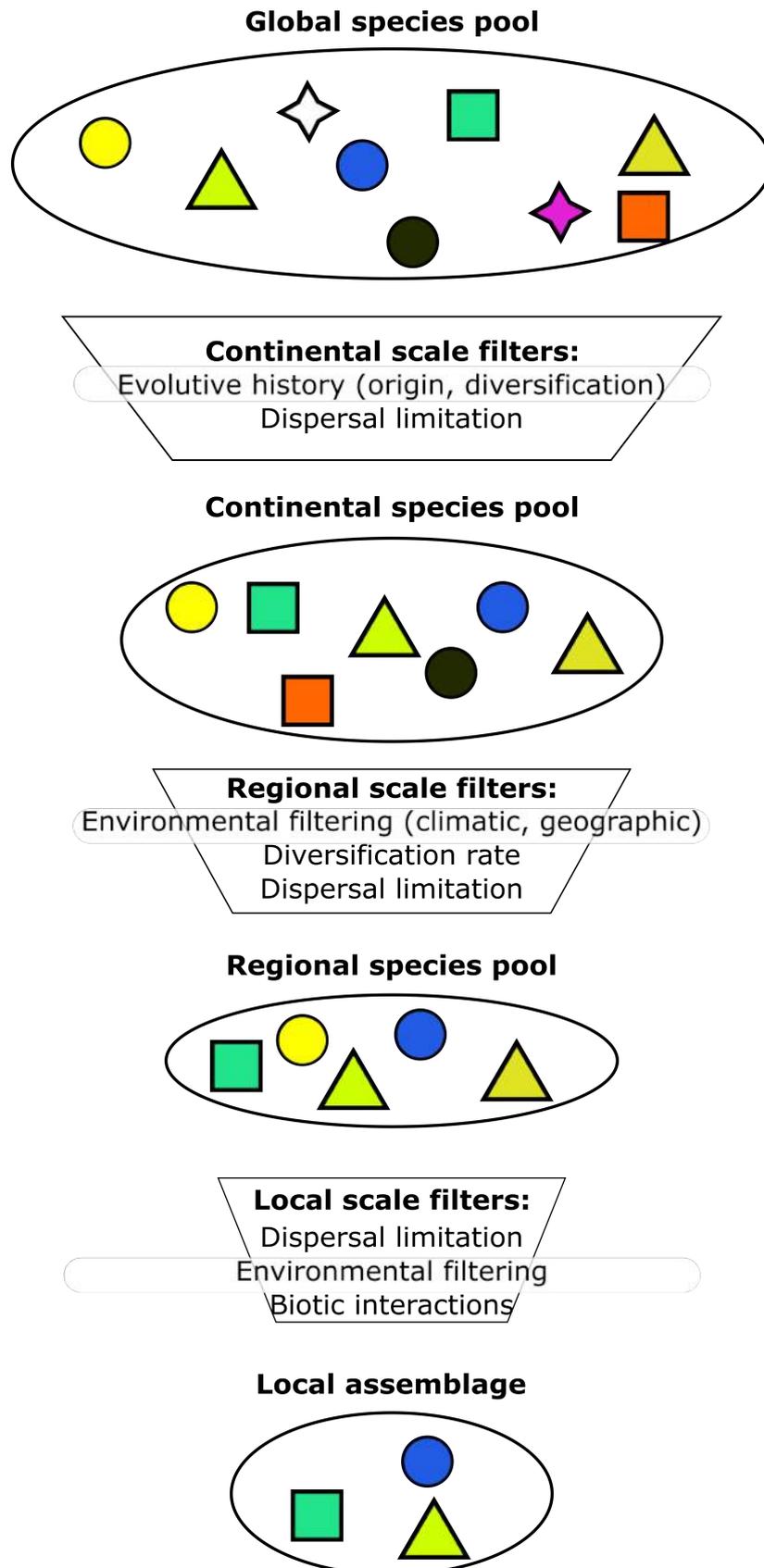
At the global scale, four main non-exclusive groups of hypotheses have been proposed to explain the latitudinal diversity gradient (LDG).

First, the observed pattern might result from random distribution of species range along a latitudinal axis bounded at the two poles, without the need of any environmental effect. This geometric rule predicts that majority of species midpoint range will fall near the centre of the axis, corresponding to tropical regions (the mid-domain effect; Figure I.4, Colwell & Hurtt, 1994).

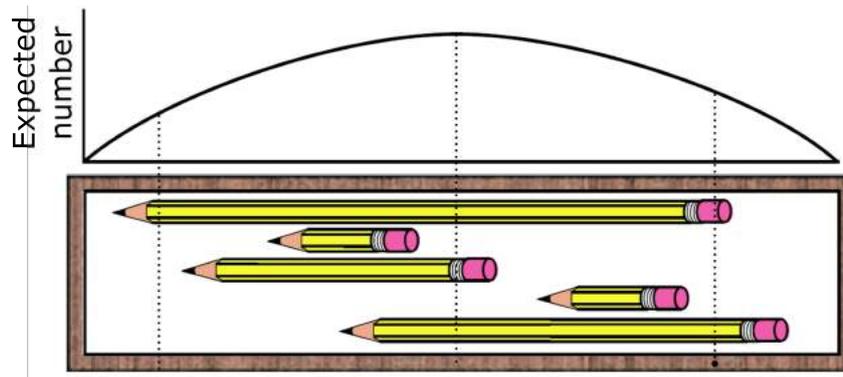
Aside from this neutral hypothesis, geographical differences between tropical and temperate regions can also contribute to the latitudinal diversity gradient. Tropical regions form a continuous entity, whereas temperate and polar regions are distributed part aside of it and form smaller entities. Larger areas are associated with higher species richness, through a greater speciation rate, a lower extinction rate and higher diversity of habitat (species-area relationship, Preston, 1962).

Tropical regions also receive higher solar radiation that can translate in higher primary productivity and in higher climatic stability and lower seasonality variability (Hutchinson, 1959; Hawkins & Porter, 2003; Currie *et al.*, 2004). The species-energy relationship has been criticized because experimental studies showed that the species richness indeed increase with energy, but stops increasing or decreases after a threshold, and it thus might only be applicable under particular conditions (Hurlbert & Stegen, 2014).

A third group of hypotheses deals with the biogeographic historic differences between tropical and temperate regions, and associated evolutionary consequences. These hypotheses stipulate that diversity patterns are caused by differential speciation and extinction rates between tropical and temperate regions (Rolland *et al.*, 2014). Two mechanisms have been proposed to explain those differences: either the tropical regions are older and more species originated from those areas (Wiens *et al.*, 2011), or diversification rates are higher in the tropical regions compared to those observed in the temperate regions (Mittelbach *et al.*, 2007; Pyron & Wiens, 2013). Some studies also suggested the existence of phylogenetic tropical niche conservatism, the tendency of tropical species to retain ecological traits related to



**Figure I.3:** Processes structuring local assemblages. The processes can be considered as a succession of filters that act at different spatial scales (continental, regional, local scales). Symbols represent species



**Figure 1.4:** Illustration of the mid-domain effect with an analogy, where species range are represented by pencils, enclosed in a bounded box. If pencil positions are randomly distributed, the expected number of overlapping pencils is greater in the middle of the box rather at its edges. Adapted from Colwell *et al.* (2016)

tropical environment (Wiens & Donoghue, 2004). This would limit their dispersal capacity to regions outside of the tropics (**dispersal limitation**). Recently, Morinière *et al.* (2016) proposed a similar mechanism for taxa that present inverse latitudinal gradients, based on temperate niche conservatism for lineages originating from temperate regions.

More recently, biotic interactions, although acting mainly at local scales, has regained importance as a mechanism that can affect global scale diversity patterns (Schemske *et al.*, 2009; Pianka, 1966). The relative stability of the climate over geological times for tropical areas allows the establishment of numerous interactions between species. In addition, this stability makes the environment more predictable and therefore relaxes the strength of selection by abiotic factors. In contrast, abiotic environment is the main constraint to species evolution in temperate regions and its variability selects for more generalist species (Stevens, 1989). The shift from the dominance of abiotic constraints to the dominance of biotic constraints in tropical areas makes the opportunity for co-evolution to be a strong driver of speciation, where optimum phenotypes always change, facilitating adaptation and speciation Schemske *et al.*, 2009; Pianka, 1966. Biotic interactions include the effect of competition that promotes speciation through niche specialization. It also includes predation, which strength in tropical ecosystems (due to the high number of predator species) can reduce the competitive exclusion of prey species and thus enhance diversity (Janzen, 1970; Connell, 1971).

Despite the apparent separation of geometrical, geographical, historical and biotic hypotheses, they are not exclusive and together contribute in shaping diversity at the global scale.

### Regional scale processes

At the regional scale, the pool of species is constrained by biogeographic, climatic and geographic variations within the considered region. Similarly to the global processes, differences in diversification rates between regions caused by geologic events can

cause difference in diversity between regions located under same latitudes. For example, ancient refuge areas during glaciation or marine incursion periods have higher species richness and endemism than other areas (Svenning & Skov, 2007; Lawes *et al.*, 2007; Reyjol *et al.*, 2007). Climatic and landscape features also restrict the identity of species that occur in a region. This combination of environmental characteristics and biotic component (the species present) have been used to define ecoregions (area with relative homogeneity of ecosystems), even if recent definitions put the emphasis more on the biotic component (Olson *et al.*, 2001; Abell *et al.*, 2008).

### Local scale processes

At the local scale, two kinds of processes, or **assembly rules**, have been proposed to determine diversity patterns, deterministic processes and neutral processes, which represent the two extremes of a spectrum.

Deterministic or niche-based processes put the emphasis on the role of abiotic and biotic environment on community composition that successively act on the species pool to define occurring species. First, species that can establish and persist under given abiotic conditions (e.g. climate, temperature, soil characteristic...) will constitute a potential set of species. This process is referred as **environmental filtering**. Then, biotic interactions between species will restrain this set of species to the observed occurring species. Traditionally, competition is considered as the main biotic constraint that will prevent species with similar ecology to co-occur (a process also called **limiting similarity**), but predation and facilitation also affect community composition. A first approach to make the distinction between abiotic and biotic constraints on species assemblages was through the use of the ratio between the number of genera found in a community and the number of species (Elton, 1946). A low species-to-genus ratio indicates that the community is composed of relatively distantly related species and is expected limiting similarity is the dominant process. On the contrary, a higher ratio is expected under environmental filtering. Its use was however greatly criticized as it highly depends of the sample size, with the species-to-genus ratio increasing with an increasing number of species (Gotelli & Colwell, 2001; Jarvinen, 1982).

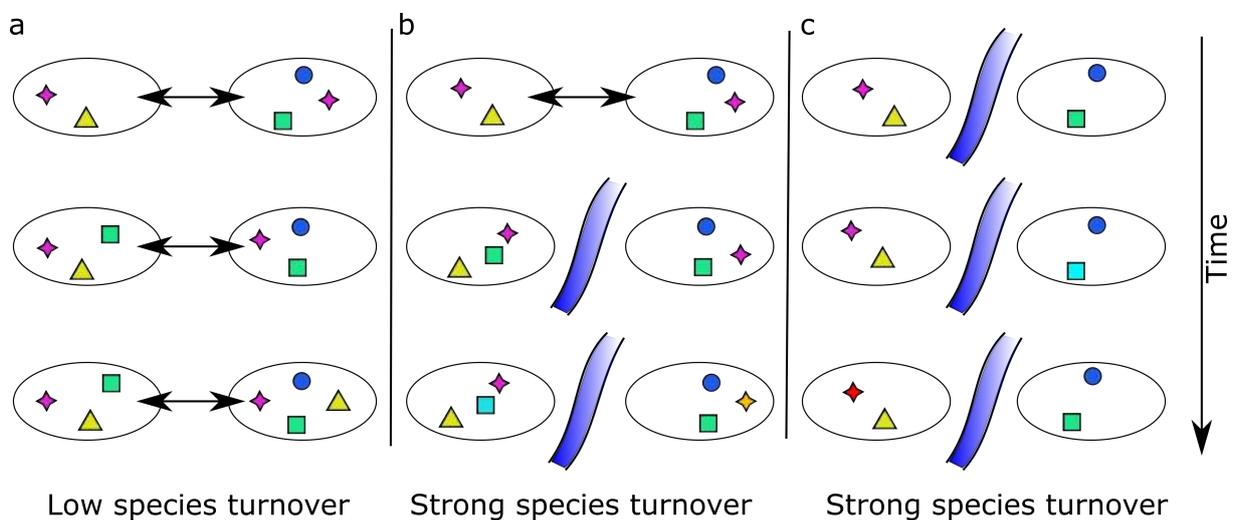
At the other end of the spectrum, the neutral processes root in Hubbell's neutral theory (Hubbell, 2011) where all individuals and species are functionally equal, so the environment impacts them equally, and only dispersal capacity and survival affect community composition. It is a dynamic equilibrium between migration from the species pool to local communities and local ecological drift that shape the community composition. Proposed as an alternative to more deterministic models, neutral hypotheses are currently used as a null model to establish random expectations against which the observed patterns are tested. Differences between these expectations and the observed patterns are often interpreted as the sign of the effect of deterministic processes in community assembly.

## I.1.6 Unlocking the “Same pattern from several processes” problem

### The process-from-pattern drawbacks

As explained above, the interpretation of clustering patterns as the result of local environmental filtering and overdispersed patterns as the result of local competition served as a basis to measure the strength of environmental and biotic processes in community assembly, but combinations of several processes can lead to the same pattern. For instance, if distant related species have converged toward the same ecology (niche convergence) and a strong environmental filtering act locally, the species-to-genus ratio will erroneously indicate that competition is the main process structuring the community. Similarly, competition between species can result in both clustering and overdispersed pattern (Mayfield & Levine, 2010). For example, if soil type is the main driver of competition between plant species and if soil preference is phylogenetically conserved, close related species will mostly compete between themselves, leading to an overdispersed pattern. However, if light is the main driver of competition, taller species will outcompete the smaller ones and if height is conserved, competition will drive clustering.

The same problem can arise for dispersal limitation between communities. It is indeed difficult to differentiate the effect of historical dispersal limitation and recent dispersal limitation without knowledge of the studied system (Figure I.5).



**Figure I.5:** Recent and historical dispersal limitation can cause the same observed pattern of taxonomic turnover between two communities (ellipses) made of two to four species (symbols). a: No dispersal limitation between the two communities. The assemblages can exchange species and it results in a low species turnover between the two assemblages. b: Recent dispersal limitation. The two assemblages have been connected but have been recently separated by a geographic barrier (e.g. a river). Species can evolve in each assemblage and it results in a strong turnover between the two assemblages. c: Historic dispersal limitation. The two assemblages have historically been separated and species have evolved within each assemblage. It causes a strong turnover of species between assemblages

## **Incorporating functional and phylogenetic information**

In the last decades, an increasing consensus emerged highlighting that biodiversity is not only the identity of species, but also encompasses their role in the ecosystem and their evolutionary history. Biodiversity can thus be described via the identity of the species (**taxonomic diversity**), their ecological function (**functional diversity**) or the evolutionary history they represent (**phylogenetic diversity**). This view roots in the field of biological conservation, where a great attention has been given to maintaining ecosystem functions in addition to species.

Phylogenetic diversity approaches root in the study of Vane-Wright *et al.* (1991) that proposed to use cladistic (or phylogenetic) relationships between species as an additional component of biodiversity for conservation assessment. Indeed, species are not equal and this distinctiveness can be approximated via phylogenetic diversity, that is related to both past history of species (extinction, speciation, colonization) but also to ecosystem functioning. In addition, the capacity of species to evolve, which is related with the past history of the species and their phylogenetic relationships, is linked to the ability of the species to adapt to changing environment in the context of global changes. Its use have expended through the development of statistical and modeling tools allowing to handle complex phylogenetic data and to buildup robust phylogenetic trees for large number of species (Smith *et al.*, 2009) based on genetic material from single genetic markers to complete genome (Kappas *et al.*, 2016; Jarvis *et al.*, 2014).

Species can also be considered by their role in ecosystem functioning, giving rise to functional diversity. Functional diversity can be assessed by measuring directly species impact on ecosystem processes (decomposition rate, carbon and nitrogen fluxes), or by measuring species role in ecosystems through **functional traits**. Functional traits are any traits that impacts fitness indirectly via its effects on growth, reproduction and survival (Violle *et al.*, 2007). They are thus linked to ecosystem processes. These functional traits have been frequently to characterize plant and aquatic insects communities for decades. Their extensions to others animals have only recently expended through the development of life-history traits and/or morphological attributes databases for various taxa. Similarly to phylogenetic diversity, functional diversity necessitates handling simultaneously several functional traits to functionally describe species and has benefited from methodological advances that facilitate the comparisons between taxonomic, phylogenetic and functional diversities. For instance,  $\beta$ -diversities metrics developed to measure species replacement (Jaccard or Sorensen) have now their equivalents for functional  $\beta$ -diversity (Villéger *et al.*, 2011b) and phylogenetic  $\beta$ -diversity (Leprieur *et al.*, 2012; Lozupone *et al.*, 2011). This allows having similar frameworks for the three diversity facets and therefore to analyze the relationships between those facets to better understand assembly rules.

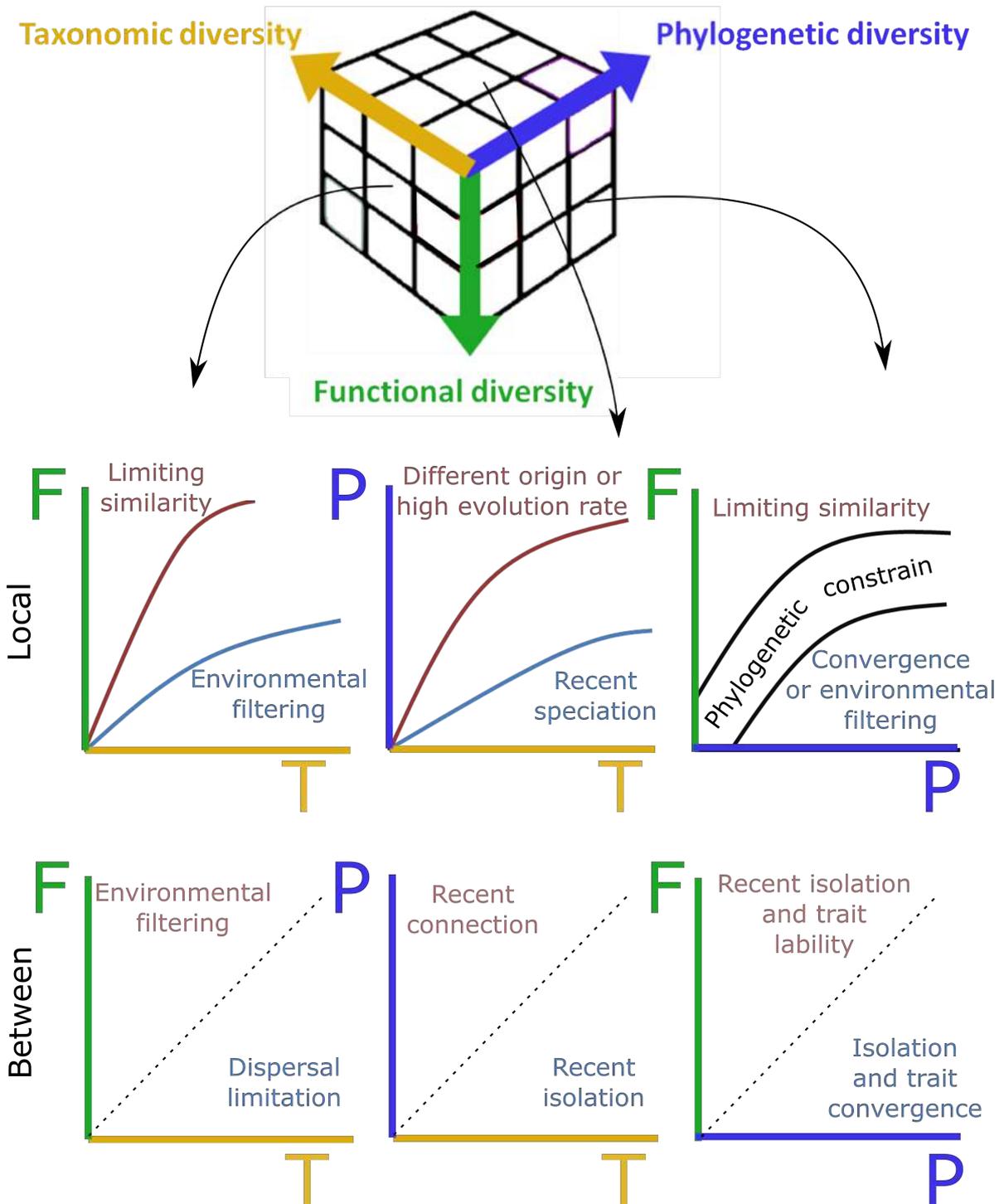
## Comparing diversity facets to understand the processes

Community composition and spatial structure are affected by both evolutionary and ecological processes. To disentangle them, the simultaneous use of the different facets of biodiversity has been proposed to overcome the “Same pattern from several processes” problem (Pavoine & Bonsall, 2011; Baraloto *et al.*, 2012; Kraft & Ackerly, 2010; Swenson & Enquist, 2009). Previous attempts focused on only on one facet (the taxonomic) or two (taxonomic and phylogenetic or taxonomic or functional). The latter cases lie on the hypothesis that functional and phylogenetic diversities are highly correlated, because functional traits are shaped through evolution. This hypothesis, that set the basis for community phylogenetic field (Webb, 2000; Webb *et al.*, 2002), is now debated, as not all traits are phylogenetically constrained, and phylogenetic **convergence** can lead to the same pattern as **phylogenetic conservatism** (Cavender-Bares *et al.*, 2004). This fact was already noticed by Webb *et al.* (2002), but trait conservatism is often assumed (and not tested), which might lead to major drawbacks.

### $\alpha$ -diversity relationships

If functional traits are conserved through the phylogeny, close related species have similar ecological attributes, and increasing taxonomic diversity of assemblages will increase (or at least not decrease) functional and phylogenetic diversities. However the different assembly processes will affect the rate at which functional or phylogenetic diversities increase compare to the increase of taxonomic diversity. In other terms, the increase of functional and phylogenetic diversities can be higher or lower than expected knowing the increase in taxonomic diversity depending on the process that mainly structure local assemblages. The expected increase can be look via the use of null models that create random communities under specific rules. There is a variety of null model, and they can be applied to taxonomic, functional and phylogenetic diversity. For example, null models applying to taxonomic diversity are based on the randomization of the abundance or the occurrence of species in communities, by redistributing individuals or species from the species pool to communities, to test if some constrains prevent species to occur in some communities (Gotelli & Graves, 1996). For functional and phylogenetic diversity, null models often permute species traits or species placement in the phylogeny to keep the observed taxonomic diversity fixed and avoid entangling the processes that affect only one diversity facet.

For example, if we look at the  $\alpha$ -diversity, increasing species richness will increase to lead to higher functional and phylogenetic richness. However, adding new species into the community with similar ecological attributes to those already present will lead to a low functional increase. Such low increase will be lower than expected under a random species selection. This can occur if environmental filtering drives local community composition (Mouillot *et al.*, 2007). The same situation occurs for phylogenetic diversity if the species derive from recent speciation: a new species that is more closely related to the species already present will not increase greatly



**Figure I.6:** Combining information on taxonomic, functional and phylogenetic diversity can help to disentangle the main processes that structure communities, both within- and between-communities. On each plot, dominant processes are indicated. For local processes ( $\alpha$ -diversity), the main expectation is that higher taxonomic diversity causes higher functional and phylogenetic diversity. Deviation from this expectation (blue and red lines) indicates that one process structure local community. Phylogenetic and functional diversity must be correlated (phylogenetic constrain or trait conservatism), but communities can deviate from it. For between-communities diversity ( $\beta$ -diversity), the more dissimilar the communities are in their species identity, the more functionally and phylogenetically dissimilar communities will be. Departure from this expectation indicates that one process is stronger than the other. Although each plot links only two facets, conclusions must be taken from the comparison of the three facets simultaneously

phylogenetic richness (Xiang *et al.*, 2004).

On the other hand, if limiting similarity rules community assembly, each species will be ecologically different to the others (as a result of competitive exclusion), and hence will greatly contribute to increasing functional richness, more than under a random species selection (Mouillot *et al.*, 2007). If species present in the community originate from different regions (following recolonization or after introduction) or if evolutionary or speciation rates are faster due to particular environmental conditions (barriers to gene flows, habitat heterogeneity) or biological attributes of species (dispersal rates, population structure), phylogenetic richness will greatly increase (Xiang *et al.*, 2004).

Lastly, the relationship between functional and phylogenetic  $\alpha$ -diversity, if deviating from the expectation that more distant species are more functionally different, can either reveals limiting similarity (if close related species occurrence causes higher functional richness than expected) or indicates dominant and strong environmental filtering that causes strong niche conservatism or trait convergence, with distant species having similar ecological traits (Safi *et al.*, 2011).

### **$\beta$ -diversity relationships**

For  $\beta$ -diversities relationships, the main expectation is that the more pairs of communities will have different species (higher  $\beta$ -diversity), the more they will be different both functionally and phylogenetically, due to increasing environmental differences and isolation by distance. Deviation from this null expectation, with one diversity facet being higher or lower than the other, can inform which processes dominate community structure.

If communities are composed of different species (high taxonomic  $\beta$ -diversity) but these species are ecologically similar (low functional  $\beta$ -diversity), this could provide hints for strong dispersal limitation between communities, preventing species to occur in both communities (Fukami *et al.*, 2005). This dispersal limitation can be caused by the presence of a barrier to the dispersion or can result from different colonization history and competitive exclusion (Fukami, 2015). On the opposite, communities with similar species composition but with some species that differ greatly in their ecological attributes can result from differences in particular environmental conditions between the two sites that select for species with specialized ecology.

Comparing taxonomic and phylogenetic  $\beta$ -diversities can help to differentiate between dispersal limitation caused by recent or ancient isolation (Weinstein *et al.*, 2014). Ancient isolations of communities result in independent evolution of communities that causes high values of dissimilarities in both diversities. If communities were recently isolated, they had the possibility to exchange species before isolation and species of the communities will be phylogenetically related. After the isolation, communities evolved independently, but not during a sufficient time to blur their past common history, thus resulting in lower phylogenetic dissimilarities than expected. On the opposite, recent connection between previously isolated assemblages can allow the exchange of some phylogenetically distant species

between communities (the one that can colonize new communities), thus reducing taxonomic dissimilarity between communities. However phylogenetic dissimilarity will stay relatively high compared to taxonomic dissimilarity.

Lastly deviations from the expected relationship between functional and phylogenetic  $\beta$ -diversities can inform on traits convergence between isolated communities in similar environmental conditions or high trait lability in recently isolated communities (Swenson *et al.*, 2012b; Yang *et al.*, 2015). This last relationship is difficult to test, as phylogenetic diversity and functional diversity are independantly computed and thus cannot be decoupled.

Here I described relationships between diversity facets two by two. However, they must all be considered altogether to have the full picture of the processes. For example, observing low dissimilarities in the 3 diversity facets will reflect communities in similar environment with no dispersal limitation, whereas high dissimilarities in taxonomic and functional diversity but lower phylogenetic dissimilarity reflects recent isolations of communities that evolve in different environments.

### **Freshwater fish as a model to study assembly rules**

This framework thus needs exhaustive information on the communities and on the species that compose them. Rich and variable communities might be promising cases to disentangle assembly processes. Within Neotropical regions, plants communities and their assembly processes have been heavenly studied (Kraft *et al.*, 2008; Swenson *et al.*, 2012a; Kraft & Ackerly, 2010; Fortunel *et al.*, 2014). In contrast, freshwater fish assemblages have long been restrained to species inventories, species-habitat relationships and anthropogenic impacts on the species. Building on those previous knowledge helps analysing assembly rules. Among freshwater organisms, fish are the best known, they benefit from a quite exhaustive taxonomic knowledge and from information (at least partial for tropical species) on species distribution. They therefore constitute an interesting biological model for studying assembly rules. Moreover, freshwater fishes are ectothermic organisms and their distribution is thus highly constrained by their environment. Moreover, their movement depends on their own capacity, but also on the configuration of the hydrological network (Landeiro *et al.*, 2011). This network also represents a closed environment for freshwater fishes, as the marine water represents barrier to their dispersal between the different drainage basins. The history of drainage basins will thus be reflected in the fish communities (Hugueny, 1989). All these characteristics thus affect how environment, space and past history structure fish assemblages and will modulate the strength of the assembly processes.

Within freshwater tropical fauna, fish from French Guiana benefited from extensive inventories in the seventies that led to the publication of the atlas of Freshwater fish from French Guiana (Planquette *et al.*, 1996; Keith *et al.*, 2000; Le Bail *et al.*, 2000) and to gather solid information about the taxonomy of this speciose fauna. This knowledge on fish taxonomy and distribution constitute the first, but essential, step to unravel the

processes that structure fish assemblages.

## **I.2 Application to French Guiana freshwater fish assemblages**

### **I.2.1 The Guianese territory**

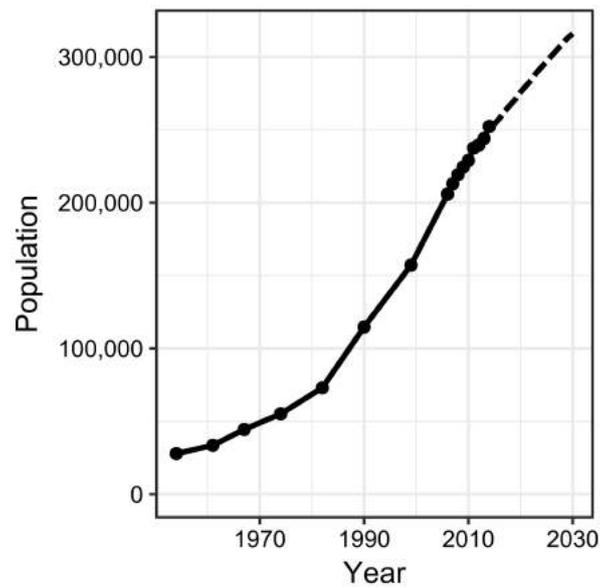
French Guiana is a French overseas department and region located on the Northwest coast of South America between Brazil and Suriname. It covers about 83,500 km<sup>2</sup>. Its coastal location close to the equator allows a relative homogeneity of the climate, with constant temperature around 25°C. Only the rainfall highly varies between months and determines two main seasons in French Guiana: the main dry season with low precipitation between September and December and two wet seasons (January to February and April to August).

The hydrological network is structured in 8 major drainage basins and small coastal creeks, that flow from South to North. The two largest drainages are the Maroni (65,830 km<sup>2</sup>) that form the Western boundary with Suriname and the Oyapock (26820 km<sup>2</sup>) at the boundary with Brazil. The network represents 112,000 km of watercourse, with 80% being small streams of less than 1 m depth and 10 m wide. The downstream part of the network is under marine influence, with tide influence reaching up to 50 km long watercourse due to the low slope of the downstream part of the watersheds (Tito de Morais & Lauzanne, 1994).

### **I.2.2 Guianese demography and anthropogenic activities**

Population in French Guiana have greatly increased since the late 1980 (Figure I.7), and is actually 4 times higher than in 1970. Until 1990, this increase was essentially caused by a high immigration rate that has reduced through the years. Even if the population growth rate slows down (+3.6% per year between 1999 and 2009, and +2.4% per year between 2009 and 2014; INSEE 2017), demographic models predict that population will exceed 300,000 inhabitants in 2030. The population is mainly concentrated on the coast, with 80% of the population located on a 20 km East-West coastal band.

This coastal band thus concentrates the majority of anthropogenic infrastructures (roads and urban area) and gathers the majority of industrial and agricultural activities that will mainly impact the downstream part of the hydrological network. On the contrary, the interior of the department is composed mainly of forest. Two main activities are conducted in the forested part of the territory: forestry and gold-mining. The forestry activity is managed by the National Forests Office (ONF) that sets rules to reduce environmental impacts of this exploitation on freshwater ecosystems. In addition, gold-mining activities are increasing (Hammond *et al.*, 2007), especially localized non-legal mining that strongly impact freshwater ecosystems.



**Figure I.7:** Evolution of Guianese population from 1954 to 2014 (—) and projection to 2030 from the Omphale model (---). Data from INSEE (03/09/2017)

### I.2.3 History and biogeography

French Guiana is part of the Guiana Shield, an upland region of South America, which formed during Precambrian around 1.7 Ma (Lujan & Armbruster, 2011). The recent ichthyofauna of French Guiana however results from more recent events of sea level oscillations caused by the alternation of hot and cold periods, that occurred during Late Pleistocene (Boujard & Tito de Morais, 1992). During hot and humid periods, sea level raised and the huge quantity of freshwater discharged by the Amazon River created continuous littoral swamps that connected all the river mouth of French Guiana and homogenized the ichthyofauna between drainage basins. These swamps also contributed to the colonization of fish species from the Amazon River to the East and from the Essequibo River and Rio Branco Riverto to the North. During subsequent cold period (-18,000 years), sea level went down and the littoral swamp reduced. The connection between the different drainage basins stopped, except between a few neighboring basins (Maroni and Mana on the West and Approuague and Oyapock on the East). The isolation of some fish led to speciation cases, probably explaining the current differentiation between Western and Eastern basins (Le Bail *et al.*, 2012; de Mérona *et al.*, 2012). In addition, river captures events in the upper portion of rivers had also favored species colonization of the headwaters from the South between Guianese and Amazonian tributaries (Cardoso & Montoya-Burgos, 2009). All these events thus contributed to the large diversity and organization of freshwater fish in French Guiana.

### I.2.4 Aquatic ecology

Guianese freshwater fish fauna counts of 405 species, with 386 strictly freshwater fish species, from 12 orders (Siluriformes and Characiformes are the most represented).

91 of these species (26%) are endemic of drainage basins located in French Guiana. Within French Guiana, wider drainage basins have more fish species (de Mérona *et al.*, 2012), and species identity differs between Eastern and Western drainage due to past history of the region (Le Bail *et al.*, 2012).

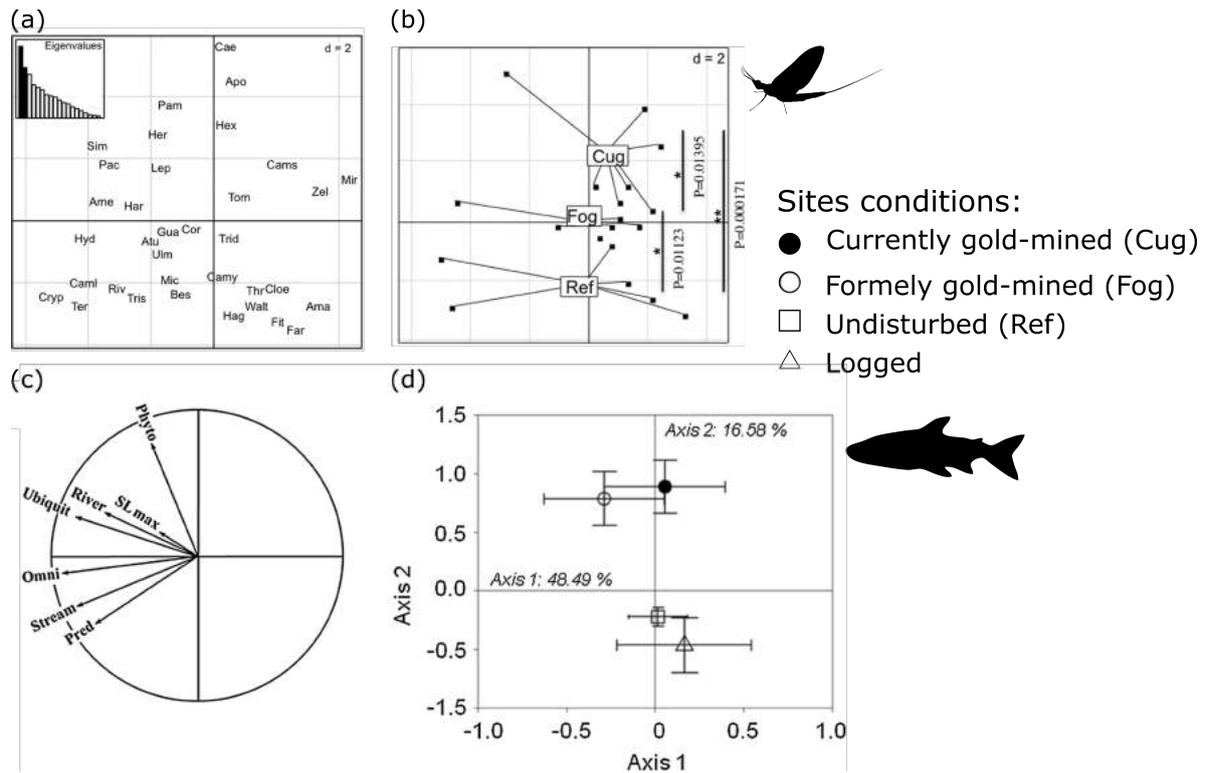
Ecological studies conducted on freshwater fish assemblages in French Guiana have been mainly conducted on the main rivers, especially on the Sinnamary and the Approuague Rivers leading to distinguish fish fauna from estuaries, middle and upper main streams and headwaters (Tito de Morais & Lauzanne, 1994; Boujard & Rojas-Beltran, 1988). At a lower scale, five types of habitats have been described (Boujard *et al.*, 1990): creeks or small streams (width 10 m and depth 1 m), rapids (high flow and important presence of rocks) and pools with different bottom substrates (eroded concave part, sedimentary convex part and intermediary).

### **Freshwater stream assemblages and anthropogenic disturbances**

Studies of stream assemblages started in the 90ies with extensive studies on the streams of the Sinnamary basin before and after the filing of the Petit-Saut's dam, an hydroelectric barrage on the middle course of the Sinnamary River, focused the attention of freshwater ecological studies toward this river and its tributaries. Those studies led to analyses fish densities according to habitat structural complexity, with higher density in habitat with higher complexity, water level and distance of the site to the river (Mérigoux & Ponton, 1999). Spatial variation in assemblages was linked with water quality (oxygen and turbidity) that affect the identity of species according to their life-history traits between streams.

More recently, the development of illegal mining activities led to extensive works on small forest streams with the aim to quantify the strength of the mining disturbance on aquatic assemblages (fish and benthic macroinvertebrates). This work conducted during Allard (2014) and Dedieu (2014) PhD theses, allowed to sample over 150 sites throughout Guiana, in both undisturbed and disturbed (gold mining and logging) environments. Physical and chemical characteristics of undisturbed and logged sites were similar, but logged sites had finer bottom particles (Dedieu *et al.*, 2014). However, gold mined sites greatly differed from undisturbed sites, with higher water turbidity and coarser bottom particles, especially gravel. This modification of the local habitat translated in a modification of assemblage structure (Dedieu *et al.*, 2015; Allard *et al.*, 2016). Both taxonomic and functional structure of assemblage differed between undisturbed and gold mined sites, with a shift from small stream specialist species to larger ubiquitous species inhabiting rivers for fish and from herbivorous and swimmers species to endobenthic burrowers and collector filterers for macroinvertebrates (Figure 1.8).

Although these works contributed to understand how tropical streams and freshwater assemblages are affected by anthropogenic disturbances, the processes that structure local assemblages remained poorly known.



**Figure 1.8:** Modification of Guianese freshwater assemblages by anthropogenic disturbances. Taxonomic structure of Ephemeroptera assemblages (a, distribution of genera; b, distribution of sites clustered by site condition based on taxonomic data) and functional structure of fish assemblages (c, correlation circle of fish functional traits; d, positioning of each site condition centroids based on functional data) differed between undisturbed and logged sites, and gold-mined sites. Abbreviations used in c: Phyto: phytophagous species; SL max: maximum standard length; Ubiquit: ubiquitous species; Omni: omnivorous species; Pred: predatory species. Figures from Dedieu *et al.* (2015) and Allard *et al.* (2016); Ephemeroptera image by George Starr (CC BY-NC-SA 3.0)

## I.3 Thesis

### I.3.1 Aim

Previous work on freshwater fish of French Guiana highlighted the impact of anthropogenic disturbances on these assemblages, both in large rivers and small streams. Understanding the processes that govern these assemblages will help evaluating future impacts of anthropogenic disturbances. The main goal of my thesis was thus to elucidate the assembly rules that govern freshwater fish assemblages of French Guiana. To do, I combined information on 4 types of data: taxonomic, functional and molecular data, and site descriptors.

### I.3.2 Taxonomic data: assemblage composition in streams and rivers

Stream fish assemblage composition was obtained from sampling during the dry seasons, with a single sampling occasion by site (except for a few sites). Fish were collected using rotenone, a non selective piscicide. Rotenone is a chemical compound from the ketone family, naturally produced by tropical plants of the Leguminosae

family found in Australia, Southeast Asia and South America, like jewel vines *Derris* spp. or lacepods *Lonchocarpus* spp. It acts by inhibiting cellular use of oxygen by gill-breathing organisms (Lindahl & Öberg, 1961). This aspect implies that it also affect not only fish, but also freshwater insects, and have thus a strong disturbing effect on local fauna. Nevertheless, it is the only method available to get a good picture of local fish assemblages (Allard *et al.*, 2014). To limit its impact, sampled sites were located closed to a confluence to dilute Rotenone and weaken its effect downstream. Moreover, the lowest possible quantity of rotenone was used, and downstream fish mortality was never observed. All the samples were authorized by the French ministry of environment (DEAL), and the Guianese National Park (PAG) when samples were taken in streams belonging to the PAG.

The sampling protocol takes place as follows: i) a portion of the stream is isolated with fine mesh (4 mm mesh size) stop nets to avoid fish escape out of the station (Figure I.9a); ii) the rotenone is introduced upstream and homogenized before and in the portion; iii) fish are collected; iv) when all fish have been collected, a last passage is made to collect fish lying at the bottom of the stream; v) nets are removed and fish stopped by them collected; vi) fish are identified directly or stored to be identified later.

The portion of the stream isolated was defined to represent one hydromorphological unit (pool, riffle, rapid, fall or run), but was sometimes constrained by the presence of obstacles in the stream, (e.g. trees and branches fallen in the stream making areas where collecting fish is not possible). Several subsequent portions were sampled on the same stream in some sites to represent the hydromorphological diversity of the stream.

Most of the samples were collected during Allard (2014) thesis, as part of a Guiana National Park-DEAL-HYDRECO project (2011-2014), but some sites have been sampled under other projects: CNRS-Nouragues projects (2008; 2010), PAG Itoupe project (2010), DIADEMA project (LabEx CEBA, 2013-2015), and Our Planet Reviewed Mitaraka project (2015).

For rivers, fish are collected using 50 meters long gill-nets with different mesh sizes (from 15 to 35 mm). In each site an overnight sampling with a standard set of 20 gillnets was achieved. Gill-nets are placed on riverbanks (Figure I.9b). The nets are taken out of the water the following morning and identified after all nets removal. Contrary to streams, each site was sampled several times among the years. These samples are conducted to comply with the European Water Framework Directive (DCE). Fish surveys are part of DEAL and OEG projects, and technically operated by Hydreco lab, with the contribution of the EDB lab members (S. Brosse and myself) in Maroni and Approuague sites.

For my thesis, I used sampling data ranging from 2007 to 2016. Although abundance data can help to disentangle some of the assembly processes (e.g. competition between species), the differences in sampling protocol and in sensibility of species to each sampling methods can bias conclusions on assemblages determinants. We thus chose to convert all abundance data into occurrence data.

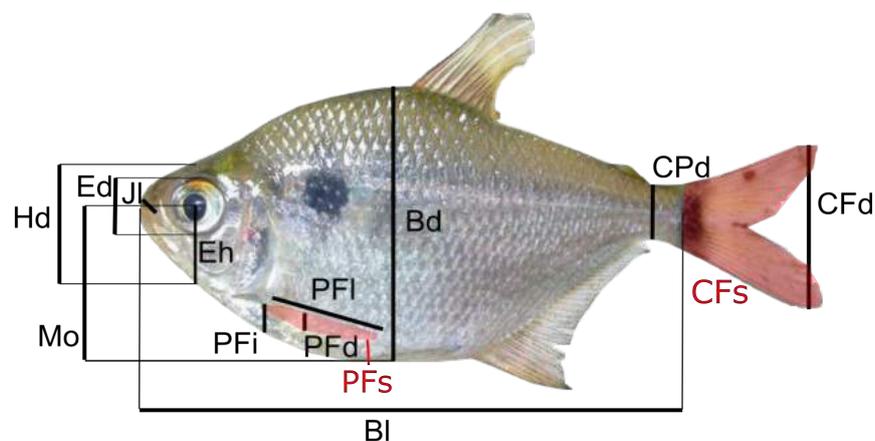


**Figure I.9:** Examples of stream (a) and river (b) sampling sites. (Photo: L. Allard and I. Cantera)

### I.3.3 Functional data: morphological measures and functional traits

To compute functional diversity of assemblages, we used 15 functional traits that account for two main functions: locomotion and nutrition. These functional traits have been widely used to assess functional diversity of fish assemblages (Villéger *et al.*, 2017; Leitão *et al.*, 2017). The lack of ecological knowledge on Guianese fishes prevented from estimating others key functions (life history strategy, feeding composition). For each species, I extracted 14 morphological measures from Toussaint *et al.* (2016) database that contain morphological measures. In addition, we took pictures of fish during 2015 and 2016 sampling sessions to complement our functional database (32 species) and to maximize the number of individuals per species, to reduce potential bias due to intraspecific variability.

The morphological measures are taken from photographs on a lateral view of the fish (Figure I.10 and Table I.1a) and characterize body shape and fin size and surface, eye and mouth size and position. In addition to these measures, the maximum length of each species was recovered from Froese & Pauly (2015).



**Figure I.10:** The 14 morphological measurements taken on each fish species to calculate the functional traits

These measures were then combined into ratios that described body shape, fins eye and mouth relative position on the body or their relative size. These ratios reflect species capacity to feed (prey capture and detection, location in the water column) and to swim (Table I.1b, Villéger *et al.*, 2017; Toussaint *et al.*, 2016).

**Table I.1:** The 14 morphological measurements used (a) and the functional traits calculated (b) on each fish species

(a) Morphological measurements

<b>Symbole</b>	<b>Name</b>	<b>Definition</b>
Bl	Body length	Standard Length
Hd	Head depth	Head depth along the vertical axis of the eye
CPd	Caudal peduncle depth	Minimal caudal peduncle depth
CFd	Caudal fin depth	Maximum depth of caudal fin
Ed	Eye diameter	Vertical diameter of the eye
Jl	Maxillary Jaw Length	Length from snout to the corner of the mouth
Bbl	Barbel maximum Length	Length of the longest barbel
PFI	Pectoral fin length	Length of the longest ray of the pectoral fin
PFi	Pectoral fin position	Vertical distance between the upper insertion of the pectoral fin to the bottom of the body
Bd	Body depth	Maximum body depth
Eh	Eye position	Vertical distance between the centre of the eye to the bottom of the body
Mo	Oral gape position	Vertical distance from the top of the mouth to the bottom of the body
CFs	Caudal fin surface	Total fin surface
PFs	Pectoral fin surface	Total fin surface
MaxLength	Maximum length	Maximum adult length (obtained from FishBase (Froese & Pauly, 2015))

Table I.1 continued

(b) Functional traits

Component	Functional traits	Measure	Relevance
	Maximum length	$\text{Log}(\text{MaxLength})$	Metabolism, trophic impacts, locomotion ability, nutrient cycling
Prey detection	Eye size	$\frac{Ed}{Hd}$	Visual acuity (Boyle & Horn, 2006)
	Barbel length	$\frac{Bl}{Bd}$	Detection of hidden preys
Prey capture	Oral gape position	$\frac{Mo}{Hd}$	Feeding position in the water column (Dumay <i>et al.</i> , 2004; Lefcheck <i>et al.</i> , 2014)
	Maxillary length	$\frac{Jl}{Hd}$	Size and strength of jaw
Position in the water column	Eye position	$\frac{Eh}{Hd}$	Position of fish and/or of its prey in the water column (Winemiller, 1991)
	Body elongation	$\frac{Bl}{Bd}$	Position of fish and/or of its prey in the water column (Winemiller, 1991)
Swimming	Body lateral shape	$\frac{Hd}{Bl}$	Relative depth of the head compared to the body
	Pectoral fin position	$\frac{Pfi}{Bd}$	Pectoral fin use for maneuverability (Dumay <i>et al.</i> , 2004)
	Pectoral fin shape	$\frac{Pfl^2}{Bd}$	Pectoral fin use for propulsion (Fulton <i>et al.</i> , 2001)
	Pectoral fin size	$\frac{Pfl^s}{Bd}$	Pectoral fin use for propulsion (Fulton <i>et al.</i> , 2001)
	Caudal peduncle throttling	$\frac{Bl}{CFd}$	Caudal propulsion efficiency through reduction of drag (Webb, 1984)
	Caudal fin aspect ratio	$\frac{CFd^2}{CFs}$	Caudal fin use for propulsion and/or direction (Gatz, 1968; Webb, 1984)
	Fin surface ratio	$\frac{PFs}{CFs}$	Fin use for swimming
	Relative fin surface	$\frac{(PFs+CFs)}{(Bl \times Bd)}$	Fin total surface compared to body lateral surface

### **I.3.4 Molecular data: molecular markers and phylogenetic relationship**

During field sampling campaigns and fish collection, a small piece of fin was collected on at least 3 individuals per species belonging to different drainage when possible. In total, 6896 individuals were sampled, belonging to 259 species. DNA from these tissue samples were extracted at EDB laboratory using salt-extraction protocol (Aljanabi & Martinez, 1997). Three mitochondrial markers were first amplified: the cytochrome c oxidase I gene (COI), the cytochrome b gene (cytb) and the mitochondrial 12S ribosomal RNA (12S rRNA, or 12S). The cytb marker was removed from the analyses as amplification failed for a substantial part of the samples, and the remaining data did not provided additional information to COI and 12S data. I also included all the sequences available on GenBank, although Guianese freshwater fish are poorly informed in Genebank (142 species have at least one of the two makers). COI and 12S were used to reconstruct phylogenetic relationships between species. The 12S was also used to build the reference database for barcoding studies. After extraction, amplification and sequences cleaning, the molecular data contains 943 sequences. COI and 12S were informed for 422 individuals, COI only was informed for 386 individual, and 135 individuals were informed only for 12S.

### **I.3.5 Sites descriptors: environment and space**

In addition to species related information, environmental characteristics of sites were obtained from field measurements and geographic information system (GIS). These descriptors can be categorized in 3 spatial scales: drainage basin, stream or river reach and local environmental features (Table I.2).

At the drainage basin scale, the identity of the drainage basin in which the site is located was used to distinguish between basins.

At the reach scale, distance from the source, slope and altitude were obtained from GIS and represent the position of the site in the upstream-downstream gradient of the hydrological network. For streams, the pH and the conductivity of the water were also considered as reach descriptors. They were measured on the field with a pH meter (WTW pH 3110 with WTW pH-SenTix 41 electrode) and a conductometer (WTW Cond 3310 with tetraCon 325 captor).

At the local scale, each stream portion was described by its hydromorphological unit (pool, riffle, run, according to the typology of Malavoi & Souchon, 2002), , the forest cover, its bottom grain size (silt, sand, gravel, pebble, boulder and bedrock), the presence of shelters for fish (wood, macrophytes, litter, under-bank and tree roots) and its width and depth. For width, at least 3 measures were taken perpendicular to stream flow, and along each line, the stream depth was measured every metre.

In rivers, local variables were not measured, because fish sampling is achieved using a net sampling with 20 50-metres long gillnets (15 to 35 mm mesh sizes). Sampling therefore occurs over the entire reach, encompassing heterogeneous local

features.

**Table I.2:** Variables used to describe streams and rivers can be classified into 3 classes of spatial scale: drainage basin, reach and local scales.

Watercourse type	Scale		
	Drainage basin	Reach	Local
Stream	Identity	Slope, distance from the source, altitude, ph, conductivity	Width, depth, bottom grain size, shelters, forest cover, hydromorphological unit
River	Identity	Distance from the source, altitude	

### I.3.6 Thesis conduct and structure

The above described data was used to investigate assembly rules of Guianese freshwater fish following these three main points:

1) Description of the diversity of streams fish assemblages: I first described spatial patterns of fish taxonomic diversity in streams, and investigated the environmental determinants of species richness and composition in these streams. I also investigated how space and environment shape the change in species composition between assemblages (manuscript A).

2) Assembly rules of freshwater assemblages: In this chapter, I elucidated the processes that structure freshwater fish assemblages, both in streams and in rivers, using all the information available on assemblages (taxonomic, phylogenetic and functional data). In a first part, I compared temperate and tropical stream assemblages (manuscript B), that are known to differ in their  $\alpha$ -diversity and  $\beta$ -diversity. Using taxonomic and functional diversities, I tested if these differences could be explained by different strength in the processes that structure assemblages. Then I focused on Guianese assemblages and used the multi-faceted approach, with the taxonomic, functional and phylogenetic data to elucidate these processes.

3) A new sampling protocol: Stream assemblage data were obtained using rotenone, a toxicant that kills local fauna, which use is now banned by the law. River assemblage data, collected using nets placed on riverbanks, only represent a small fraction of the fauna and cause substantial fish mortality. Developing a new sampling protocol that can be used in both streams and rivers and that provides an exhaustive image of assemblages is a crucial step to pursue ecological studies in French Guiana. I thus tested the efficiency of eDNA metabarcoding, a new and promising molecular sampling method, to detect fish species, recover assemblage composition and to describe ecological patterns (manuscript C).

For each part, a summary of the corresponding manuscript is given to introduce the chapter.





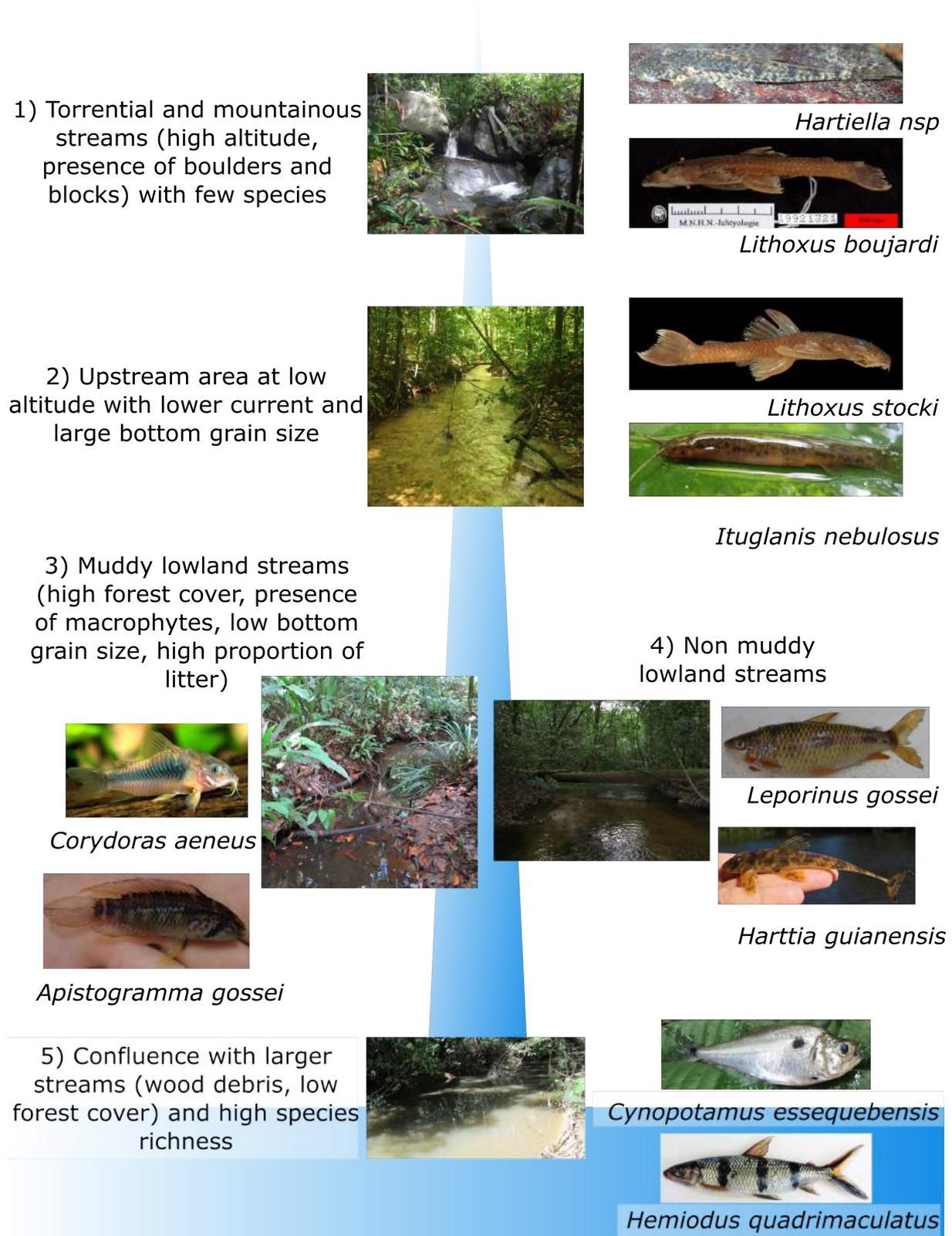
# Chapter II: Diversity of freshwater fish assemblages in undisturbed small streams

## II.1 Summary

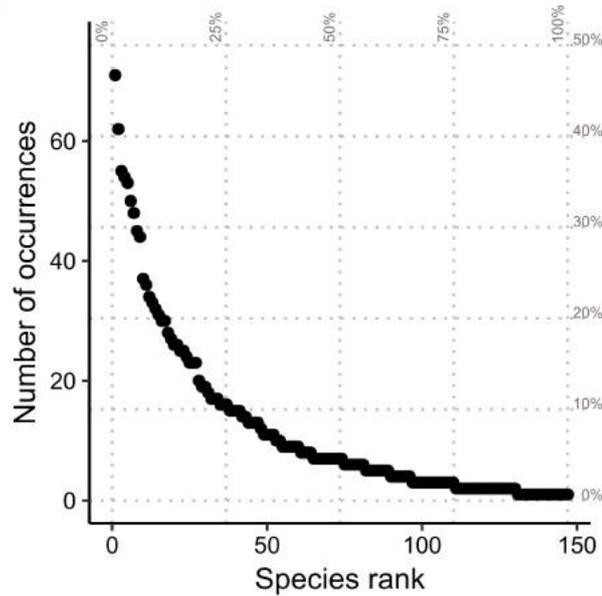
Compared to the temperate freshwater ecosystems that set the basis for freshwater ecological concepts, tropical streams have received less attention, and studies and management of these streams are highly driven by the temperate concepts. This situation is even more pronounced for small tropical streams and the fish assemblages they host. Indeed tropical lowland streams have easier access for sampling and they host more species with economical interest than the small upstream watercourses. In French Guiana, the first ecological studies of freshwater fish assemblages have thus been devoted to large rivers (especially the Sinnamary and the Approuague Rivers). In contrast, fish assemblages of the small streams have been mainly studied through the effect of anthropogenic disturbances on fish assemblage composition (Allard *et al.*, 2016). Without information on the structure of freshwater fish assemblages and its determinants, predictions of fish assemblage and evaluating the impact of anthropogenic disturbances on them remain limited.

We used fish assemblage structure and environmental descriptors for 152 undisturbed sampling sites to describe the patterns of diversity in these streams (species richness and assemblage variation). We then evaluated the effect of environment and space (distance between sites) on these patterns. Each site was described at three spatial scales: the drainage basin scale (corresponding to the identity of basin), the reach scale (position in the upstream-downstream gradient and physic-chemical characteristic) and the site scale (forest cover, shelters presence, bottom grain size, width and depth). For the site scale, we used two different approaches to analyze species richness and assemblage variation: for species richness analyses, we calculated a habitat diversity index using the different categories of shelters and grain size, and classes of width and depth, whereas for assemblage variation, we kept separate these variables. This choice was guided by the niche theory that stipulates that the number of species in a site is related to the habitat diversity, whereas the identity of species is related to the habitat characteristics. In addition to these site-specific descriptors, we also took into account spatial relationships between the sites, based on the distance along the stream between sites located within a same drainage basin.

Species richness in the sites increased from upstream to downstream and in sites with higher habitat diversity. These patterns also hold for in the main fish orders (Characiformes and Siluriformes), testifying that the global pattern did not result from the replacement of different groups of fish. This upstream-downstream gradient was however marked by a succession of five groups of species that characterize five main types of stream habitats (Figure II.1): altitude and torrential streams (1), small (2) and large (3) non-torrential streams, muddy streams (4) and confluence areas with larger



**Figure II.1:** Small streams of French Guiana can be divided into five main types areas along the upstream to downstream gradient. Main characteristics of the area are indicated on the left and some characteristic species of the area are on the right. (Photo: L. Allard, S. Brosse, I. Cantera, K. Cilleros, F. Melki, Guyane Wild Fish, M.N.H.N.)



**Figure II.2:** Rank occurrence plot for the fish assemblages studied in this chapter. Horizontal dashed lines represent the relative number of sites (152 sites in total) and vertical dashed lines represent the relative number of species (147 species in total)

streams (5). This zonation of stream fish species can thus be used as a reference point for future studies on anthropogenic disturbances.

For assemblage composition analyses, space and environment explained similar part of variation (11% and 8% respectively). Contrary to our predictions, the drainage basin did not greatly affect assemblage composition variation. Spatial relationships within watershed were the main responsible for assemblage composition variation, suggesting that dispersion of small stream species is limited by larger streams or rivers. For the environment component, the assemblage composition variation was not explained by few highly influential variables, but all the environmental descriptors slightly affected assemblage variation. This contrasts with some other studies (Mesquita *et al.*, 2006; Nakagawa, 2014) that found few variables explaining assemblage variation. The difference may lie in the fact that species distributions are affected by different components of the environment. This contribution of all environmental components to fish assemblage diversity highlights that anthropogenic disturbances, often modifying several environmental characteristics at the same time, can have great impacts on fish assemblage.

Our study also underlined that a large amount of information remains unknown since the variation explained by our models does not exceed 25%. This can be explained by the high number of species with low occurrence (about 3/4 of the species occurred in less than 10 sites, Figure II.2), that can limit the explanatory power of multivariate analyses. Removing the rarest species (species occurring in less than 5 sites) did not impact greatly the variation explained (from 25% to 30%). Removing a higher proportion of species by increasing the minimum occurrence threshold might increase the explanatory power of species, but in turn, information on those species are lost. Rare species can occupy important function in the ecosystems

(Mouillot *et al.*, 2013) but are however the species that are the most prone to go extinct. The equilibrium between the explanatory power and the loss of information is thus an important thing to consider and depending on the desired goal, one might be favored over the other.

## II.2 Manuscript A

### **Disentangling spatial and environmental determinants of fish species richness and assemblage structure in Neotropical rainforest streams**

*Freshwater Biology*, **62**(10): 1707–1720

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### **INTRODUCTION**

The spatial patterns of biological diversity result from factors acting at different scales. According to the hierarchical filter model proposed by Tonn (1990), large or regional scale processes, related with climatic or biogeographic differences between regions, will define a species pool, *i.e.* a set of candidate species that could occur at smaller scale (Srivastava, 1999). From this regional species pool, species that actually occur in local assemblage are selected through local processes. Those processes encompass biotic interactions between species (predation, competition and facilitation) and the effect of the local environment on local assemblages (Jackson *et al.*, 2001). The determinant role of the local environment on species persistence relies on the Hutchinsons niche concept (Hutchinson, 1957), which still has strong support in the current development of niche modelling approaches in both terrestrial and aquatic ecosystems (e.g. Elith & Leathwick, 2009). For instance, in freshwaters, species distribution within drainage basins can be explained by the variability in the environment along the stream, especially temperature or hydromorphology, creating distinct fish assemblages across this upstream-downstream gradient. These assemblages differ either by the replacement of species due to changing environments and isolation by the distance, or by the gradual addition of species caused by a gradual increase in the size or/and in the diversity of local habitat (Ibarra *et al.*, 2005; Rahel & Hubert, 1991). Such upstream-downstream environment-driven changes in species assemblages are consistent with some of the predictions of the River Continuum Concept (Vannote *et al.*, 1980) that set the basis for further models (e.g. the Riverine Ecosystem Synthesis, Thorp *et al.*, 2006; the Stream Biome Gradient Concept, Dodds *et al.*, 2015) that have informed the currently accepted integrated view of river functioning. In addition to this deterministic effect of the spatial variability on assemblages, stream fish assemblages are also influenced by the hydrological stochasticity (succession of droughts and floods or high- and low-flows between years or seasons), which can override deterministic processes (Grossman *et al.*, 1982, 2010).

Such templates, developed in temperate rivers, have recently been extended to the tropics where the overall patterns of species richness and assemblage structure across the upstream-downstream gradient are broadly comparable to those known in temperate rivers. For instance, Araújo *et al.* (2009) found an increase in species richness and a change in the species composition in fish assemblages along the upstream-downstream gradient in a large Brazilian river. Similarly, Petry & Schulz (2006) reported an upstream- downstream fish zonation in the Sinos River (Southeast Brazil). However, despite an increase in research undertaken on the ecology of tropical rivers, most of information has been obtained from the lowland sections of major river systems, whereas the fish assemblages of headwater streams have been much less studied (Anderson & Maldonado-Ocampo, 2011). This bias is probably due to the easier accessibility to lowland streams for sampling and to the limited commercial interest of the, often small-bodied, fishes inhabiting low-order streams compared to the large and heavily exploited downstream species (Allan *et al.*, 2005). However, large rivers represent only a small part of the river network. For instance, in French Guiana, more than 70% of the permanent river network is represented by streams < 10 m wide (Dedieu *et al.*, 2014). Moreover, such small rivers host about a half of the species inhabiting each river drainage, as shown by Junk *et al.* (2007) for the Amazon, or by Allard *et al.* (2016) for the five main Guianese river basins.

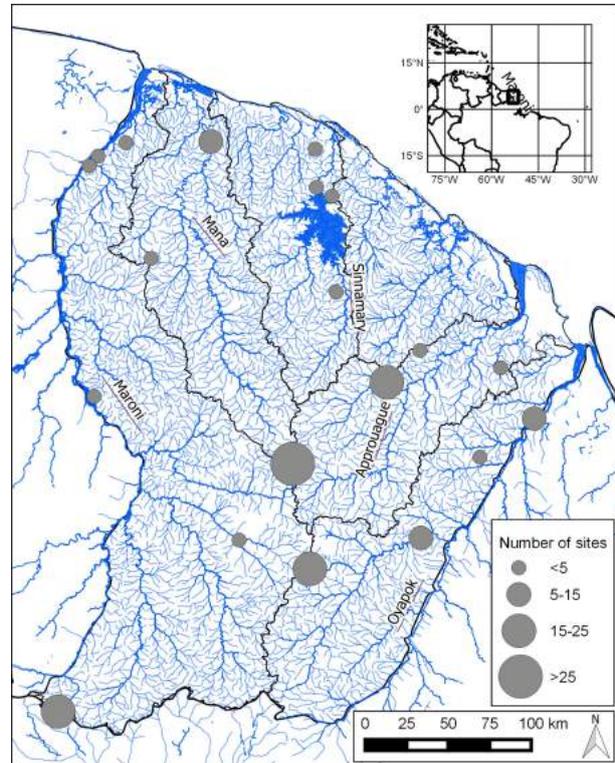
To date, most studies have been devoted to measuring the impact of human disturbance on the fish assemblages in small tropical streams (Allard *et al.*, 2016; Dias *et al.*, 2010; Mol & Ouboter, 2004). The lack of conceptual understanding of the determinants of assemblage structure in these small streams nevertheless limits our ability to predict the fish assemblages they host and hence the impact of ongoing anthropogenic disturbances (e.g. mining, deforestation and urbanisation) on those assemblages. Here, we quantified the determinants of species richness and composition of freshwater fish assemblages in non-impacted headwater streams across the major Guianese drainages. We considered environmental determinants encompassing different hierarchical scales, from drainage basins, to reaches, to microhabitats, while considering river network connectivity that can affect fish distributions (Elith & Leathwick, 2009). We therefore predicted that local species richness (1) increases with stream size (from upstream to downstream) and (2) is higher in sites with high environmental diversity. We also tested if these predictions about determinants of diversity held for the two most speciose fish orders (Characiformes and Siluriformes) or if the global pattern observed resulted from the overlap of different responses across fish clades. Moreover, as the fish community composition differs among river drainages according to the biogeographic history of the region (Le Bail *et al.*, 2012; Lujan & Armbruster, 2011), the pool of species inhabiting the upper reaches should be constrained by the regional context. We hence predict that species assemblage composition (3) differs primarily between drainage basins with different biogeographic histories; (4) is secondly influenced by the sites connectivity within each drainage and; (5) is finally affected by the reach position in the upstream-downstream continuum and the local scale habitat characteristics that

both determine the environmental niche of the species.

## METHODS

### Fish data collection

We achieved complete fish inventories in 152 stream sites in French Guiana (Figure II.3). All the streams considered are small (first to third order) perennial streams flowing in a primary forest environment. Their width varied between 1 and 10 m and depth from 0.1 to 1 m on average. None of the streams were disturbed by human activities as the entire drainage upstream of the sampling sites was free of settlements. The fish surveys were conducted under several different research projects conducted between 2010 and 2015, and each site was sampled once. All sites were sampled during the dry seasons to ensure similar hydrological conditions and optimal detection rate of the species (Allard *et al.*, 2016). We did not consider inter-annual or inter-seasonal variability that would require repeated samples on the same sites at different seasons and during several years. Most of the sites are remote, and multiple sampling was not possible due to the heavy logistic needed to access to the sites. Moreover, sampling during the rainy season was not possible because of river overflows. The sampling protocol was standardised for all sites. Each site was a homogeneous hydrological unit (pool, run, riffle, rapid, waterfall), and its length was on average  $27.49 \pm 17.73$  m (mean  $\pm$  SD). It was proportional to stream width and was  $6.48 \pm 8.84$  times longer than stream width. Since little is known on the home range of Neotropical tropical fishes, the length of our stream sites was the longest possible given that the streams are obstructed by fallen trees and/or dense vegetation where distinguishing fish is difficult. We nevertheless considered the length of sampled stream sites sufficient, as it was similar to that used to analyse fish assemblages in first to fifth order North American streams (Grossman *et al.*, 1998; Hoeinghaus *et al.*, 2007). Fish were collected using rotenone (PREDATOX<sup>®</sup>: a 6.6% emulsifiable solution of rotenone extracted from *Derris elliptica* by Saphyr, Antibes, France) a non selective piscicide, which is traditionally used to catch fishes by Amazonian tribes. Such a method is currently the only way to collect exhaustive information on local fish assemblages, because electrofishing is



**Figure II.3:** Map of French Guiana showing the location of the study sites. Point size is proportional to the number of sites sampled in each area

not efficient in those low conductivity streams (Allard *et al.*, 2014). Nets such as seine and cast nets were not efficient in streams cluttered with fallen trees and branches and visual observation strongly underestimated nocturnal and cryptic species (Allard *et al.*, 2014). We were therefore allowed by the authorities (French ministry of environment and National Amazonian Park) to use rotenone pending the development of an efficient alternative to this destructive method, such as environmental metabarcoding (Valentini *et al.*, 2016). We nevertheless reduced the impact of our rotenone samples by paying a particular attention to releasing as little toxicant as possible to avoid fish mortality downstream from the section studied. This dose was sufficient to detect the entire fish community (Allard, 2014). As there is no published estimation of the efficiency of the method to measure fish abundance according to species behaviour or to environmental characteristics of the sites, we transformed species abundance into species occurrence to control for potential differences in fish capture efficiency according to sites or species as recommended by Oberdorff *et al.* (2001).

In each stream, one to three subsequent sites with homogeneous hydromorphological units were selected. Subsequent hydrological units (*i.e.* sites) were separated using two fine mesh (4 mm) stop nets. A particular attention was devoted to set stop nets simultaneously to avoid fish movement between sites. Rotenone was released a few metres upstream of the stop net located upstream from the upstream site. When two or three subsequent sites were sampled, one or two operators were in charge of collecting fish in each site allowing collecting fish simultaneously from all the subsequent sites with a single rotenone release. At the end of each sampling session we searched for fishes lying on the bottom or hidden in the leaves and debris. In almost all sites, cryptic fish were collected, including highly cryptic Siluriformes such as *Farlowella*, *Harttiella*, *Lithoxus*, or *Ancistrus*, bottom-dwelling fishes such as *Ituglanis* or *Loricaria*, and litter- bank fishes such as small killifishes. Several species of Gymnotiformes inhabiting small Guianese streams, although known to be resistant to rotenone, were also caught.

### **Description of sites**

We characterised each site by three groups of variables defined according to the scale at which they were measured (Table II.1). At the regional scale, we distinguished sites by drainage (from West to East: Maroni, Mana, Sinnamary, Approuague and Oyapock; Figure II.3). At the reach scale, we measured chemical characteristics of the water (pH and conductivity) using pH meter (WTW pH 3110 with WTW pH-SenTix 41 electrode) and conductometer (WTW Cond 3310 with tetraCon 325 captor), and we extracted topographic metrics (distance from the source, slope and altitude) from a GIS (QGIS Development Team, 2016). At the site scale, we measured the percentage of forest canopy cover visually as in Dedieu *et al.* (2014) and classified each site to an hydromorphological unit (pool, run, riffle, rapid and waterfall) according to Delacoste *et al.* (1995). We then measured the local stream habitat variables, including substratum granulometry, shelter availability and channel morphology. The substratum

**Table II.1:** Drainage basin, reach and local scale variables measured at each site. Codes used for each variable are in bold. Distance from the source, slope and altitude were derived from GIS. Conductivity and pH were measured with conductometer and pH meter in the field. Percent of forest coverage, bottom particles (Sand, Gravel, Pebble, Boulder, Bedrock) and of each shelter type (wood, aquatic macrophytes, litter, under-banks, tree roots, open water) were visually estimated. Stream width was measured on transects perpendicular to stream flow and depth was measured every meter on these transects.

<b>Scale</b>	<b>Variables (mean <math>\pm</math> SD [min – max])</b>		
Drainage basin	Identity	Maroni, Mana, Sinnamary, Approuague, Oyapock	
Reach	Distance from the source (km) ( <b>Dis</b> )	3.23 $\pm$ 3.71 [0.4 – 16]	
	Slope (%) ( <b>Slp</b> )	5.24 $\pm$ 3.86 [0.49 – 18.8]	
	Altitude (m) ( <b>Alt</b> )	182.84 $\pm$ 149.45 [28.21 – 632.57]	
	pH	5.88 $\pm$ 0.94 [3.75 – 7.65]	
	Conductivity (S.cm <sup>-1</sup> ) ( <b>Cnd</b> )	30.13 $\pm$ 15.84 [8.4 – 108]	
Local	Forest cover (%) ( <b>Foc</b> )	68.78 $\pm$ 29.86 [0 – 100]	
	Hydromorphological unit	Run, riffle, rapid, fall, pool	
	Bottom particle grain size (%)	Silt (< 0.05mm)	
		Sand (0.05 – 2mm)	
		Gravel ( <b>Grv</b> ) (2 – 10mm)	
		Pebble ( <b>Pbb</b> ) (1 – 3cm)	
		Boulder ( <b>Bld</b> ) (3 – 50cm)	
	Available shelters (%)	Bedrock ( <b>Bdk</b> ) (> 50cm)	
		Wood	
		Macrophyte ( <b>Mac</b> )	
Litter ( <b>Lit</b> )			
Under-banks ( <b>Ubk</b> )			
Depth (m)	Tree roots ( <b>Trt</b> )		
	Open water ( <b>Owa</b> )		
	Mean: 0.24 $\pm$ 0.16 [0.01 – 1.13] ( <b>mDp</b> )		
	CV: 0.43 $\pm$ 0.16 [0 – 0.93] ( <b>cvDp</b> )		
	< 0.2m		
	[0.2, 0.4m[		
	[0.4, 0.6m[		
[0.6, 0.8m[			
[0.8, 1m[			
$\geq$ 1m			
Width (m)	Mean: 3.45 $\pm$ 2.03 [0.95 – 10] ( <b>mWd</b> )		
	CV: 0.19 $\pm$ 0.12 [0 – 0.57] ( <b>cvWd</b> )		
	< 2m		
	[2, 4m[		
	[4, 6m[		
	[6, 8m[		
	[8, 10m[		
$\geq$ 10m			

granulometry was described by estimating visually the percentage of streambed particle grain size cover (silt, sand, pebble, boulder and bedrock were defined according to Cailleux [1954] methodology; see Table II.1 for size classes). Such visual assessment of stream bed particle size clustering has often been used to analyse the relationships between freshwater fish and their physical habitat (e.g. Brosse & Lek, 2000; Grossman *et al.*, 2006). Shelter availability (presence of wood debris, macrophytes, litter, under-banks, tree roots) was measured as a percent coverage of each shelter type as in Allard *et al.* (2016) and the percent coverage without shelter was categorised as open water. Channel morphology was recorded using stream width and depth. We measured stream width on at least three transects perpendicular to the stream flow, and every 5 m if the sampled site was longer than 10 m. Stream depth was recorded every metre across transects. We then calculated the mean and the coefficient of variation (CV) of the depth and width for each site. Those local habitat variables, known to be the main predictors of fish habitat (Allard *et al.*, 2016; Brosse & Lek, 2000; Gorman & Karr, 1978; Grossman *et al.*, 2006) were used to analyse the species composition of the sites. In contrast, the niche theory predicts that the species diversity in a site is more related to the overall environment and its structural diversity than to its different components (Hutchinson, 1957; Kovalenko *et al.*, 2011; Stein *et al.*, 2014). The structural diversity of the site was estimated using the ShannonWiener equitability index ( $-\sum_{i=1}^S p_i \log(p_i) / \log(S)$ ; Shannon, 1948). We calculated this index separately for grain size classes and percentages of shelter types. We did the same for depth and width after sorting them into six classes. A regular increment of 2 m for width and 0.2 m for depth was chosen as it provided a balanced representation of all width and depth classes (see Table II.1 for classes). Those four diversity metrics were summed to obtain a single habitat structural diversity measure for each site, with a maximum value of four reflecting maximum diversity. The habitat structural diversity index and overall descriptors of the environment (distance from the source, slope, altitude, pH, conductivity, forest canopy cover and hydromorphology) were used to explain species richness patterns.

### **Spatial relationships among sites within drainage basins**

We described the spatial structure of the sites within individual drainages with distance-based Morans eigenvector maps (dbMEM, Borcard & Legendre, 2002; Dray *et al.*, 2006). We used the spatial coordinates of the sites to calculate an in-stream distance matrix using the shortest river path between sites. We then derived the dbMEM variables from the distance matrix taking into account the drainage membership of the sites (Declerck *et al.*, 2011). For each drainage, a set of dbMEM variables was computed to describe the spatial structure of the sites within the drainage. Sites from others drainages are given a zero value. We did not use Euclidean distances as they do not represent the dendritic structure of streams and thus provide irrelevant distances to explain assemblage variation for strictly aquatic species which dispersal capacities are strongly constrained by water availability

(Landeiro *et al.*, 2011; Yunoki & Velasco, 2016).

### Statistical analyses

To explain variation in species richness between sites, we used generalised linear-mixed effect models (GLMM) with a Poisson distribution of error terms and with a log link function to explore the relationship between species richness and environmental variables (distance from the source, slope, altitude, pH, conductivity, vegetation cover, hydromorphology and habitat structural diversity). A GLMM was preferred to a classical GLM to account for the spatially nested pattern of our data (site within reach within drainage) and to remove the dependency between sites within a reach (Rhodes *et al.*, 2009). Reach membership was treated as a random effect nested within the drainage membership random effect. Between-drainage and between-reach within drainage variations were modelled by random intercepts only. All variables were centred and scaled to SD. We used a model-selection approach (Burnham & Anderson, 2002) to determine the most important factors in explaining variation in species richness. No prior knowledge about the factors explaining the variation in species richness was used. We thus tested all possible combinations of the eight variables, resulting in 257 candidate models (all combinations + a null model). We used Akaike's information criterion (AICc; Burnham & Anderson, 2002) corrected for small samples to rank the models, with the lowest AICc indicating the best model, and we computed for each model its Akaike weights ( $w_i$ ). As the Akaike weight of the best model was low ( $w_i < 0.5$ ), we retained models with a  $\Delta\text{AICc}$  to the best model  $\leq 4$ . We then used model averaging to estimate parameters, standard errors (SE), variable relative importance and 95% confidence intervals (Burnham & Anderson, 2002). For each model, we computed marginal  $R^2$  ( $R_m^2$ , variance explained by fixed factors) and conditional  $R^2$  ( $R_c^2$ , variance explained by the entire model, *i.e.* by fixed and random factors), following Nakagawa & Schielzeth (2013). To test for congruence across clades, we repeated this analysis on the two most speciose fish orders (Characiformes and Siluriformes, that account for 45% and 30% of the species in our database respectively). Perciformes were not considered here since recent phylogenies have revealed that the previous definition of Perciformes is not valid because it included several phylogenetically distinct valid fish orders (Betancur-R *et al.*, 2013; Sanciangco *et al.*, 2016). Those valid orders did not contain sufficient species in our data to be analysed independently (see Table II.S1 in Supporting Information).

For composition variation, we used a detrended correspondence analysis (DCA) to select the best ordination method (ter Braak & Šmilauer, 2002) and choose between a linear (redundancy analysis) and a unimodal (canonical correspondence analysis, CCA) response. DCA revealed that a unimodal model was the most suitable (gradient length  $> 4$  standard deviation, indicating a complete species turnover along the axis) and we thus used CCA to explore the relationships between assemblage composition variation and spatial and environmental variables (Lepš & Šmilauer, 2003).

We used variation partitioning to separate the effect of overall spatial configuration of sites (drainage-scale and dbMEM spatial variables) and the environment (reach- and site-scale) on the overall species assemblage composition (Borcard *et al.*, 1992). We first selected variables using forward selection as recommended by Blanchet *et al.* (2008): variables were included in the CCA model as long as they did not exceed a  $p > .05$  (p-values were assessed using 999 permutations) and if the  $R_{adj}^2$  of the tested model did not exceed the  $R_{adj}^2$  of the CCA model including all the tested variables. We then calculated the percentage of variation explained by each retained variable while taking into account all the other variables for a given scale (conditional  $R_{adj}^2$ ). We then ran a final CCA with selected variables and calculated  $R_{adj}^2$  for the four components (drainage, dbMEM, reach and site) by permutations of constraining matrix using varcanv software (Peres-Neto *et al.*, 2006). We also tested if the unique parts explained by the spatial and environment components differed. We assessed significance of fractions and difference between components with 999 permutations. We performed the analysis on the entire assemblages (all the 147 species were considered) and on a reduced dataset, where rare species (occurring in less than five sites, *i.e.* occurrence  $< 3\%$  of the sites, see Table II.S1) were removed. In this reduced dataset 58 rare species, which might affect the quality of the CCA, were removed.

Then, to evaluate how species composition differs between sites and reaches, we classified species into groups based on their scores extracted from the two-first axes of the CCA including only reachand local-scale variables, using K-means partitioning. We tested two to fifteen clusters. The final number of groups was selected based on the simple structure index (ssi), with the highest value indicating the best partition (Dolnicar *et al.*, 2000). Although maximal resolution was achieved for a clustering of nine groups, five-group clustering provided similar ecological information (Figures II.S1 and II.S2). We hence considered five groups as the best compromise between species group clustering and interpretability of the results.

We then compared fish species identity between the different drainages to confront the results provided by the CCA and variation partitioning to the known differentiation of fish community composition between river drainages in French Guiana. We computed the turnover component of Jaccards dissimilarity index between the different drainages to remove the effect of richness difference between them (Baselga, 2012). We then used the dissimilarity values in hierarchical clustering analysis with the complete linkage method, based on maximising the correlation between the original distances and the cophenetic distances (Pearsons correlation:  $r = 0.86$ ; Farris, 1969). As for the previous analysis, we repeated these steps on Characiformes and Siluriformes, with single linkage ( $r = 0.79$ ) and average linkage ( $r = 0.81$ ) methods respectively.

All analyses were performed using R software (R Core Team, 2015) with the packages “adespatial” version 0.0-8 (Dray *et al.*, 2017), “lme4” version 1.1-12 (Bates *et al.*, 2015), “MuMIn” version 1.15.6 (Barton, 2016) and “vegan” version 2.4-2 (Oksanen *et al.*, 2016).

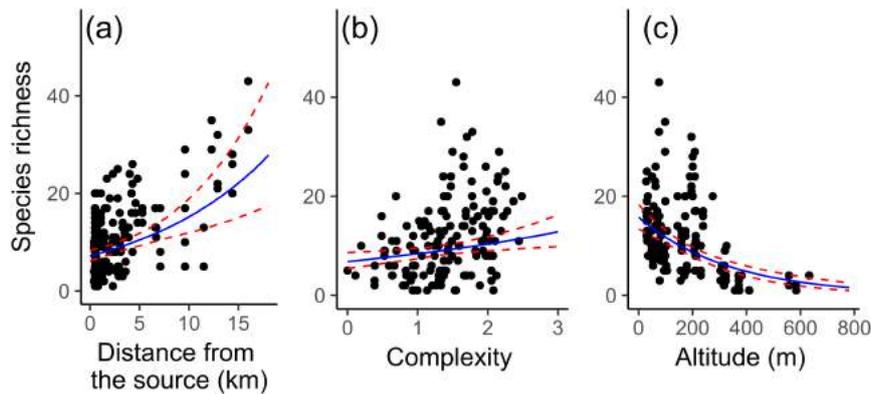
## RESULTS

We collected 147 species belonging to 10 orders of fish (Table II.S1). Characiformes and Siluriformes were the most represented orders, with 66 and 45 species respectively, and together represent more than 75% of the species. At the drainage scale, the Maroni River drainage was the richest in stream species (108 species recorded), followed by the Approuague (79 species), the Sinnamary (65 species), the Oyapock (52 species) and the Mana River drainage had the lowest number of stream species (43 species). Sixty-one species were only found in a single drainage. On average,  $12 (\pm 8)$  species were collected in each site, and species richness ranged from 1 to 43 per site. Among the 257 candidate GLMM predicting species richness, eight were retained as the best models ( $\Delta AICc < 4$ , Table II.2). The distance from the source, the altitude and the habitat structural diversity were retained in the eight models, whereas the hydromorphological unit was never retained.

The best model retained two reach scale variables, distance from the source and altitude, and one local scale variable, the habitat structural diversity. In this model, the variance of the intercept for the drainage random effect was null and the variance for the reach random effect was estimated at 0.15. The 95% confidence intervals of these three variables did not include zero indicating a significant effect of these variables on species richness (Table II.2). In contrast, the effect of forest canopy cover, the conductivity and the pH were not significant as the 95% confidence intervals included zero for those variables. Species richness increased with the distance from the source ( $0.27 \pm 0.057$  (estimate  $\pm$  SE); Figure II.4a) and to a lower extent with habitat structural diversity ( $0.11 \pm 0.042$ ; Figure II.4b), and decreased with the altitude ( $-0.43 \pm 0.066$ ; Figure II.4c). Considering the two main fish orders (Characiformes and Siluriformes) showed that their species richness followed the same increasing pattern according to the distance from the source as the whole fish fauna (Figure II.S3). The species richness in Characiformes also decreased with altitude, while the species richness in Siluriformes increased with habitat structural diversity.

**Table II.2:** The eight best models retained to explain variation in species richness. The models are ranked by their Akaike's information criterion corrected for small samples (AICc). For each model, the variables retained are indicated by a '+'. The Akaike weight ( $w_i$ ) the  $R_m^2$  and  $R_c^2$  are indicated. For each variable, the model-averaged coefficient (estimate  $\pm$  SE), its relative importance (the sum of model  $w_i$  in which the variable is included) and its 95% confidence interval (CI) are given.  $R_m^2$  refers to the marginal  $R^2$  (variance explained by fixed factors) and  $R_c^2$  refers to the conditional  $R^2$  (variance explained by the entire model, *i.e.* by fixed and random factors)

Model	Variable							AICc	$w_i$	$R_m^2$	$R_c^2$
	Distance from the source	Altitude	Slope	Forest cover	Conductivity	pH	Diversity				
1	+	+					+	873.30	.33	.59	.83
2	+	+	+				+	874.90	.15	.59	.83
3	+	+		+			+	875.14	.13	.59	.83
4	+	+			+		+	875.49	.11	.59	.83
5	+	+				+	+	875.49	.11	.59	.83
6	+	+	+	+			+	876.66	.06	.59	.83
7	+	+	+			+	+	877.00	.05	.59	.83
8	+	+	+			+	+	877.06	.05	.59	.83
Estimate $\pm$ SE	0.27 $\pm$ 0.057		-0.43 $\pm$ 0.066	-0.016 $\pm$ 0.042	-0.0062 $\pm$ 0.026	0.0014 $\pm$ 0.023	0.0018 $\pm$ 0.023	0.11 $\pm$ 0.042			
Importance	1	1	.31	.19	.16	.16	1				
95% CI	0.16 – 0.38	-0.56 – -0.31	-0.18 – 0.075	-0.13 – 0.069	-0.10 – 0.12	-0.10 – 0.12	0.029 – 0.20				



**Figure II.4:** Relationships between species richness and the distance of the site from the source (a), the habitat diversity (b) and the altitude (c). Fitted values (solid lines) and 95% confidence interval (dashed lines) are derived from the averaged estimates of the Poisson generalised linear-mixed effect models

After independent forward selections of variables, the five reach scale variables and 14 out of the 18 local scale variables were retained (Table II.3). When we partitioned variation in species composition between the three different scales and the spatial structure derived from dbMEM for the full set of species, drainage membership, reach position and local habitat characteristics explained 5.2%, 7.7% and 10.1% of the variation respectively, and the spatial component explained 10.8% of the variation (Table II.4). When accounting for the other variables, the spatial structure of sites explained the highest amount of species composition variation (7.6%), local habitat characteristics and the drainage membership explained similar variation (4.1% and 4.0% respectively) and the reach-scale variables explained the lowest variation (1.9%). Overall, the spatial structure (drainage and dbMEM variables) explained a similar percentage of variation than the environment component ( $R_{adj}^2$  spatial = 10.9%;  $R_{adj}^2$  environment = 7.9%;  $p = .37$ ). When removing the rare species (species occurring in less than five sites), the percentage of explained variation increased slightly (5% of gain) and the ranking between the different components did not differ, with the spatial component explaining the highest variation in species composition (Tables II.S3 and II.S4).

The forward selection step on all environmental variables (reach and local-scales) excluded silt, sand, under-bank shelters, tree roots and CV of width from CCA analysis. The CCA on all assemblages constrained by all of the retained variables was significant ( $F = 2.22$ ,  $p = .001$ ) and the full set of constraining variables explained 13.9% of the variation in assemblage composition (raw  $R^2 = 27.5\%$ ; Figure 3). We clustered species in five groups with the K-means analysis according to the simple structure index criterion (Table II.S1 and Figure II.S1). The first group of species (group 1 in Figure II.5) was composed of only two species (*Hartiella n. sp.* and *Lithoxus boujardi*) that are characteristic of torrential mountainous upstream sites, with high altitude, marked slopes and waterfalls (Figure 4). Group 2 (19 species) represented species inhabiting upstream sites with less torrential and mountainous characteristics (Figure II.6). Those species are replaced downstream by more ubiquitous ones (group 3, 63 species). Species in group 4 (40 species) preferentially

**Table II.3:** Spatial, reach- and local-scales variables retained after the forward selection step and used in the variation partitioning analysis of overall fish assemblage. The cumulative  $R_{adj}^2$  value given for each variable corresponds to the  $R_{adj}^2$  of the CCA model containing the variable and all the previous ones. The conditional  $R_{adj}^2$  represents the percentage of variation explained by theselected variable while taking into account all the other variables for a given scale. Excluded variables are given in italics

Scale	Variable	Cumulative $R^2$ (%)	Conditional $R_{adj}^2$ (%)
Reach	Altitude	2.8	2.7
	Distance from the source	5.4	2.1
	pH	6.8	0.4
	Conductivity	7.3	0.5
	Slope	7.7	0.3
Local	Mean width	2.2	1.2
	Open water	3.6	0.3
	Boulder	4.5	0.4
	Forest cover	5.2	0.6
	Mean depth	5.8	0.4
	Hydromorphological unit	7.2	1.7
	Macrophytes	7.7	0.6
	Depth CV	8.0	0.3
	Litter	8.4	0.7
	Pebble	8.9	0.5
	Wood	9.2	0.4
	Gravel	9.5	0.3
	Width CV	9.8	0.3
	Bedrock	10.1	0.4
	<i>Under-bank</i>	<i>10.3</i>	-
	<i>Roots</i>	<i>10.3</i>	-
	<i>Sand</i>	<i>10.2</i>	-
<i>Silt</i>	<i>10.2</i>	-	
Spatial	dbMEM 9	2.3	2.4
	dbMEM 1	4.1	1.8
	dbMEM 4	5.5	1.8
	dbMEM 5	6.6	1.2
	dbMEM 6	7.6	1.1
	dbMEM 7	8.4	1.3
	dbMEM 8	9.6	1.2
	dbMEM 3	10.2	0.6
	dbMEM 2	10.8	0.7

inhabit muddy lowland streams covered by dense canopy, with a high percentage of litter and macrophytes. The last group of species contained those that mainly occur in downstream sites with wide and deep morphology and located close to the confluence with a larger river (group 5, 23 species; Figure II.6). The same zonation was found when removing the rare species from the analysis (Figure II.S4).

Species turnover between drainages was moderate, with dissimilarities ranging from 0.13 to 0.59 (mean  $\pm$  SD:  $0.41 \pm 0.14$ ). Hierarchical classification based on species turnover separated western (Maroni and Mana) and eastern (Approuague and Oyapock) river drainages (Figure II.7). The Sinnamary River drainage remained distinct from these two groups. Characiformes species turnover was on average slightly lower than that of the entire fauna ( $0.36 \pm 0.13$ ) but remained of the same

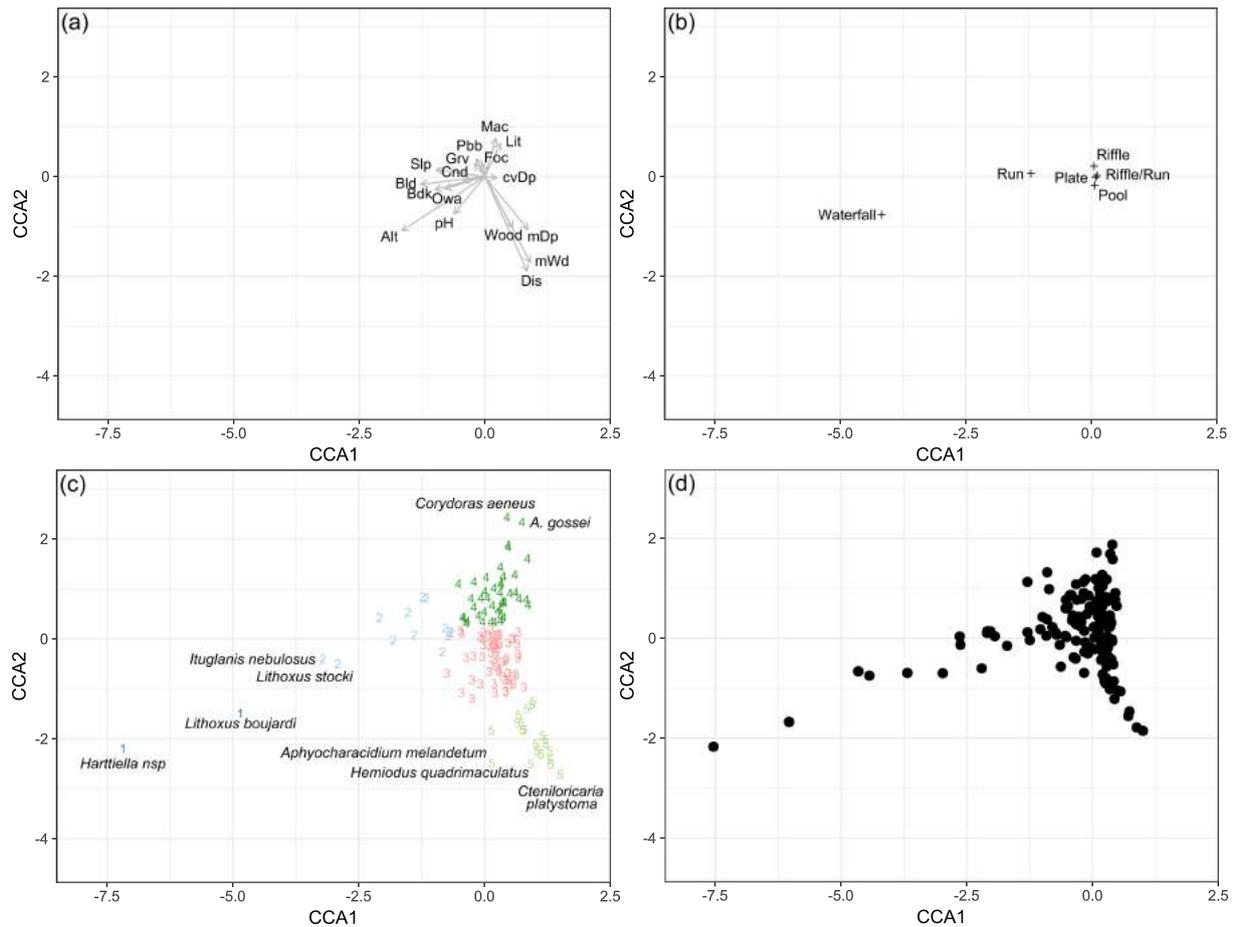
**Table II.4:** Variation partitioning of the fish species occurrence matrix, between the four components (drainage, spatial, reach and local). This partition was achieved on the entire assemblage data after forward selection of the variables.  $R^2$  and  $R_{adj}^2$  values are expressed in percentage

	$R^2$	$R_{adj}^2$	<b>p</b>
Total explained variation	44.0	25.0	
<b>Global effects</b>			
Drainage	8.0	5.2	.001
Spatial	16.6	10.8	.001
Reach	11.1	7.7	.001
Local	21.8	10.1	.001
<b>Pure-effects</b>			
Drainage	5.2	4.0	.001
Spatial	10.8	7.6	.001
Reach	4.1	1.9	.001
Local	13.0	4.1	.001
<b>Shared effects</b>			
Drainage $\cap$ Spatial	< 0	< 0	
Drainage $\cap$ Reach	1.1	1.1	
Drainage $\cap$ Local	3.6	< 0	
Spatial $\cap$ Reach	1.7	1.5	
Spatial $\cap$ Local	2.5	1.3	
Reach $\cap$ Local	2.1	1.9	
Drainage $\cap$ Spatial $\cap$ Reach	< 0	< 0	
Drainage $\cap$ Spatial $\cap$ Local	< 0	3.8	
Drainage $\cap$ Reach $\cap$ Local	< 0	4.0	
Spatial $\cap$ Reach $\cap$ Local	1.8	1.4	
Drainage $\cap$ Spatial $\cap$ Reach $\cap$ Local	2.2	< 0	
Unexplained variation	56.0	75.1	

magnitude (range: 0.15 – 0.59) and no clear distinction between the river drainages was found. In contrast, Siluriformes experienced strong turnover between river drainages ( $0.52 \pm 0.12$ , range: 0.36 – 0.73). The distinction between the groups Mana-Maroni and Approuague-Oyapock was marked and the Siluriformes fauna of the Sinnamary River drainage remained intermediate between the two groups.

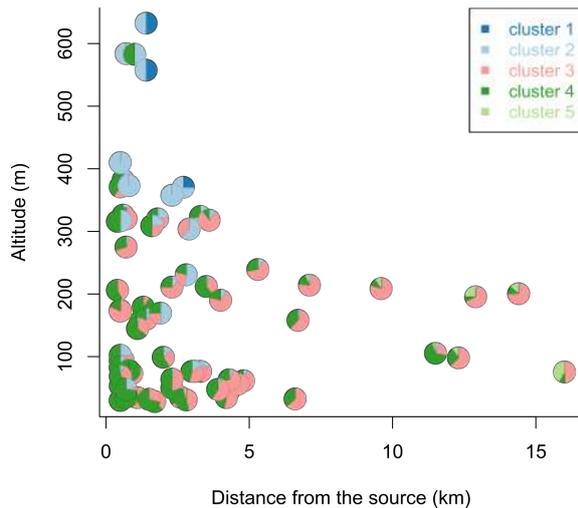
## DISCUSSION

Despite the marked differences in species richness and composition between tropical and temperate fish faunas (Lévêque *et al.*, 2007; Oberdorff *et al.*, 2011), Neotropical rainforest stream fish assemblages were found to be shaped similarly to those of temperate streams. Indeed, local species richness exhibited the expected increase along the upstream-downstream gradient, which was also associated with an increase in local habitat structural diversity. Such a gradient in species richness has also been reported elsewhere in the Neotropical streams and rivers (de Mérona *et al.*, 2012; Mol *et al.*, 2007; Ponton & Copp, 1997; Terra *et al.*, 2016), but in addition to this upstream-downstream pattern, we here show that habitat structural diversity of local

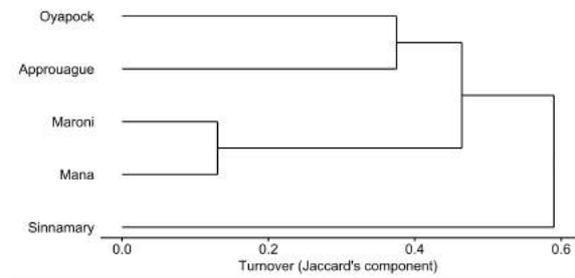


**Figure II.5:** Canonical correspondence analysis (CCA) ordination illustrating fish species assemblage constrained by all environmental variables along the two-first axis (axis 1: 17% of the explained variation,  $p = .001$ ; axis 2: 12% of the explained variation,  $p = .001$ ). Upper left panel (a) quantitative environmental variables are represented by arrows with abbreviations as in Table II.1; upper right panel (b) hydromorphological units; bottom left panel (c) species position in the CCA ordination. Species were grouped according to K-means analysis with  $K = 5$ . *Apistogramma gossei* is abbreviated *A. gossei*. See Table S1 for complete information about species membership for each cluster; bottom right panel (d) position of sites in the CCA ordination

habitat had a positive, although slight, effect on local species richness. Habitat structural diversity includes a wide range of components (from flow velocity to substrate granulometry) and it thus can be described by several measurements (Gorman & Karr, 1978; Mérioux & Ponton, 1999; Willis *et al.*, 2004). Here, we quantified habitat structural diversity as the variability in channel morphology (width and depth), granulometry and shelter availability. Indeed, hydromorphological heterogeneity caused by local variations of the slope or by the presence of woody debris creates areas of reduced current velocity that may be used as refuges for species (Fausch, 1993). Habitat heterogeneity may also act via a trophic pathway where debris and crevices in substrate favour higher diversities of stream invertebrates and periphyton that can be consumed by a variety of fish species with different feeding modes and habits (Downes *et al.*, 1998; Robson & Chester, 1999). Unlike the effect of the distance from the source, of the altitude and of the habitat structure diversity, which significantly influenced the local species richness, the role of river drainage identity was negligible. Hence, historical and macroevolutionary



**Figure II.6:** Stream sites position along the distance from the source and the altitudinal gradients. For each site, the percentage of species belonging to each cluster is represented (see Figure II.5 and Table II.S1 for details on species clustering). Sites belonging to the same stream reach were grouped together to simplify the figure



**Figure II.7:** Hierarchical clustering of the species turnover between the five drainages (Jaccard's turnover component) with complete linkage method

processes that shape the size of the species pool occurring in a river drainage (Jackson *et al.*, 2001), hardly affected the local species richness.

Our results also highlighted that the overall species richness pattern was congruent within the two major fish orders (Characiformes and Siluriformes; Figure II.S3). Species richness in both orders increased from upstream to downstream. The number of Siluriformes species also increased with habitat structural diversity, but we did not detect such a relationship for Characiformes. The increase in diversity in streambed materials (grain size, shelters) primarily favoured the diversity of benthic species. Most of the Siluriformes species are benthic whereas Characiformes species often occupy open waters (Winemiller *et al.*, 2008). This overall difference of fish position in the water column between the two orders might explain the lack of relationship between habitat structural diversity and Characiformes species richness. We also detected a lower richness of Characiformes in the rapids, probably due to the morphology of most Characiformes that is more suited to swimming in open waters, but less to the turbulent water of rapids (Winemiller *et al.*, 2008). Apart from these differences, the overall fish richness pattern also holds for the two main fish orders, meaning that the overall species richness gradient resulted from a gradual increase in diversity rather than a species replacement between different fish orders.

Turning from the determinants of species richness to the determinants of assemblage composition revealed that the environment and spatial effects explained no more than 25% of the composition of species assemblages. Although this value is less than previous studies that reported a much higher explained variation (e.g. Brosse *et al.*, 2013; Terra *et al.*, 2016), it should be noted that the effect we report is corrected for sample size and for the number of variables, which was not the case in

previous studies. Without such correction, our explained variation reached 44% (see Table II.4), a value comparable to of earlier studies. Although correcting for sample size and for the number of variables lowered the amount of variance explained, it permitted an unbiased measurement of the effect and of the importance of the environmental variables on fish assemblage composition (Peres-Neto *et al.*, 2006), and the patterns of assemblage variation we detected were nonetheless significant. The large amount of unexplained variance recorded in the present study, but also in the literature on the determinants of fish assemblage structure in Neotropical streams (e.g. Brosse *et al.*, 2013; Terra *et al.*, 2016), has often been afforded to the low occurrence of most species (c.a. 75% of species occurred in < 10% of the sites), a common problem in tropical community ecology (Hercos *et al.*, 2013; ter Steege *et al.*, 2013). Nevertheless, removing rare species (occurring in less than five sites) only slightly increased the explained percentage of variation in species composition, meaning that unexplained variance was only slightly affected by rare species. An alternative explanation is that temporal variability in the environment promotes some stochasticity in the species composition of assemblages (de Mérona *et al.*, 2012; Grossman *et al.*, 1982). Testing for environmental stochasticity would need repeated samples on the same sites, which is problematic given the destructive nature of rotenone sampling. The development of a non-destructive alternative to rotenone samples is therefore an urgent priority.

Among the influential determinants of assemblage composition, spatial and environmental factors explained similar amounts of variation, which contradicted our expectation that spatial factors should override environmental variables. Although almost half of the species were found in a single drainage, thereby giving rise to a strong spatial effect, the remaining half of the fauna was made up of widespread species, thus constituting a common core to all the river drainages and offsetting the role of the spatial component. This contrasts with the findings of Brosse *et al.* (2013) based on a few sites from the same mountainous area. In that particular case, the regional effect (drainage membership) was of primary importance to explain variation in fish assemblages. Extending our analysis the entire Guianese region therefore provided a more comprehensive view of the determinants of assemblage structure, paralleling therefore the work of Garzon-Lopez *et al.* (2014) on the scale dependence effect on tropical trees assemblages. Although the regional determinants were not the major force driving fish assemblages, they nonetheless had a marked effect on assemblage variation. The species turnover between drainages was responsible for the clustering of the river drainages along an East- West gradient, reflecting the biogeographic history of the region (Boujard & Tito de Morais, 1992; Le Bail *et al.*, 2012). This clustering of river drainages was mirrored in their stream fish assemblages, and was particularly marked in the Siluriformes. Indeed, a substantial part of the siluriform fauna inhabiting streams is made of strictly rheophilic taxa restricted to the upstream parts of rivers (Cardoso & Montoya-Burgos, 2009; Lujan & Armbruster, 2011). Their low dispersal ability probably accentuates the regional effect observed in the Siluriformes. This regional effect was, however, weaker compared to

the spatial structure of the assemblages within each drainage, which explained the highest part of variation in assemblage composition. The low regional effect compared to the strong spatial structure within drainages probably reflects dispersal limitations between different streams belonging to the same drainage (Cilleros *et al.*, 2016; Vitorino Júnior *et al.*, 2016). Indeed, the main channels of the rivers and the presence of rapids can act as dispersal barriers for some small-bodied stream fishes either by way of a distance effect (Datry *et al.*, 2016) or due to predation by or competition with species inhabiting large rivers (Wisz *et al.*, 2013).

The environmental characteristics also affected species composition of fish assemblages. This effect did not result from a few environmental characteristics having a dominant effect, but rather from the combination of several environmental characteristics having slight, but nonetheless significant, effects. We therefore hypothesise that there is no consistent response of all species to environmental variables, but more probably species-specific responses to particular variables, with the result that almost all variables have some significant albeit low explanatory power. This multifactorial contribution of the environment to the fish community composition contrasts with the situation found in temperate streams, where the fish composition is determined by a few dominant environmental features related to stream morphology and streambed substratum size (Mesquita *et al.*, 2006; Nakagawa, 2014; but see Johnson *et al.*, 2007). Although we did not identify a strong environmental gradient shaping fish assemblages, we distinguished five successive groups of species along the upstream-downstream gradient (Figure II.6), revealing an upstream-downstream species succession equivalent to that reported in temperate streams (e.g. Allan & Castillo, 2007). In addition, a particular fauna was found close to the confluence with larger rivers. This last zone might be composed of a specific fish fauna (Albanese *et al.*, 2004), or be a transition between streams and rivers and hence host a mixed fauna (Fernandes *et al.*, 2004). This last situation holds for Guianese streams where we did not detect species strictly inhabiting these zones, but a mix between stream and river fishes, with the occurrence of species usually found in large rivers such as *Hemiodus quadrimaculatus* or *Hypostomus gymnorhynchus* (Le Bail *et al.*, 2012).

The pattern of stream fish zonation we describe could constitute a benchmark for future studies measuring the impact of anthropogenic disturbances on Neotropical forest streams. Moreover, the contribution of almost all the environmental descriptors of the local environment to both species richness and assemblage composition suggests that modifications to only a single component of the environment might alter fish assemblage composition. This is of particular importance since damming, mining and logging are already known to affect the characteristics of Neotropical streams (de Mérona *et al.*, 2005; Dedieu *et al.*, 2014; Dias *et al.*, 2010), and are thus very likely to influence of both species richness and composition of stream fish assemblages.

## **ACKNOWLEDGMENTS**

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**Table II.S1:** List of the species studied, cluster membership (cluster numbers are as in Figure 3) and species occurrence in the 152 sampling sites)

Order	Genus species	Cluster	Occurrence (%)
Beloniformes	<i>Potamorhaphis guianensis</i> (Jardine 1843)	4	0.7
Characiformes	<i>Acestrorhynchus falcatus</i> (Bloch 1794)	3	9.2
Characiformes	<i>Anostomus brevior</i> (Géry 1963)	5	2.0
Characiformes	<i>Aphyocharacidium melandetum</i> (Eigenmann 1912)	5	0.7
Characiformes	<i>Astyanax bimaculatus</i> (Linnaeus 1758)	2	7.2
Characiformes	<i>Astyanax validus</i> (Géry, Planquette & Le Bail 1991)	4	5.9
Characiformes	<i>Bryconamericus aff. hyphesson</i>	4	3.9
Characiformes	<i>Bryconamericus guyanensis</i> (Zarske, Le Bail & Géry 2010)	3	34.9
Characiformes	<i>Bryconops affinis</i> (Günther 1864)	3	32.9
Characiformes	<i>Bryconops caudomaculatus</i> (Günther 1864)	5	2.6
Characiformes	<i>Bryconops melanurus</i> (Bloch 1794)	3	8.6
Characiformes	<i>Characidium zebra</i> (Eigenmann 1912)	3	28.9
Characiformes	<i>Charax gibbosus</i> (Linnaeus 1758)	5	0.7
Characiformes	<i>Copella carsevennensis</i> (Regan 1912)	4	35.5
Characiformes	<i>Creagrutus melanzonus</i> (Eigenmann 1909)	5	0.7
Characiformes	<i>Creagrutus planquettei</i> (Géry & Renno 1989)	2	1.3
Characiformes	<i>Cynopotamus essequibensis</i> (Eigenmann 1912)	5	2.6
Characiformes	<i>Cyphocharax helleri</i> (Steindachner 1876)	3	4.6
Characiformes	<i>Cyphocharax spilurus</i> (Günther 1864)	5	1.3
Characiformes	<i>Erythrinus erythrinus</i> (Bloch & Schneider 1801)	4	17.1
Characiformes	<i>Gasteropelecus sternicla</i> (Linnaeus 1758)	3	7.2
Characiformes	<i>Hemibrycon surinamensis</i> (Géry 1962)	3	17.8
Characiformes	<i>Hemigrammus boesemani</i> (Géry 1959)	4	1.3
Characiformes	<i>Hemigrammus guyanensis</i> (Géry 1959)	5	2.0

Table II.S1 continued

Characiformes	<i>Hemigrammus ocellifer</i> (Steindachner 1882)	3	9.2
Characiformes	<i>Hemigrammus rodwayi</i> (Durbin 1909)	3	3.9
Characiformes	<i>Hemigrammus unilineatus</i> (Gill 1858)	4	16.4
Characiformes	<i>Hemiodus quadrimaculatus</i> (Pellegrin 1909)	5	1.3
Characiformes	<i>Hoplerythrinus unitaeniatus</i> (Spix & Agassiz 1829)	4	2.0
Characiformes	<i>Hoplias aimara</i> (Valenciennes 1847)	3	19.7
Characiformes	<i>Hoplias malabaricus</i> (Bloch 1794)	4	11.2
Characiformes	<i>Hyphessobrycon borealis</i> (Zarske, Le Bail & Géry 2006)	4	12.5
Characiformes	<i>Hyphessobrycon copelandi</i> (Durbin 1908)	3	2.0
Characiformes	<i>Hyphessobrycon roseus</i> (Géry 1960)	3	5.9
Characiformes	<i>Hyphessobrycon simulatus</i> (Géry 1960)	5	2.0
Characiformes	<i>Hypomasticus despaxi</i> (Puyo 1943)	5	4.6
Characiformes	<i>Jupiaba abramoides</i> (Eigenmann 1909)	3	20.4
Characiformes	<i>Jupiaba keithi</i> (Géry, Planquette & Le Bail 1996)	3	8.6
Characiformes	<i>Jupiaba meunieri</i> (Géry, Planquette & Le Bail 1996)	5	0.7
Characiformes	<i>Leporinus friderici</i> (Bloch 1794)	5	1.3
Characiformes	<i>Leporinus gossei</i> (Géry, Planquette & Le Bail 1991)	3	5.3
Characiformes	<i>Leporinus granti</i> (Eigenmann 1912)	3	8.6
Characiformes	<i>Leporinus lebaili</i> (Géry & Planquette 1983)	4	1.3
Characiformes	<i>Leporinus nijsseni</i> (Garavello 1990)	3	0.7
Characiformes	<i>Leporinus pellegrini</i> (Steindachner 1910)	5	0.7
Characiformes	<i>Melanocharacidium cf. blennioides</i>	5	2.0
Characiformes	<i>Melanocharacidium dispilomma</i> (Buckup 1993)	5	3.9
Characiformes	<i>Microcharacidium eleotrioides</i> (Géry 1960)	4	10.5
Characiformes	<i>Moenkhausia aff. grandisquamis</i>	3	3.3
Characiformes	<i>Moenkhausia aff. intermedia</i>	3	2.6

Table II.S1 continued

Characiformes	<i>Moenkhausia chrysargyrea</i> (Günther 1864)	3	15.1
Characiformes	<i>Moenkhausia collettii</i> (Steindachner 1882)	3	16.4
Characiformes	<i>Moenkhausia georgiae</i> (Géry 1965)	5	5.9
Characiformes	<i>Moenkhausia hemigrammoides</i> (Géry 1965)	4	7.2
Characiformes	<i>Moenkhausia moisae</i> (Géry, Planquette & Le Bail 1995)	3	10.5
Characiformes	<i>Moenkhausia oligolepis</i> (Günther 1864)	3	31.6
Characiformes	<i>Moenkhausia surinamensis</i> (Géry 1965)	3	15.1
Characiformes	<i>Myloplus ternetzi</i> (Norman 1929)	5	3.3
Characiformes	<i>Nannostomus bifasciatus</i> (Hoedeman 1954)	3	11.2
Characiformes	<i>Parodon guyanensis</i> (Géry 1959)	3	2.0
Characiformes	<i>Phenacogaster wayana</i> (Le Bail and Lucena 2010)	3	10.5
Characiformes	<i>Poptella brevispina</i> (Reis 1989)	3	15.8
Characiformes	<i>Pristella maxillaris</i> (Ulrey 1894)	5	3.3
Characiformes	<i>Pyrrhulina filamentosa</i> (Valenciennes 1847)	4	40.8
Characiformes	<i>Roeboexodon geryi</i> (Myers 1960)	5	2.0
Characiformes	<i>Tetragonopterus rarus</i> (Zarske, Géry & Isbrücker 2004)	5	1.3
Characiformes	<i>Thayeria ifati</i> (Géry 1959)	3	4.6
Cichliformes	<i>Aequidens tetramerus</i> (Heckel 1840)	4	4.6
Cichliformes	<i>Apistogramma gossei</i> (Kullander 1982)	4	1.3
Cichliformes	<i>Cleithracara maronii</i> (Steindachner 1881)	4	3.3
Cichliformes	<i>Crenicichla albopunctata</i> (Pellegrin 1904)	4	19.7
Cichliformes	<i>Crenicichla johanna</i> (Heckel 1840)	3	2.6
Cichliformes	<i>Crenicichla saxatilis</i> (Linnaeus 1758)	4	6.6
Cichliformes	<i>Geophagus camopiensis</i> (Pellegrin 1903)	3	0.7
Cichliformes	<i>Guianacara geayi</i> (Pellegrin 1902)	3	7.2
Cichliformes	<i>Guianacara owroewefi</i> (Kullander & Nijssen 1989)	3	2.0

Table II.S1 continued

Cichliformes	<i>Krobia aff. guianensis sp1</i>	3	3.9
Cichliformes	<i>Krobia aff. guianensis sp2</i>	4	21.1
Cichliformes	<i>Krobia itanyi</i> (Puyo 1943)	4	11.2
Cichliformes	<i>Nannacara aureocephalus</i> (Allgayer 1983)	4	15.1
Cichliformes	<i>Satanoperca rhynchitis</i> (Kullander 2012)	3	2.6
Cyprinodontiformes	<i>Anablepsoides gaucheri</i> (Keith, Nandrin & Le Bail 2006)	2	1.3
Cyprinodontiformes	<i>Anablepsoides holmiae</i> (Eigenmann 1909)	3	2.0
Cyprinodontiformes	<i>Anablepsoides igneus</i> (Huber 1991)	2	21.7
Cyprinodontiformes	<i>Anablepsoides lungi</i> (Berkenkamp 1984)	4	7.9
Cyprinodontiformes	<i>Laimosemion agilae</i> (Hoedeman 1954)	4	11.8
Cyprinodontiformes	<i>Laimosemion cf. geayi</i>	2	0.7
Cyprinodontiformes	<i>Laimosemion geayi</i> (Vaillant 1899)	4	22.4
Cyprinodontiformes	<i>Laimosemion xiphidius</i> (Huber 1979)	4	9.9
Gobiiformes	<i>Eleotris pisonis</i> (Gmelin 1789)	4	1.3
Gymnotiformes	<i>Brachyhypopomus beebei</i> (Schultz 1944)	4	3.3
Gymnotiformes	<i>Eigenmannia virescens</i> (Valenciennes 1836)	3	6.6
Gymnotiformes	<i>Electrophorus electricus</i> (Linnaeus 1766)	3	2.0
Gymnotiformes	<i>Gymnotus carapo</i> (Linnaeus 1758)	4	46.7
Gymnotiformes	<i>Gymnotus coropinae</i> (Hoedeman 1962)	4	29.6
Gymnotiformes	<i>Hypopomus artedi</i> (Kaup 1856)	4	9.9
Gymnotiformes	<i>Hypopygus lepturus</i> (Hoedeman 1962)	4	2.6
Gymnotiformes	<i>Japigny kirschbaum</i> (Meunier, Jégu & Keith 2011)	3	3.3
Gymnotiformes	<i>Sternopygus macrurus</i> (Bloch & Schneider 1801)	3	24.3
<i>Incertae sedis in Ovalentaria</i>	<i>Polycentrus schomburgkii</i> (Müller & Troschel 1849)	4	0.7
Myliobatiformes	<i>Potamotrygon orbignyi</i> (Müller & Henlé 1841)	3	0.7
Siluriformes	<i>Ancistrus aff. hoplogenyis</i> (Günther 1864)	5	1.3

Table II.S1 continued

Siluriformes	<i>Ancistrus aff. temminckii</i> (Valenciennes 1840)	4	1.3
Siluriformes	<i>Ancistrus cf. leucostictus</i> (Günther 1864)	2	17.1
Siluriformes	<i>Batrochoglanis raninus</i> (Valenciennes 1840)	3	18.4
Siluriformes	<i>Callichthys callichthys</i> (Linnaeus 1758)	4	5.9
Siluriformes	<i>Cetopsidium orientale</i> (Vari, Ferraris & Keith 2003)	3	4.6
Siluriformes	<i>Chasmocranus brevior</i> (Eigenmann 1912)	2	4.6
Siluriformes	<i>Chasmocranus longior</i> (Eigenmann 1912)	3	5.3
Siluriformes	<i>Corydoras aeneus</i> (Gill 1858)	4	2.0
Siluriformes	<i>Corydoras amapaensis</i> (Nijssen 1972)	4	1.3
Siluriformes	<i>Corydoras geoffroy</i> (Lacepède 1803)	3	12.5
Siluriformes	<i>Corydoras guianensis</i> (Nijssen 1970)	5	2.0
Siluriformes	<i>Corydoras spilurus</i> (Norman 1926)	5	1.3
Siluriformes	<i>Cteniloricaria platystoma</i> (Günther 1868)	5	0.7
Siluriformes	<i>Farlowella reticulata</i> (Boeseman 1971)	3	5.9
Siluriformes	<i>Farlowella rugosa</i> (Boeseman 1971)	3	1.3
Siluriformes	<i>Glanidium leopardum</i> (Hoedeman 1961)	3	3.3
Siluriformes	<i>Harttia fowleri</i> (Pellegrin 1908)	2	0.7
Siluriformes	<i>Harttia guianensis</i> (Rapp Py-Daniel & Oliveira 2001)	3	2.6
Siluriformes	<i>Harttiella longicauda</i> (Covain & Fisch-Muller 2012)	3	1.3
Siluriformes	<i>Harttiella lucifer</i> (Covain & Fisch-Muller 2012)	2	3.9
Siluriformes	<i>Harttiella nsp</i>	1	3.3
Siluriformes	<i>Helogenes marmoratus</i> (Günther 1863)	4	36.2
Siluriformes	<i>Heptapterus bleekeri</i> (Boeseman 1953)	2	3.9
Siluriformes	<i>Hypostomus gymnorhynchus</i> (Norman 1926)	5	1.3
Siluriformes	<i>Ituglanis amazonicus</i> (Steindachner 1882)	4	4.6

Table II.S1 continued

Siluriformes	<i>Ituglanis nebulosus</i> (de Pinna & Keith 2003)	2	23.7
Siluriformes	<i>Lithoxus boujardi</i> (Muller & Isbrücker 1993)	1	4.6
Siluriformes	<i>Lithoxus planquettei</i> (Boeseman 1982)	3	9.9
Siluriformes	<i>Lithoxus stocki</i> (Nijssen & Isbrücker 1990)	2	4.6
Siluriformes	<i>Megalechis thoracata</i> (Valenciennes 1840)	4	0.7
Siluriformes	<i>Ochmacanthus cf. alternus</i> (Myers 1927)	5	0.7
Siluriformes	<i>Ochmacanthus reinhardti</i> (Steindachner 1882)	4	0.7
Siluriformes	<i>Otocinclus mariae</i> (Fowler 1940)	5	1.3
Siluriformes	<i>Phenacorhamdia tenuis</i> (Mees 1986)	4	5.3
Siluriformes	<i>Pimelodella cristata</i> (Müller & Troschel 1849)	3	13.2
Siluriformes	<i>Pimelodella geryi</i> (Hoedeman 1961)	3	5.3
Siluriformes	<i>Pimelodella procera</i> (Mees 1983)	3	9.9
Siluriformes	<i>Pseudancistrus brevispinis</i> (Heitmans, Nijssen & Isbrücker 1983)	3	4.6
Siluriformes	<i>Rhamdia quelen</i> (Quoy & Gaimard 1824)	4	8.6
Siluriformes	<i>Rineloricaria stewarti</i> (Eigenmann 1910)	3	5.9
Siluriformes	<i>Tatia brunnea</i> (Mees 1974)	3	1.3
Siluriformes	<i>Tatia intermedia</i> (Steindachner 1877)	3	3.9
Siluriformes	<i>Trachelyopterus galeatus</i> (Linnaeus 1766)	4	1.3
Siluriformes	<i>Zungaro zungaro</i> (Humboldt 1821)	2	0.7
Synbranchiformes	<i>Synbranchus marmoratus</i> (Bloch 1795)	3	2.0

**Table II.S2:** Models retained to explain variation in the number of Characiformes (a) and Siluriformes (b). The models are ranked by their Akaike's Information Criterion corrected for small samples (AICc). All the models with an AICc within 4 units from the best model are indicated. For each model, the Akaike weight ( $w_i$ ) is indicated, the variables included are noted with + and the  $R_m^2$  and  $R_c^2$  are indicated. For each variable, the model-averaged coefficient (estimate  $\pm$  SE), its relative importance (the sum of model  $w_i$  in which the variable is included) and its 95% confidence interval (CI) are indicated. For hydromorphological unit, detailed results for each type are given below the table. Abbreviations used: Dis, distance from the source; Alt, altitude; Cpx, complexity; Slp, slope; Foc, forest cover; Cnd, conductivity; Hdu, hydromorphological unit.

## (a) Characiformes

Model	Variable								AICc	$w_i$	$R_m^2$	$R_c^2$
	Dis	Alt	Slp	Foc	Cnd	pH	Cpx	Hdu				
1	+	+						+	699.47	.23	.97	.98
2	+	+						+	699.66	.21	.97	.98
3	+	+				+		+	701.57	.08	.97	.98
4	+	+		+				+	701.71	.07	.97	.98
5	+	+				+	+	+	701.75	.07	.97	.98
6	+	+			+			+	701.78	.07	.97	.98
7	+	+	+					+	701.77	.07	.97	.98
8	+	+		+				+	702.01	.06	.97	.98
9	+	+			+			+	702.02	.06	.97	.98
10	+	+	+					+	702.02	.06	.97	.98
Estimate	0.43 $\pm$	-0.66 $\pm$	-4.17x10-4 $\pm$	0.0021 $\pm$	-0.0012 $\pm$	-0.0063 $\pm$	0.043 $\pm$	†				
$\pm$ SE	0.074	0.11	0.033	0.027	0.035	0.036	0.062					
Importance	1	1	.14	.14	.14	.15	.47	1				
95% CI	0.28 –	-0.88 –	-0.18 – 0.18	-0.12 –	-0.20 –	-0.20 –	-0.058 –	†				
	0.57	0.44		0.15	0.18	0.12	0.40					

† Estimate  $\pm$  SE (95% CI) for each unit. Pool (intercept): 1.40  $\pm$  0.17 (1.07 – 1.73); Plate: -0.21  $\pm$  0.17 (-0.56 – 0.13); Run: -0.56  $\pm$  0.32 (-1.19 – 0.08); Riffle: 0.21  $\pm$  0.31 (-0.41 – 0.83); Riffle/run: -0.0072  $\pm$  0.16 (-0.32 – 0.30); Waterfall (No species of Characiformes was found in this hydromorphological unit): -19.37  $\pm$  3295 (-6533 – 6494).

Table II.S2 continued

(b) Siluriformes

Model	Variable									AICc	w <sub>i</sub>	R <sub>m</sub> <sup>2</sup>	R <sub>c</sub> <sup>2</sup>
	Dis	Alt	Slp	Foc	Cnd	pH	Cpx	Hdu					
1	+							+		535.58	.19	.22	.28
2	+			+				+		536.68	.11	.24	.27
3	+							+		537.12	.09	.23	.28
4	+	+						+		537.25	.08	.23	.27
5	+			+				+		537.62	.07	.22	.28
6	+					+		+		537.72	.07	.23	.28
7	+			+				+		538.62	.04	.24	.27
8	+	+		+				+		538.67	.04	.24	.27
9	+			+	+			+		538.76	.04	.24	.27
10	+				+			+		538.86	.04	.24	.28
11	+			+		+		+		538.87	.04	.24	.27
12	+							+	+	538.98	.04	.30	.32
13	+	+						+		539.00	.03	.24	.28
14	+					+		+		539.06	.03	.23	.27
15	+									539.29	.03	.18	.27
16	+	+				+		+		539.39	.03	.23	.28
17	+	+			+			+		539.44	.03	.23	.27
Estimate	0.27 ±	0.0091 ±	0.017 ±	0.0041 ±	-2x10 <sup>-4</sup> ±	0.012 ±	0.15 ±	‡					
± SE	0.051	0.036	0.044	0.027	0.029	0.040	0.06						
Importance	1	.22	.27	.17	.17	.24	.97	.04					
95% CI	0.17 –	-0.091 –	-0.063 –	-0.010 –	-0.14 – 0.14	-0.084 –	0.032 –	‡					
	0.37	0.18	0.19	0.15		0.19	0.28						

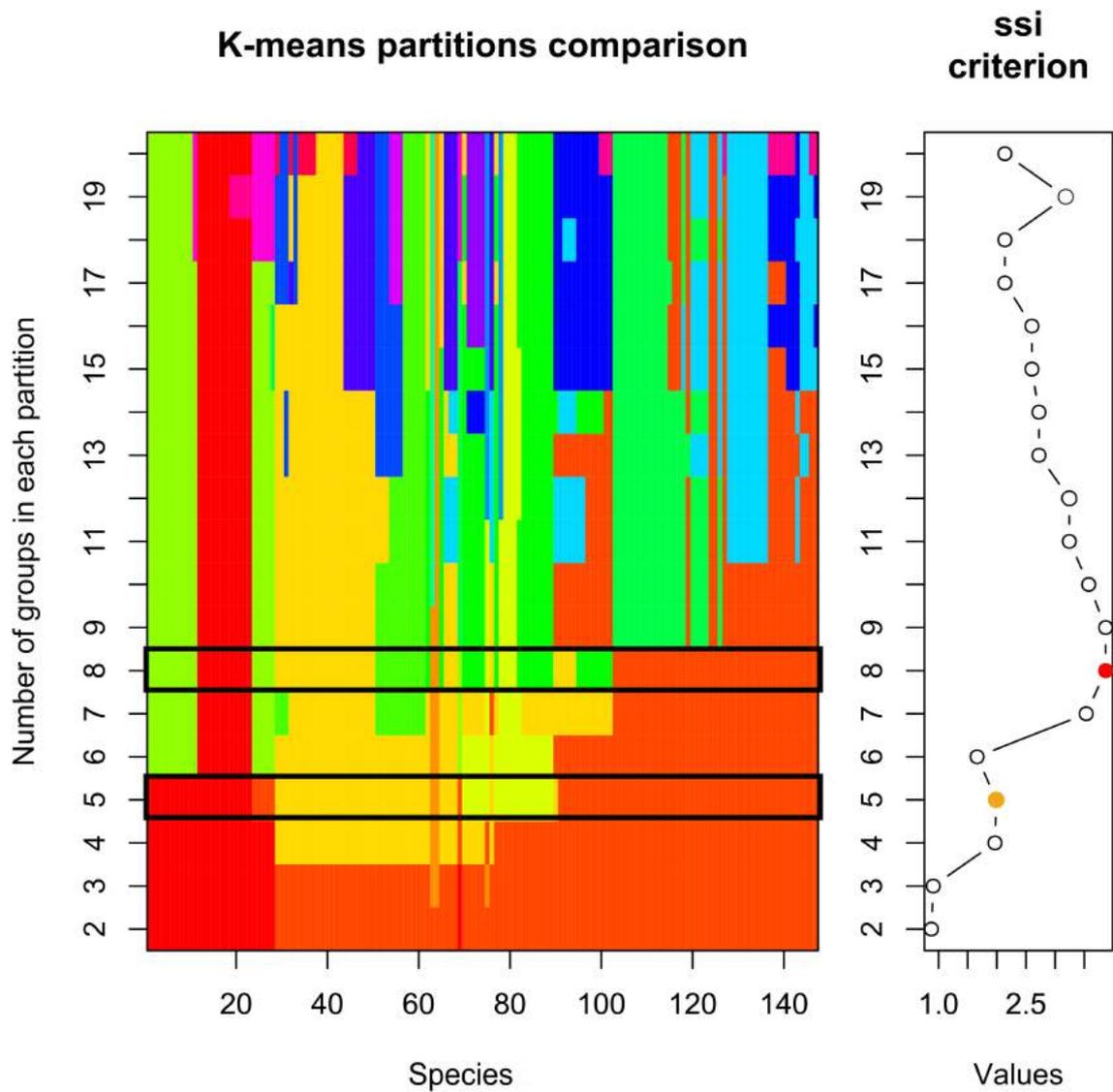
‡ Estimate ± SE (95% CI) for each unit. Pool (intercept): 1.40 ± 0.17 (1.07 – 1.73); Plate: -0.21 ± 0.17 (-0.56 – 0.13); Run: -0.56 ± 0.32 (-1.19 – 0.08); Riffle: 0.21 ± 0.31 (-0.41 – 0.83); Riffle/run: -0.0072 ± 0.16 (-0.32 – 0.30); Waterfall (No species of Characiformes was found in this hydromorphological unit): -19.37 ± 3295 (-6533 – 6494).

**Table II.S3:** Spatial, reach- and local- scales variables retained after the forward selection step and used in the variation partitioning analysis of fish assemblage while removing the rare species (found in less than 5 sites). The cumulative  $R^2_{adj}$  value given for each variable corresponds to the  $R^2_{adj}$  of the CCA model containing the variable and all the previous ones. The conditional  $R^2_{adj}$  represents the percentage of variation explained by the selected variable while taking into account all the other variables for a given scale. Excluded variables are given in italics

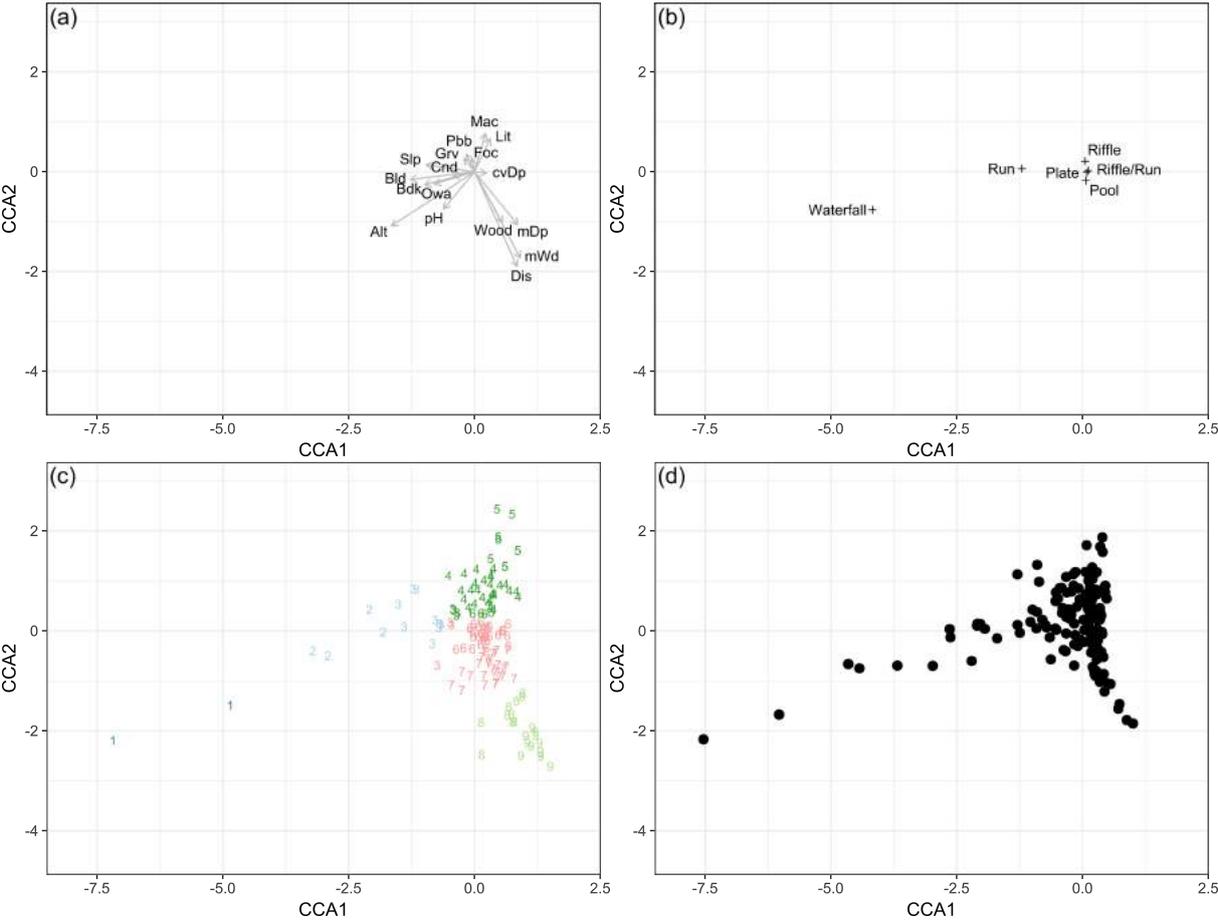
Scale	Variable	Cumulative $R^2$ (%)	Conditional $R^2_{adj}$ (%)
Reach	Altitude	4.2	3.9
	Distance from the source	7.1	2.5
	pH	9.0	0.8
	Conductivity	9.9	0.9
	Slope	10.4	0.5
Local	Mean width	2.6	1.2
	Boulder	4.6	0.6
	Open water	6.0	0.5
	Forest cover	7.0	0.8
	Macrophytes	7.9	0.8
	Depth CV	8.6	0.4
	Litter	9.1	0.7
	Pebble	9.8	0.6
	Wood	10.3	0.5
	Gravel	10.7	0.4
	Mean depth	11.2	0.4
	Hydromorphological unit	13.1	0.3
	Width CV	13.5	0.4
	Under-bank	13.8	0.4
	<i>Bedrock</i>	<i>14.0</i>	-
	<i>Roots</i>	<i>14.1</i>	-
<i>Sand</i>	<i>14.1</i>	-	
<i>Silt</i>	<i>14.2</i>	-	
Spatial	dbMEM 4	1.9	2.4
	dbMEM 1	3.8	2.0
	dbMEM 8	5.0	1.9
	dbMEM 7	6.8	1.8
	dbMEM 5	7.9	1.2
	dbMEM 3	8.9	1.0
	dbMEM 9	9.7	0.8
	dbMEM 6	10.3	0.6
	<i>dbMEM 2</i>	<i>10.7</i>	-

**Table II.S4:** Variation partitioning of the fish species occurrence matrix after removing species occurring in less than 5 sites, between the four components (drainage, spatial, reach and local). This partition was achieved on the entire assemblage data after forward selection of the variables.  $R^2$  and  $R^2_{adj}$  values are expressed in percentage

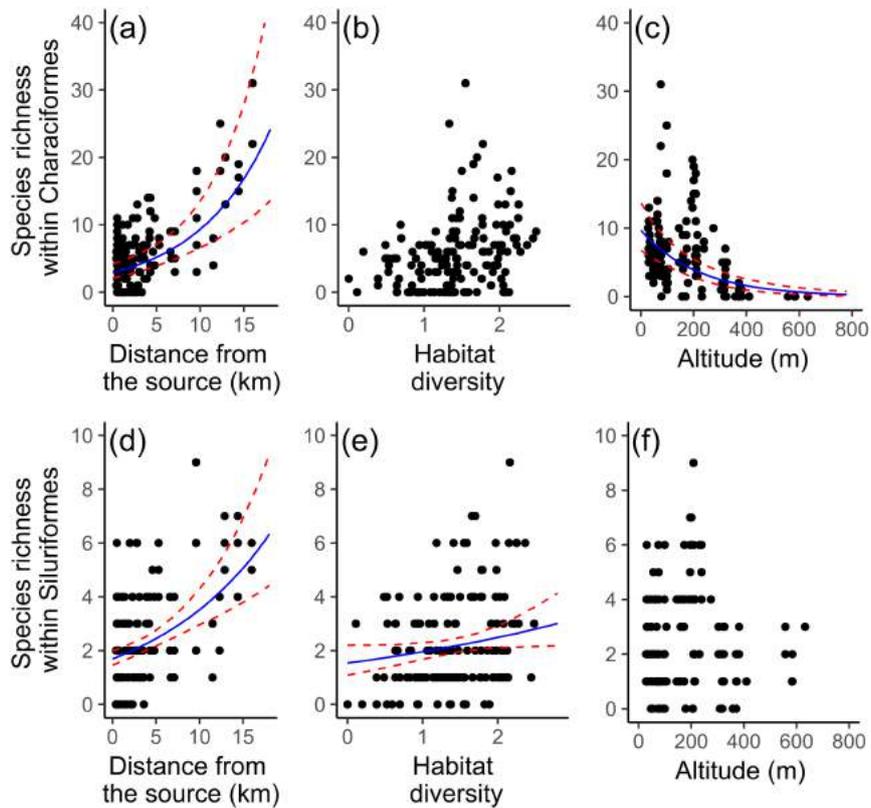
	$R^2$	$R^2_{adj}$	<b>p</b>
Total explained variation	44.8	29.8	
<b>Global effects</b>			
Drainage	8.7	6.2	.001
Spatial	14.9	10.3	.001
Reach	13.3	10.4	.001
Local	24.0	13.9	.001
<b>Pure-effects</b>			
Drainage	6.2	5.4	.001
Spatial	9.8	7.4	.001
Reach	4.6	2.9	.001
Local	13.0	5.4	.001
<b>Shared effects</b>			
Drainage $\cap$ Spatial	< 0	< 0	
Drainage $\cap$ Reach	1.1	1.0	
Drainage $\cap$ Local	5.2	< 0	
Spatial $\cap$ Reach	1.7	1.5	
Spatial $\cap$ Local	2.4	1.4	
Reach $\cap$ Local	3.0	2.7	
Drainage $\cap$ Spatial $\cap$ Reach	< 0	< 0	
Drainage $\cap$ Spatial $\cap$ Local	< 0	2.4	
Drainage $\cap$ Reach $\cap$ Local	< 0	2.9	
Spatial $\cap$ Reach $\cap$ Local	2.6	2.3	
Drainage $\cap$ Spatial $\cap$ Reach $\cap$ Local	2.9	< 0	
Unexplained variation	54.2	70.2	



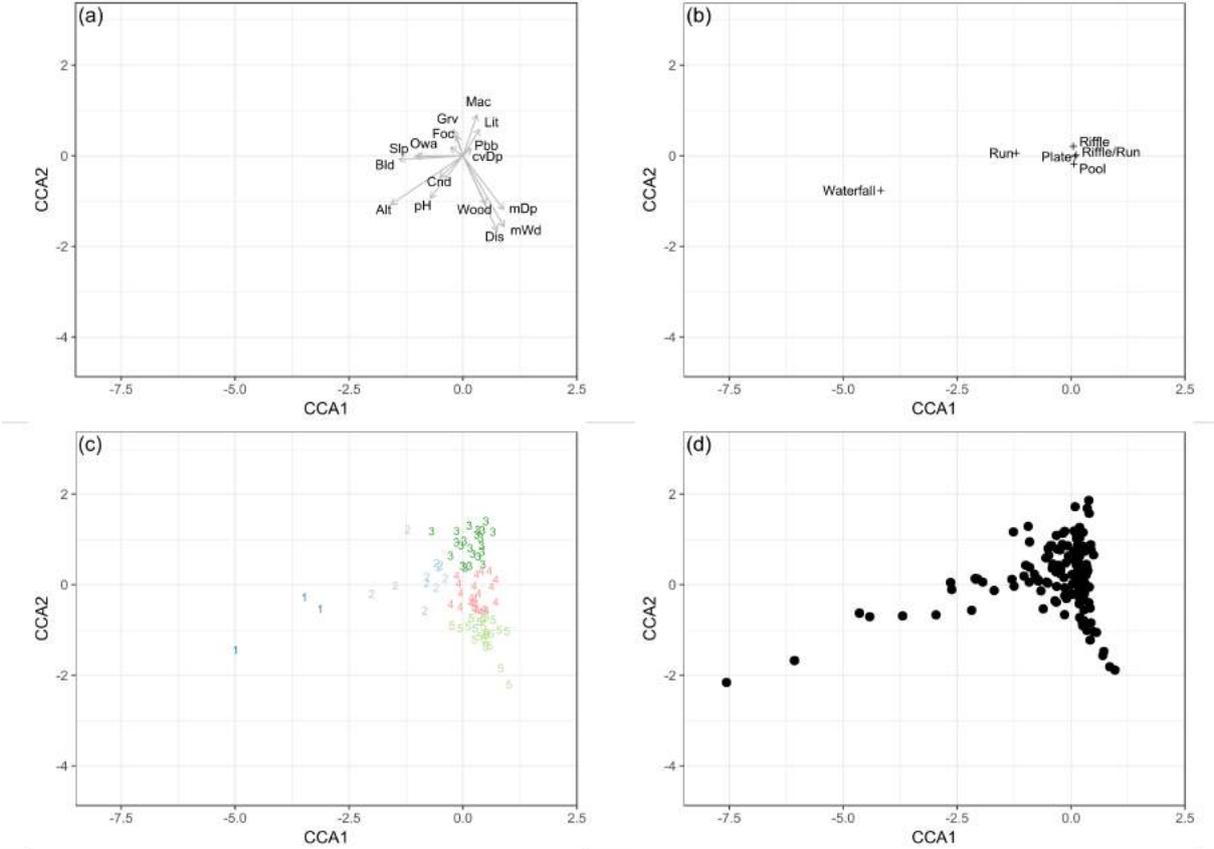
**Figure II.S1:** Species clusters (left panel) and simple structure index (ssi) values (right panel) for K ranging from two to fifteen. The first local maxima (●) and the maximal value of ssi (●) are indicated by dots and the corresponding clustering are highlighted on the left panel by black rectangles



**Figure II.S2:** Canonical correspondence analysis (CCA) ordination illustrating fish species assemblage constrained by all environmental variables. Upper left panel (a): quantitative environmental variables are represented by arrows with abbreviations as in Table II.1; Upper right panel (b): hydromorphological units; Bottom left panel (c): species position in the CCA ordination. Species were labelled with figures according to K-means analysis with K = 9 and colours correspond to the five groups used in the main text. (d): site position in the CCA ordination



**Figure II.S3:** Relationships between the number of Characiformes (a,b,c) and Siluriformes species (d,e,f) and the distance of the site from the source (a,d), the local habitat diversity (b,e) and the altitude (c,f). Fitted values (solid lines) and 95% confidence bands (dashed lines) are derived from the averaged estimates of the Poisson GLMM and are represented only for significant variables



**Figure II.S4:** Canonical correspondence analysis (CCA) ordination illustrating fish species assemblage constrained by all environmental variables while removing the species occurring in less than 5 sites. Upper left panel (a): quantitative environmental variables are represented by arrows with abbreviations as in Table II.1; Upper right panel (b): hydromorphological units; Bottom left panel (c): species position in the CCA ordination. Species were grouped according to K-means analysis with K = 5. (d): site position in the CCA ordination





# Chapter III: Assembly processes in Guianese freshwater fish assemblages

## III.1 Assembly processes between temperate and tropical freshwater fish assemblage

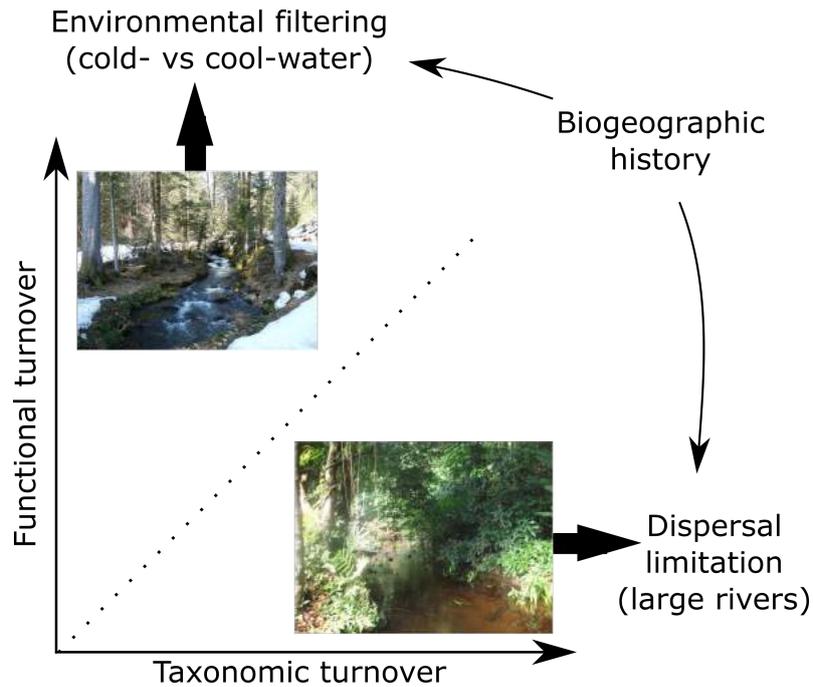
### III.1.1 Summary

Tropical regions are generally characterized by a higher species richness than temperate ones (Latitudinal Diversity Gradient, see I, but also by a higher turnover of species between assemblages (Soininen *et al.*, 2007). Differences in the strength of dispersal limitation and environmental filtering have been proposed to explain differences in beta-diversity patterns between regions. However, their relative roles remain unclear and have been shown to differ between tropical and temperate environments (Kraft *et al.*, 2011; Myers *et al.*, 2013). Using the same framework of multi-faceted diversity can help disentangle the strength of dispersal limitation and environmental filtering in these assemblages.

Using the data available for French Guiana (used in this work) and for continental France (French National Agency for Water and Aquatic Environments data for species occurrence and Toussaint *et al.* global functional database), I compare the relative strength of dispersal limitation and environmental filtering in structuring fish assemblages of small streams between the two regions. Assembly rules being sensitive to the spatial scale, I first constrained the two datasets to have about the same spatial structure and environmental characteristics. Once the sites have been selected, I compared the taxonomic and functional structures of fish assemblages to determine whether dispersal limitation and environmental filtering act in the same way in the two regions. We predict that, if taxonomic and functional turnovers between assemblages are higher in the tropical region than in the temperate region, dispersal limitation and environmental filtering have the same relative strength in the two regions. On the contrary, if functional and taxonomic turnovers are decoupled, the relative strengths of the two processes are differing between the two regions.

As expected and reported for other taxa (Ricklefs & O'Rourke, 1975; Novotny *et al.*, 2006), species richness was higher in the Guianese assemblages, at both local and regional scales, and this caused a higher functional richness for Guianese fauna compared to the French one. The higher functional richness thus represented an addition of functional attributes between the two regions caused by higher number of available niches and not by the presence of functionally distinct species.

Turning to  $\beta$ -diversity revealed that taxonomic turnover between Guianese assemblages was higher than taxonomic turnover between French assemblages. On the contrary, functional turnover was higher between French assemblages. In addition, comparisons with simulations showed that, on average, both taxonomic and functional turnovers were higher than null expectations, except for taxonomic turnover for French



**Figure III.1:** Temperate and tropical streams show different relationship between taxonomic and functional turnover, revealing different processes shaping fish assemblages. In tropical regions large rivers can act as dispersal barriers for stream fish species, promoting therefore dispersal limitation processes. In temperate streams the thermic gradient between cold upstream water and cooler downstream water that act as an environmental filter for fish assemblages. (Photo: Ji-Elle, K. Cilleros)

assemblages. Based on the above stated hypotheses, we concluded that dispersal limitation and environmental filtering did not have the same relative strength in the two regions: dispersal limitation is the main process that structures Guianese fish assemblages, whereas environmental filtering mainly constrains French assemblages.

This difference might be rooted in the difference in the habitat heterogeneity between the two regions and their past history. Guianese streams might be more locally diverse in their environment but more environmentally homogeneous between them than their temperate counterparts that exhibit a strong variability between lowland and piedmont zones, explaining the higher functional local richness but a lower functional turnover. French fish fauna also results from recent post-glacial recolonization by species with strong dispersal capacities, which can reduce the strength of dispersal limitation. In contrast, Guianese fauna is composed of species that originate from highly dynamic modification of the hydrological network (Lujan & Armbruster, 2011) and from recent dispersal events from different biogeographical regions (Amazon basin and Orinoco basin) that could have favored a high taxonomic turnover.

This first work on assembly rules and could benefit of two main improvements for further studies. Firstly, the incorporation of the phylogenetic information might help resolve underlying processes structuring local assemblages. Indeed, here we did not look if local assemblages were assembled under limiting similarity, environmental filtering or neutral process. We might expect a difference in the main process structuring local assemblages due to difference in environmental conditions between

regions (especially in seasonality and variability) and past history. Secondly, to confirm these results, it will be important to look at the relationships between beta-diversity and environmental dissimilarities and/or geographical distance between sites. If environmental filtering is dominant across assemblages, sites must have different environmental conditions even at close distance, whereas, under dispersal limitation, sites with similar environment should be separated by large distance.

The difficulty is how to define the environment in this case: does the environment must be described independently in each region or are the same measures needed? The former might be easier to implement when using databases, but such standardized environmental data is not available and this heterogeneity might blur the effect of assembly processes. The later will focus mainly on GIS derivable measures (distance from the source or to sea, slope, land use) and instream characteristics (size, bottom grain size) common to all streams. It will be easier to implement and comparisons will be based on same set of variables but this approach can miss environmental variables that affect the most assemblage structure. Depending of the organisms, one of the two solution can be favored (e.g. for tree, landscape features and soil content can be standardized across regions and can thus be used for environmental data), but for fish, the second might be the less difficult and stream characteristic are often correlated with the upstream-downstream gradient that can be described easily with GIS data.

### III.1.2 Manuscript B

#### **Taxonomic and functional diversity patterns reveal different processes shaping European and Amazonian stream fish assemblages**

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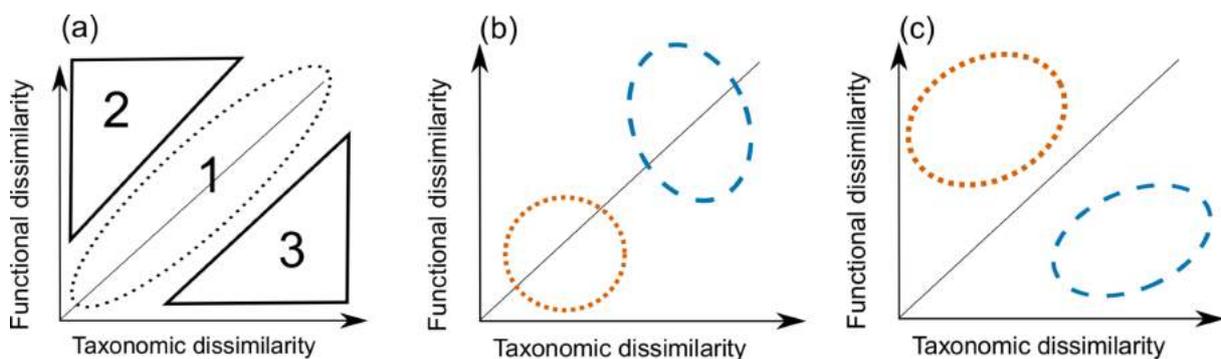
#### **INTRODUCTION**

One of the most striking characteristics of tropical regions is their species richness compared to temperate areas. The causes of these differences have been debated for more than a century, and three nonexclusive groups of hypotheses have been proposed to explain the patterns observed, i.e. historical, evolutionary and ecological hypotheses. Historical and evolutionary causes are closely linked and have been handled through a biogeographic context. Ecological hypotheses refer to mechanisms and factors that promote species presence and coexistence through their ecology, including higher available energy (Wright, 1983), more diverse habitats (Preston, 1962; MacArthur & Wilson, 1967) and/or decreased abiotic harshness in tropical regions (Fischer, 1960).

In addition to the higher local species richness in tropical than in temperate assemblages, a higher turnover in species between localities has also been reported (Soininen *et al.*, 2007). For instance, Kraft *et al.* (2011) reported a global decrease in  $\beta$ -diversity with increasing latitude, and a stronger species turnover occurs in the tropics compared to the temperate areas for both frog and fungus assemblages (Dahl *et al.*, 2009; Bahram *et al.*, 2013). Dispersal limitation and environmental filtering are the two main processes proposed to explain these patterns (Nekola & White, 1999), but their relative roles remain unclear and have been shown to differ between tropical and temperate environments (Kraft *et al.*, 2011; Myers *et al.*, 2013). For instance, Myers *et al.* (2013) reported that tropical tree assemblages are mainly structured by dispersal limitation, whereas temperate assemblages are more influenced by environmental filtering.

Functional diversity approaches have been proposed as a way to disentangle these processes, by examining the correlation between taxonomic and functional facets of diversity (Devictor *et al.*, 2010). Indeed, the assemblages filtered by the environmental characteristics should differ functionally according to the local environmental characteristics, therefore promoting functional turnover between assemblages. In contrast, assemblages dominated by dispersal limitation evolve

independently in a homogeneous environment and should therefore differentiate distinct species with similar strategies. Based on this, we predict that (1) if functional turnover and taxonomic turnover cover a similar range, environmental filtering and dispersal limitation have a similar strength (Figure III.2a), (2) if functional turnover is higher than taxonomic turnover, environmental filtering is stronger than dispersal limitation, and (3) if taxonomic turnover is higher than functional turnover, dispersal limitation is stronger than environmental filtering. Using this theoretical framework, we also predict that if both the taxonomic and the functional turnover in the tropics are higher than in the temperate zones, environmental filtering and dispersal limitation act in the same way in both environments (Figure III.2b). In contrast, if despite an increase in taxonomic turnover between temperate and tropical zones, we show an opposite trend for functional turnover, this would indicate a prominent role of dispersal limitation in tropical assemblages and of environmental filtering in temperate assemblages (Figure III.2c). Such a framework deserves to be built using phylogenetic diversity approaches (Graham & Fine, 2008), but no robust global phylogeny is currently available for South American fishes, especially Guianese ones.



**Figure III.2:** Theoretical framework (a) and expectations (b, c) about the relative strength of environmental filtering and dispersal limitation based on the relationship between taxonomic and functional turnover. (a) Assemblages with similar taxonomic and functional turnover (area 1) are structured with similar strengths of environmental filtering and dispersal limitation. Outside this area, pairs of assemblages with higher functional turnover are structured by different environmental filters and promote species with different ecological strategies (area 2), whereas pairs with higher taxonomic turnover are dominated by dispersal limitation, with distinct species sharing similar strategies (area 3). We hence expect that a higher functional turnover in tropical assemblages (blue dashed line) than in temperate ones (orange dotted line) will reveal that both processes act in the same way in the two regions (b). In contrast, a higher functional turnover in temperate assemblages will reveal that the relative strength of dispersal limitation and environmental filtering differs between the two regions (c)

Here, we compared taxonomic and functional diversity of French and Guianese stream fish assemblages. We selected a set of samples having similar topographic characteristics (altitude, stream width) and similar spatial structure (extent and grain size) between the two regions to ensure their comparability (Steinbauer *et al.*, 2012). We first quantified local and regional taxonomic and functional  $\alpha$ -diversity in the two regions. Then, we compared the spatial patterns in taxonomic and functional turnover between France and French Guiana, and inferred the processes shaping assemblage structure according to the relationships between taxonomic and functional turnover in temperate and tropical streams.

## METHODS

### Fish data

Analyses were based on complete fish inventories in small streams in both French Guiana and continental France. In both cases, fish abundances were converted into species occurrences to avoid potential bias due to differences in sampling techniques and potential differences in sampling efficiency between sites and between climate zones (Oberdorff *et al.*, 2001; Macnaughton *et al.*, 2015). Species lists obtained at the same location from 2008 to 2013 were merged to get the most complete image of fish assemblages. The Guianese database contained the occurrence of freshwater fish species in 84 stream reaches dispersed throughout French Guiana (Figure III.3a). The fish surveys that were used to build this database were conducted under several different research projects (CNRS-Nouragues, PAG-DEAL-HYDRECO project, CEBA-DIADEMA project). The sampling protocol was standardized for all sites (Allard *et al.*, 2016). At each site, a river section located upstream of a confluence was isolated using two fine mesh (4 mm) stop nets. Fish were collected after releasing a small quantity of rotenone a few metres upstream from the first net. At the end of each sampling session we searched for fishes lying on the bottom or hidden in the leaves and debris. In almost all sites, cryptic fish were collected, including highly cryptic silurids such as *Farlowella* or *Hartiella*, bottom-dwelling fishes such as *Ituglanis* or *Loricaria*, and litter bank fishes such as small killifishes (see Table III.S1 for lists of species). Gymnotids, although known as resistant to rotenone, were also captured in almost all the sites. For continental France, we used a database compiled by the French National Agency for Water and Aquatic Environments (Onema). Extensive surveys were conducted on stream sections with a standardized two-pass removal electrofishing protocol [see Poulet *et al.* (2011)]. All the fish captures were in accordance with French laws and guidelines concerning live animals, and all the experiments were approved by the Direction of Environment of the French ministry of Environment (DEAL), the French Guiana National Park (Parc Amazonien de Guyane), and the Onema.

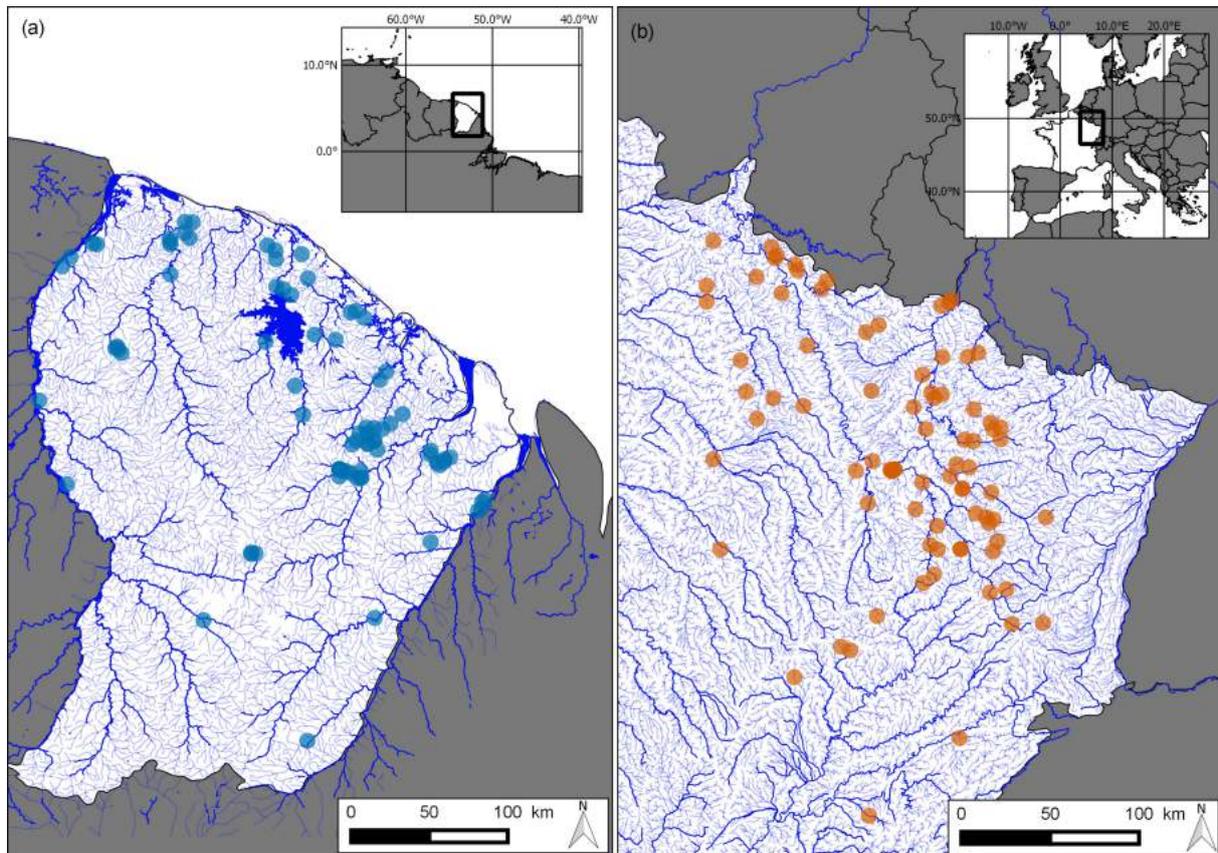
### Site selection

These two databases were built in order to characterize local fish assemblages in the two regions. French and Guianese sampling sites were thus selected to balance environmental, spatial and topographic discrepancies between the two regions that can bias comparisons between distant areas. Among the French sites, we selected those having the same physical characteristics (slope, altitude and distance from the source) and sampled during the same time period (2008 – 2013) as Guianese sites. All sites were upstream sites located between 0.5 and 16 km from the source. They belonged to lowland and piedmont rivers and were located from 27 to 633 m above sea level, with a slope between 0.57‰ and 18.77‰. Then, to ensure a similar spatial structure between Guianese and French sites, we translated Guianese site

coordinates to the part of mainland France where the spatial congruence was the best, corresponding here to the north-east of the country. We then selected French sites with more than four species (because the functional space was calculated in four dimensions, see below) that best matched Guianese site location (Figure III.3b). The resulting selection of 84 French sites showed similar spatial structure to the Guianese sites (see Table III.S2). This selection procedure provided two comparable databases of freshwater fish species occurrence in 84 assemblages for each region.

### Functional data

We characterized fish for two main functions, locomotion and food acquisition, combining 14 morpho-anatomic measures taken on a picture (lateral view) of one adult individual per species using ImageJ software<sup>1</sup> and fish size was described using the log-transformed maximum body length (values taken from Fishbase (Froese & Pauly, 2015); see Figure III.S1, Table ?? and Toussaint *et al.* (2016) for details). Juveniles were not considered as morphological changes can occur during ontogeny. The effect of intraspecific variability on our results was assessed with all the species with several individuals measured (*i.e.* all French species and 17 Guianese species). Intraspecific variation in each trait was lower than interspecific variation for all the



**Figure III.3:** The location of the 84 studied fish assemblages within French Guiana (a) and continental France (b)

<sup>1</sup><https://imagej.nih.gov/ij/index.html>

species, with interspecific variability accounting for  $75.10 \pm 18.83\%$  (mean  $\pm$  SD) of the total variability. Moreover, a simulation analysis randomly selecting individuals within species (999 replicates) showed that the French functional richness reported here was never underestimated, and overestimated in 30% of the local sites. The Guianese fish functional richness never differed. This means that intraspecific variability did not affect our conclusions, however, in 30% of the sites it did accentuate the magnitude of the difference between French and Guianese functional richness.

Morpho-anatomic measures were combined as 14 unitless ratios (see Table III.S1b). Their use has first been recommended by Webb (1984) and Winemiller (1991), and these unitless ratios still remain the most commonly used to describe fish locomotion and nutrition (e.g. Bellwood *et al.*, 2014; Brandl *et al.*, 2015; Leitão *et al.*, 2016; Toussaint *et al.*, 2016). We then computed functional distance matrix between species with Gowers distance based on the morphological ratios and the fish size to consider missing data. We used a principal coordinate analysis (PCoA) on the functional distance matrix encompassing 192 species (155 species from French Guiana and 37 species from continental France) to obtain coordinates of the species in a multidimensional space (Villéger *et al.*, 2008; Leitão *et al.*, 2016). The method of Maire *et al.* (2015) was used to evaluate the best compromise to represent the functional information and keep the number of dimensions as low as possible (and hence keep considering assemblages with a few species). It revealed that a 4-D approach was the most appropriate. We, therefore, kept the first four principal components to describe the functional space, representing 80.45% of the inertia (32.18%, 22.51%, 16.19% and 9.58% for the first four axes of the PCoA respectively). Allometric or isometric relationships existing between morpho-anatomic measures can bias species ordination in a multivariate space toward a first component representing a size effect rather than a shape or functional difference between species (Winemiller, 1991; Baur & Leuenberger, 2011). We thus checked that species ordination was not driven by a few variables having a preponderant effect by inspecting correlations between functional traits and the four PCoA components (see Table III.S4). For instance, fish body length has been considered as an integrative functional variable (Winemiller & Rose, 1992), but here its effect did not blur the effect of the others variables.

### **Taxonomic and functional diversity measures**

We defined regional richness as the total richness found in each of the two regions considered, with the number of species found in all the French and Guianese sites, respectively, being the regional taxonomic richness and the percentage of convex hull volume occupied by all the species from one region being the functional richness (Cornwell *et al.*, 2006; Villéger *et al.*, 2008). Local richness ( $\alpha$ -diversity) was defined as the taxonomic richness (the number of species) and functional richness (the volume occupied in the functional space by the species present in one local assemblage) in one stream reach assemblage.

Pairwise taxonomic  $\beta$ -diversity (hereafter  $\beta T_j$ ) between two assemblages was computed using Jaccard's dissimilarity index. This index is widely used in ecology (Koleff *et al.*, 2003; Myers *et al.*, 2013) and its meaning is easily understandable (the percentage of unshared species between two assemblages). It is based on a broad-sense measure of dissimilarity (Koleff *et al.*, 2003), representing the sum of the species replacement between two assemblages (hereafter called turnover) and the difference in species richness between them (**nestedness**). The turnover was calculated according to Baselga (2012):

$$\text{taxonomic turnover} = \frac{2 \times \min(b, c)}{a + 2 \times \min(b, c)}$$

Taxonomic turnover is null when one assemblage contains all the species from the other and equals 1 (*i.e.*  $\beta T_j$ ) when assemblages have no species in common.

We then calculated the relative contribution of the turnover component to overall dissimilarity ( $p_{\text{turn}}$ ) to get an easily interpretable measure of the importance of species turnover on  $\beta$ -diversity (Toussaint *et al.*, 2014), with:

$$p_{\text{turn}} = \frac{\text{taxonomic turnover}}{\beta T_j}$$

The  $p_{\text{turn}}$  metric is defined only if dissimilarity is not null, meaning that a pair of assemblages differs by at least one species. It is null when assemblages are nested and it equals 1 when assemblages have the same species richness (taxonomic nestedness component is null) or when they have no species in common.

We computed pairwise functional  $\beta$ -diversity ( $\beta F_j$ ) within the same framework as for taxonomy, but using convex hull volumes of assemblage instead of species richness (Villéger *et al.*, 2011a). So  $\beta F_j$  can be computed with the a, b and c quantities with a being the intersection of volumes of two assemblages, and b and c the volumes unique to each assemblage. Functional dissimilarity will be equal to 1 when volumes do not overlap and 0 when volumes are identical. The same decomposition in turnover and nestedness of functional dissimilarity were also computed as for taxonomy (Villéger *et al.*, 2013). Functional turnover here represents the overlap within functional spaces between assemblages. The relative contribution of the functional turnover component is computed in the same way as its taxonomic counterpart.

### Statistical analysis

We compared functional richness at the regional scale while accounting for differences in species richness. We randomly selected as many Guianese species as French species and then calculated the functional richness that they represented. We repeated this 9,999 times and compared the regional functional richness of continental France to this distribution. For the local functional richness, we used the same approach by replacing the French species by the selected Guianese species, thus conserving the occurrence matrix. We then calculated local functional richness

for each site and compared, for each site, the French local functional richness to the simulated values for the corresponding sites.

To check for the potential effect of the difference in the number of drainage basins between the two regions on taxonomic and functional diversity measures, taxonomic pairwise comparisons were classified in either intra- or inter-basin comparison. We tested whether intra-basin and inter-basin comparisons significantly differed for each region with a permutation procedure as implemented in 'diffmean' function in R package 'simba' (Jurasinski & Retzer, 2012) with 9,999 permutations.

We tested the difference in  $\beta T_j$ ,  $\beta F_j$  and their turnover component between continental France and French Guiana using analysis of homogeneity of multivariate dispersion based on distance-to-centroids (Anderson *et al.*, 2011). In this context,  $\beta$ -diversity was defined as the average distance of sites from the centroid formed by all the sites of a group. Distance-to-centroid and average dissimilarity were highly correlated (Pearson's correlation  $> .92$ ,  $p < .001$  for each metric). We thus used Jaccard's framework to represent graphically  $\beta$ -diversity.

Taxonomic and functional dissimilarity and turnover were regressed against the logarithm of the pairwise geodesic distance between sites to test the difference in spatial pattern between continental France and French Guiana. We used the Mantel test with 9,999 permutations and the Spearman correlation coefficient to test the significance of the relationship between dissimilarity and distance. Intercept and slope were estimated with a 95% confidence interval by bootstrap with 9,999 replicates (Davidar *et al.*, 2007). Differences in slope and intercept between continental France and French Guiana were tested using 9,999 permutations, following Nekola & White (1999) with 'diffslope' and 'diffic' functions. For each run, couples of value dissimilarity/turnover - distance were randomly attributed to one of the two regions and the difference in slope or intercept was calculated. The differences calculated were used as a null distribution to compare with observed difference.

We tested whether observed turnover differed from random expectations using two null models to disentangle which processes had a predominant influence on community assembly between environmental filtering, dispersal limitation and stochastic processes. The first model tested if the observed taxonomic turnover differed from random sampling of species from the regional pool and evaluated the strength of both dispersal limitation and environmental filtering compared to stochasticity. For this, we constructed random assemblages by maintaining the species richness observed in the sites and the species occurrence in the database using a trial swap algorithm (Miklós & Podani, 2004). The second model tested if observed functional turnover differed from expectation where species traits are randomly distributed among assemblages and thus discriminate environmental filtering from stochastic processes. We permuted species identity in the species-trait matrix while maintaining the sites-species matrix and thus the local species richness and taxonomic turnover. For each null model, we calculated a standardized effect size (SES) as the difference between observed values and the mean of 999 simulated values, divided by the standard deviation of the simulated values. Pairs with an SES

value between -1.96 and 1.96 did not differ significantly from random expectations. The medians of SES values were tested for departure from zero to assess if environmental filtering or dispersal limitation govern community assembly (Stegen *et al.*, 2012). In contrast, no departure from zero would indicate that stochastic processes govern community assembly.

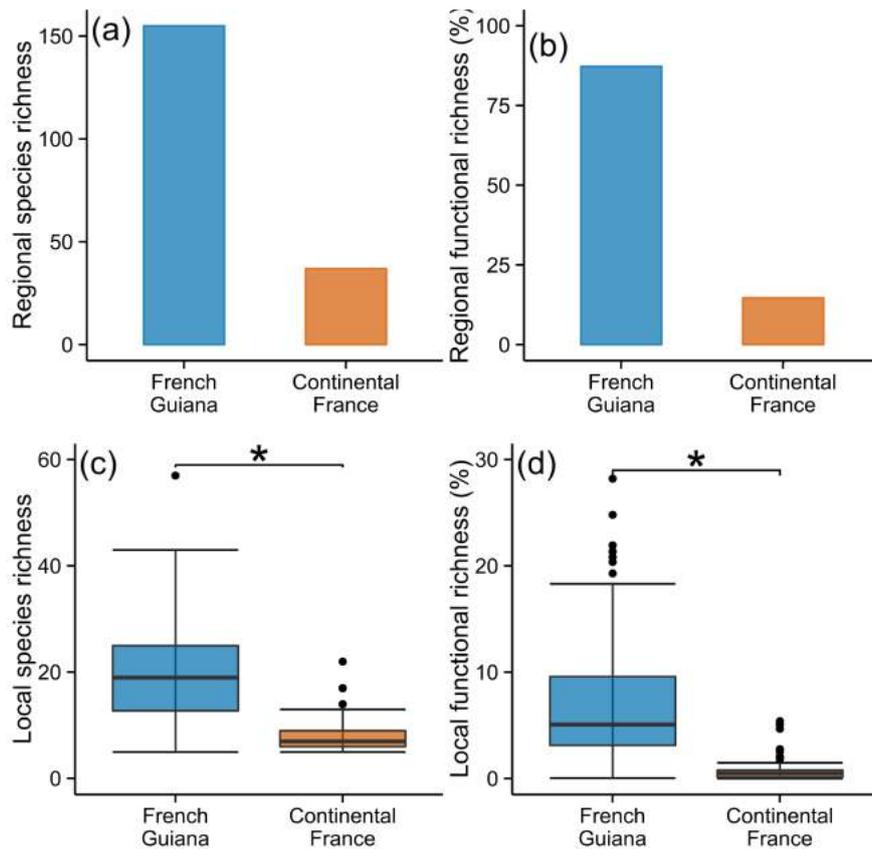
## RESULTS

Regional species richness was higher in French Guiana than in continental France, with 155 and 37 species respectively (Figure III.4a). Regional functional richness showed a similar pattern. Guianese species occupied 87.29% of the total functional volume, whereas French species accounted for 14.77% of this volume (Figure III.4b). It should be noted that the ratio between Guianese and French diversity increased when turning from taxonomic to functional diversity (4.19 vs. 5.91). Moreover, the functional space occupied by French species was largely included in the functional space of Guianese species. Indeed, only 4.88% of the French functional space was not shared with Guianese functional space (Figure III.5, see Figure III.S2). However, controlling for species richness differences between the two regions revealed that the regional functional richness of French Guiana was not different from the functional richness of continental France ( $p = .21$ ), and hence the functional differences observed between the two regions were due to a species richness effect.

At the local scale, French assemblages contained fewer species ( $7.88 \pm 3.05$ ) than Guianese assemblages ( $19.92 \pm 9.32$ ) (Figure III.4c; Wilcoxon rank-sum test:  $z = 9.46$ ,  $p < .001$ ) which were less functionally diverse ( $.74 \pm 1.02\%$  vs.  $7.26 \pm 6.28\%$ ) (Figure III.4d; Wilcoxon rank-sum test:  $z = 9.64$ ,  $p < .001$ ). Again, the distinction between Guianese and French streams was more marked from a functional than from a taxonomic point of view. Accounting for species richness differences by randomly replacing French species by Guianese species showed that only four French sites had a lower local functional diversity ( $p < .05$ ). The functional diversity of the 80 remaining French sites did not significantly differ from a random replacement of French species by Guianese species. Hence, the functional differences reported between Guianese and French sites were mainly due to a species richness effect.

Testing the effect of the difference in the number of drainage basins revealed that both  $\beta T_j$  and taxonomic turnover showed higher mean inter-basin dissimilarity values than intra-basin values for continental France (permutation tests:  $\beta T_j$ ,  $F = 52.4$ ,  $p < .001$ ; taxonomic turnover,  $F = 43.6$ ,  $p < .001$ ) and French Guiana ( $\beta T_j$ ,  $F = 14.2$ ,  $p < .001$ ; taxonomic turnover,  $F = 44.6$ ,  $p < .001$ ).

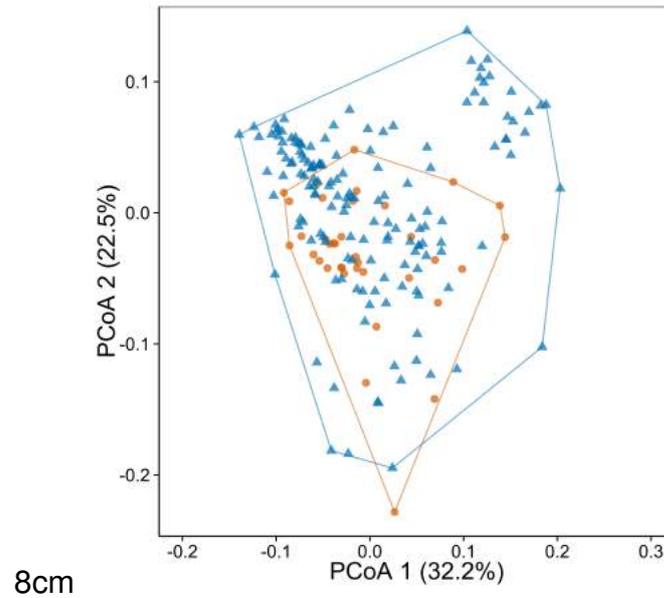
$\beta T_j$  and taxonomic turnover were higher for Guianese assemblages ( $\beta T_j = .84 \pm .096$ ; taxonomic turnover =  $.76 \pm .14$ ) than for French assemblages ( $\beta T_j = .65 \pm .16$ ; taxonomic turnover =  $.52 \pm .23$ ) (Figure III.6a; homogeneity of multivariate dispersion test on average distance-to-centroids:  $F = 117.17$ ,  $p < .001$  for  $\beta T_j$  and  $F = 86.77$ ,  $p < .001$  for taxonomic turnover). Contribution of turnover to taxonomic dissimilarity was high for both regions, but its contribution was higher for French Guiana ( $.91 \pm$



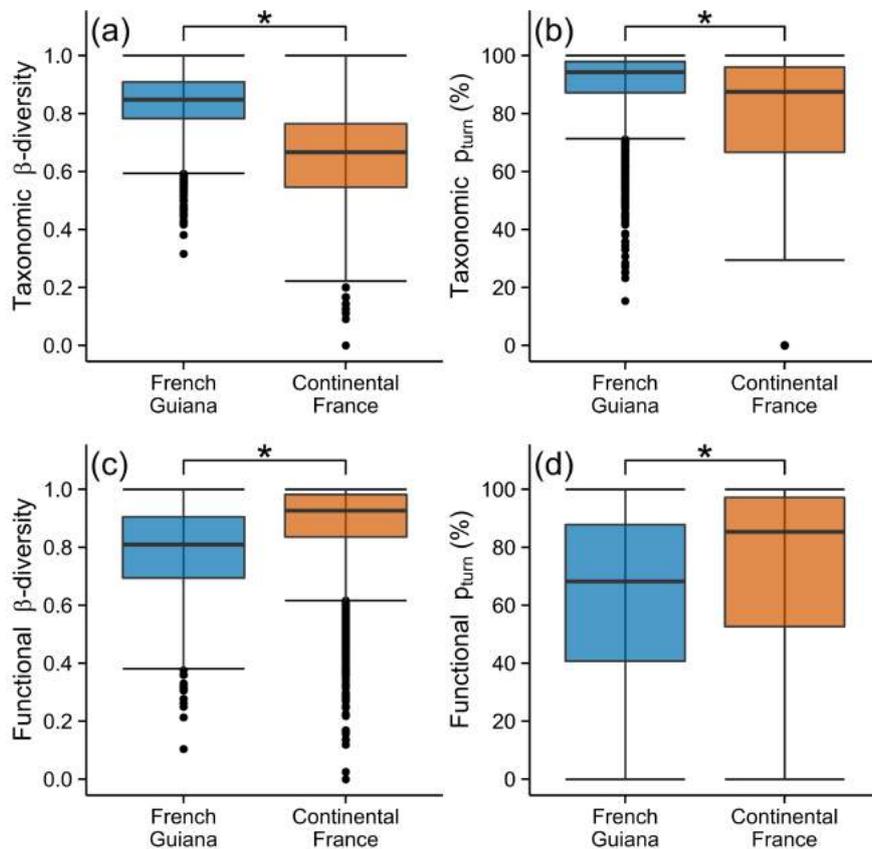
**Figure III.4:** Taxonomic and functional diversity of fish at regional scale (a, b) and at local scale (c, d). The functional richness represents the percentage of the convex hull volume occupied by all the species of one region (b) or of one site (d) out of the total convex hull volume of all the species. Significant differences between the two regions are indicated above the boxes (\*)

.11) than for continental France ( $.77 \pm .26$ ) (Figure III.6b; permutation test:  $F = 738$ ,  $p < .001$ ). In contrast,  $\beta F_j$  and functional turnover were greater for French assemblages ( $\beta F_j = .88 \pm .14$ ; functional turnover =  $.63 \pm .33$ ) than for Guianese assemblages ( $\beta F_j = .79 \pm .14$ ; functional turnover =  $.49 \pm .25$ ) (Figure III.6c; homogeneity of multivariate dispersion test on average distance-to-centroids:  $F = 77.99$ ,  $p < .001$  for  $\beta F_j$  and  $F = 47.42$ ,  $p < .001$  for functional turnover). Contribution of turnover to functional dissimilarity was lower for French Guiana ( $.62 \pm .29$ ) than for continental France ( $.70 \pm .33$ ) (Figure III.6d; permutation test:  $F = 114$ ,  $p < .001$ ).

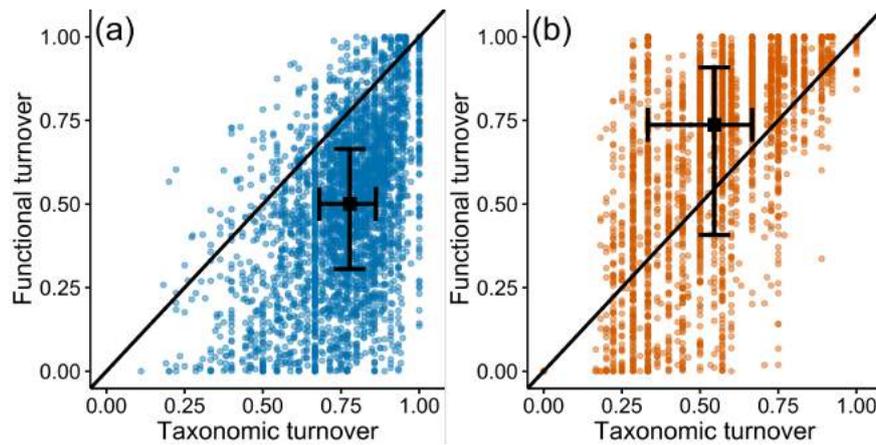
For both French Guiana and continental France,  $\beta F_j$  and  $\beta T_j$  were significantly correlated (French Guiana:  $r = .60$ ,  $p < .001$ ; continental France:  $r = .56$ ,  $p < .001$ ) as well as taxonomic and functional turnover components (French Guiana:  $r = .49$ ,  $p < .001$ ; continental France:  $r = .72$ ,  $p < .001$ ). However, the magnitude of the taxonomic and of the functional  $\beta$ -diversity and turnover differed between the two regions. For French Guiana, taxonomic  $\beta$ -diversity and turnover were higher than functional  $\beta$ -diversity and turnover ( $\beta F_j - \beta T_j$ : Wilcoxon signed-rank test:  $z = -21.62$ ,  $p < .001$ ; turnover: Figure III.7a;  $z = 48.05$ ,  $P < .001$ ). In contrast, in continental France, functional  $\beta$ -diversity and turnover were higher than taxonomic  $\beta$ -diversity and



**Figure III.5:** Ordination of fish species along the axes of the first two principal components (representing 54.7% of the total inertia) based on the functional distance matrix computed with Gower's distance on the 15 morpho-anatomic measures. Convex hulls for continental France (●) and French Guiana (▲) are represented



**Figure III.6:** Pairwise taxonomic and functional  $\beta$ -diversity among the 84 fish assemblages (a, c) and the relative contribution of the turnover component to overall dissimilarity ( $p_{turn}$ ) (b, d) for continental France and French Guiana. Significant differences between the two regions are indicated above the boxes (\*), and based on the homogeneity of multivariate dispersion test on average distance-to-centroids and permutation procedure



**Figure III.7:** Relationships between pairwise taxonomic and functional turnover among the 84 fish assemblages for French (a) and continental France (b). Squares and bars represent medians and 25<sup>th</sup>/75<sup>th</sup> percentiles respectively. The line 1:1 is represented

turnover ( $\beta F_j - \beta T_j$ :  $z = 48.05$ ,  $p < .001$ ; turnover: Figure III.7b;  $z = -.52$ ,  $p < .001$ ).

We found significant distance decay relationships for the Guianese  $\beta T_j$  and taxonomic turnover for both regions (Table III.1). Slopes of the regressions of  $\beta T_j$  and taxonomic turnover did not differ between continental France and French Guiana ( $\beta T_j$ :  $p = .074$ ; taxonomic turnover:  $p = .12$ ), but the intercept differed ( $\beta T_j$ :  $p < .001$ ; taxonomic turnover:  $p < .001$ ), being higher for French Guiana. Both slopes and intercepts differed for  $\beta F_j$  (slope:  $p = .0033$ ; intercept:  $p = .023$ ), with higher intercept and a more pronounced distance decay for French assemblages. The slope for functional turnover in French assemblages was higher than the slope for French Guiana ( $p < .001$ ), but the intercept did not differ between the two regions ( $p = .13$ ).

For French Guiana, null models showed that 23 comparisons of 3486 for taxonomic turnover were significantly different from random expectations. The median of distribution differed significantly from zero (median = .10,  $z = 5.99$ ,  $p < .001$ ). A greater number of pairs of communities were different from random expectation when looking at functional diversity turnover while conserving the observed taxonomic turnover (131 out of 3486). The median of simulated functional turnover distribution was significantly different from zero (median = .19,  $z = 7.79$ ,  $p < .001$ ). In continental France, 197 taxonomic turnover measures were not consistent with random assembly. Taxonomic turnover distribution showed a median of .052 that did not differ from zero ( $z = .47$ ,  $p = .64$ ). A lower number of pairs of communities were different from random expectation when looking at the functional diversity facet while conserving the observed taxonomic  $\beta$ -diversity (113 pairs). However, the median of the simulated distribution was significantly different from zero (functional turnover: median = .50,  $z = 17.88$ ,  $p < .001$ ).

## DISCUSSION

A major limitation in studies comparing biodiversity patterns from distant areas is the availability of databases collected at similar grain and extents. The total species pool and the distance between samples are affected by the geographical extent of the

**Table III.1:** Regression parameters of the relationship between diversity metrics ( $\beta T_j$ ,  $\beta F_j$  and turnover) of the fish faunas and the pairwise geodesic distance (logged value) between sites for continental France and French Guiana estimated by bootstrap based on 9,999 permutations. Spearman correlation coefficient  $r$  and  $p$  were obtained from Mantel tests with 9,999 permutations

Metric	Region	Intercept (95% CI)	Slope (95% CI)	$r$	$p$
$\beta T_j$	Continental France	.54 (.51 – .57)	.026 (.019 – .032)	.069	.069
	French Guiana	.75 (.73 – .76)	.021 (.017 – .024)	.13	.0043
Taxonomic turnover	Continental France	.37 (.33 – .41)	.034 (.025 – .043)	.087	.034
	French Guiana	.64 (.61 – .66)	.029 (.024 – .034)	.14	.001
$\beta F_j$	Continental France	.80 (.77 – .84)	.018 (.010 – .026)	.015	.39
	French Guiana	.76 (.74 – .79)	.0064 (.0016 – .011)	.026	.31
Functional turnover	Continental France	.48 (.42 – .54)	.034 (.021 – .049)	.034	.23
	French Guiana	.44 (.41 – .48)	.010 (.0024 – .018)	.040	.20

studies (Steinbauer *et al.*, 2012), and grain size affects  $\alpha$ -diversity through a species-area effect, with an increase in habitat heterogeneity (Devictor *et al.*, 2010).  $\beta$ -diversity is also sensitive to grain size as smaller grains are less species-rich than larger ones and thus more likely to share fewer species between each other than larger grains (Garzon-Lopez *et al.*, 2014). In addition, to compare community assembly processes across regions, data should also encompass similar environmental or topographic gradients between regions (e.g. altitude range or stream order), as these gradients have a profound effect on assemblages structure (Melo *et al.*, 2009; Jaramillo-Villa *et al.*, 2010). Here, these potential biases were controlled by collecting European and South American data at the same grain size and by constraining European data to fit the spatial extent and the topographic gradient encompassed by South American sites, therefore allowing us to explore differences between temperate and tropical faunas independently of scale and environmental gradient effects.

The higher taxonomic diversity we report in Guiana compared to France at both regional and local grains is known for most taxa (Ricklefs & O'Rourke, 1975; Novotny *et al.*, 2006). A similar trend is also forecast for functional richness, which may be positively related to the number of species (Safi *et al.*, 2011; Lamanna *et al.*, 2014). The differences we report between temperate and tropical environments increased when turning from taxonomic to functional diversity, at both regional and local scales. However, when controlling for the number of species, the Guianese fish fauna is not more functionally diverse than the French fauna. This was also the case at the local

scale and suggests that the higher functional diversity in Guiana mainly results from an addition of functional attributes across the region. The greater functional richness found in the tropical region was hence not driven by a triggered functional diversity of the tropical species, but more probably by a higher diversity of habitats which allows a greater diversity of ecological niches and thus a greater number of species (Koeck *et al.*, 2014).

Turning from  $\alpha$ - to  $\beta$ -diversity patterns sheds light on the assembly processes that structure the fish faunas. Species replacement between sites (turnover) was higher in Guianese assemblages, which are thus composed of a higher number of species whose identity differed in a large part between sites. In contrast, functional turnover was higher in French assemblages than in Guianese ones. According to our predictions, Guianese fish assemblages result from a prominent effect of dispersal limitation that promotes a stronger taxonomic turnover than functional turnover (Swenson *et al.*, 2012b; Myers *et al.*, 2013). However, French assemblages appear more strongly influenced by environmental filtering that promotes lower taxonomic turnover than functional turnover (Swenson *et al.*, 2012a; Myers *et al.*, 2013). Null models confirmed this with species more aggregated within assemblages than expected in French Guiana and a greater functional dissimilarity than expected between assemblages in continental France.

The differences in diversity patterns and processes governing assemblage structure between the two regions might root in their biogeographical history and evolutionary causes. First, tropical climates have been hypothesized to have been more stable than temperate climates and thus led a longer time for species diversification than temperate regions (Gaston & Blackburn, 1996). Moreover, faster diversification in tropical regions has been found in equal time periods, linked with less drastic extinction events during Quaternary climatic fluctuations (Rolland *et al.*, 2014). Secondly, the Guianese ichthyofauna results from the ancient complex biogeographical history of the Neotropical region, involving events of marine incursions, uplift of the Andes and the raising of palaeoarches that modified South America's hydrological network (Lujan & Armbruster, 2011), and from more recent dispersal events via coastal routes from the Amazon in the south and from the Orinoco in the Northwest that occurred during the Miocene (Hubert & Renno, 2006; Cardoso & Montoya-Burgos, 2009). The current fauna thus presents different species pools that contribute to the high regional taxonomic  $\alpha$ - and  $\beta$ -diversity in French Guiana. The studied French fauna on the other hand results mainly from a more recent recolonization of Western Europe by a single Danubian species pool after the Last Glacial Maximum (26,500 – 19,000 years ago) (Taberlet *et al.*, 1998; Kotlík & Berrebi, 2001). The recolonization from a single pool of species hence reduced both the richness in species and the taxonomic turnover across Western Europe.

Although differences in biogeographical history explain taxonomic patterns, they do not provide information on assembly rule processes. Here, we show that in Guianese streams, despite the density of the river network that could favour faunistic exchanges between basins, the local fish assemblages were affected of by both inter-

and intra-basin dispersal limitation. Despite this high turnover in species composition, we report a moderate functional turnover in Guianese streams. Functional diversity represents the range of ecological strategies of the species (Violle *et al.*, 2007; Cadotte *et al.*, 2011; Mouillot *et al.*, 2011) and thus niche availability in local habitats. The high local species richness in Guianese streams hence testifies for a high local diversity of niches, but the same niches are probably shared by most sites explaining therefore the low functional  $\beta$ -diversity (Weinstein *et al.*, 2014). The limited effect of environmental filtering may be due to the relative homogeneity in available habitats between sites. Guianese fish fauna, even historically resulting from different biogeographical origins, is thus composed of species that converged in the use of similar niches, and the co-occurrence of those species being allowed by the high complexity in local habitats.

In the French fish assemblages, functional turnover was higher than taxonomic turnover meaning that the fauna exhibited local adaptations to different environments. Such environmental filtering might be due to the low similarity of available niches between sites (Weinstein *et al.*, 2014). Indeed, under temperate climates, the thermal regime of streams can be markedly different between lowland and piedmont areas, therefore constituting a strong environmental filter for the fauna. Cool-water (lowland) and cold-water (piedmont and mountain) streams are indeed inhabited by phylogenetically and functionally distinct fish faunas (Blanchet *et al.*, 2014). Moreover, the reduced strength of dispersal limitation in France also has historical roots, as the French fauna results from a recent post-glacial recolonization by high-dispersal species (Griffiths, 2006; Hof *et al.*, 2008), that are hence likely to cross longer distances than Guianese stream fishes.

The use of phylogenetic diversity in combination with taxonomic and functional diversity may allow a more complete view of community assembly rules to be assessed (Weinstein *et al.*, 2014). An assumption behind this framework is that closely related species tend to have similar functional traits and ecological niches (Webb, 2000). If true, then environmental filtering should result in communities with more closely related species than randomly assembled communities. Conversely, communities with more distant-related species than random communities should be driven by competitive interactions. So, developing and using a robust phylogeny of freshwater fish present in both regions (or for all fish species) will be a major future step in comparative analyses of assembly processes.

Beyond their conceptual interest, understanding the processes affecting species assembly are also informative in a biodiversity conservation context (Mouillot, 2007; Rossetto *et al.*, 2008). Indeed, faunas assembled through dispersal limitation, such as Guianese stream fishes, are sensitive to local extinctions, because the recolonization of disturbed sites from distant assemblages is almost impossible. Therefore, the restoration of sites after human disturbance might not result in the re-establishment of the initial fauna. This might explain why local studies report limited post-disturbance recolonization, even after a complete recovery of the stream physical and chemical characteristics (Brosse *et al.*, 2011; Allard *et al.*, 2016). Particular attention should

therefore be devoted to the conservation of pristine sites to avoid local species extirpations. In contrast, in Europe, restoring and managing habitat diversity may be a priority, as assemblages being less constrained by dispersal limitations could have more chance to recolonize restored streams.

### **ACKNOWLEDGMENTS**

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**Table III.S1:** List of the fish species in continental France (a) and French Guiana (b) studied regions

## (a) Continental France

<b>Order</b>	<b>Genus species</b>
Anguilliformes	<i>Anguilla anguilla</i> (Linnaeus, 1758)
Cypriniformes	<i>Abramis brama</i> (Linnaeus, 1758)
Cypriniformes	<i>Alburnoides bipunctatus</i> (Bloch, 1782)
Cypriniformes	<i>Alburnus alburnus</i> (Linnaeus, 1758)
Cypriniformes	<i>Barbatula barbatula</i> (Linnaeus, 1758)
Cypriniformes	<i>Barbus barbus</i> (Linnaeus, 1758)
Cypriniformes	<i>Blicca bjoerkna</i> (Linnaeus, 1758)
Cypriniformes	<i>Carassius auratus</i> (Linnaeus, 1758)
Cypriniformes	<i>Carassius gibelio</i> (Bloch, 1782)
Cypriniformes	<i>Chondrostoma nasus</i> (Linnaeus, 1758)
Cypriniformes	<i>Cobitis taenia</i> (Linnaeus, 1758)
Cypriniformes	<i>Cyprinus carpio</i> (Linnaeus, 1758)
Cypriniformes	<i>Gobio gobio</i> (Linnaeus, 1758)
Cypriniformes	<i>Leucaspius delineatus</i> (Heckel, 1843)
Cypriniformes	<i>Leuciscus idus</i> (Linnaeus, 1758)
Cypriniformes	<i>Leuciscus leuciscus</i> (Linnaeus, 1758)
Cypriniformes	<i>Phoxinus phoxinus</i> (Linnaeus, 1758)
Cypriniformes	<i>Rhodeus amarus</i> (Bloch, 1782)
Cypriniformes	<i>Rutilus rutilus</i> (Linnaeus, 1758)
Cypriniformes	<i>Scardinius erythrophthalmus</i> (Linnaeus, 1758)
Cypriniformes	<i>Squalius cephalus</i> (Linnaeus, 1758)
Cypriniformes	<i>Telestes souffia</i> (Risso, 1827)
Cypriniformes	<i>Tinca tinca</i> (Linnaeus, 1758)
Esociformes	<i>Esox lucius</i> (Linnaeus, 1758)
Gadiformes	<i>Lota lota</i> (Linnaeus, 1758)
Gasterosteiformes	<i>Gasterosteus aculeatus</i> (Linnaeus, 1758)
Gasterosteiformes	<i>Pungitius pungitius</i> (Linnaeus, 1758)
Perciformes	<i>Gymnocephalus cernua</i> (Linnaeus, 1758)
Perciformes	<i>Lepomis gibbosus</i> (Linnaeus, 1758)
Perciformes	<i>Perca fluviatilis</i> (Linnaeus, 1758)
Petromyzontiformes	<i>Lampetra planeri</i> (Bloch, 1784)
Salmoniformes	<i>Oncorhynchus mykiss</i> (Walbaum, 1792)
Salmoniformes	<i>Salmo trutta</i> (Linnaeus, 1758)
Salmoniformes	<i>Thymallus thymallus</i> (Linnaeus, 1758)
Scorpaeniformes	<i>Cottus gobio</i> (Linnaeus, 1758)
Siluriformes	<i>Ameiurus melas</i> (Rafinesque, 1820)
Siluriformes	<i>Silurus glanis</i> (Linnaeus, 1758)

Table III.S1 continued

(b) French Guiana

Order	Genus species
Beloniformes	<i>Potamorhaphis guianensis</i> (Jardine, 1843)
Characiformes	<i>Acestrorhynchus falcatus</i> (Bloch, 1794)
Characiformes	<i>Anostomus brevior</i> (Gry, 1963)
Characiformes	<i>Astyanax bimaculatus</i> (Linnaeus, 1758)
Characiformes	<i>Astyanax leopoldi</i> (Gry, Planquette & Le Bail, 1991)
Characiformes	<i>Astyanax validus</i> (Gry, Planquette & Le Bail, 1991)
Characiformes	<i>Bryconamericus aff. hyphesson</i>
Characiformes	<i>Bryconamericus guyanensis</i> (Zarske, Le Bail & Gry, 2010)
Characiformes	<i>Bryconops aff. caudomaculatus</i> (Gnther, 1864)
Characiformes	<i>Bryconops affinis</i> (Gnther, 1864)
Characiformes	<i>Bryconops caudomaculatus</i> (Gnther, 1864)
Characiformes	<i>Bryconops melanurus</i> (Bloch, 1794)
Characiformes	<i>Characidium zebra</i> (Eigenmann, 1909)
Characiformes	<i>Creagrutus melanzonus</i> (Eigenmann, 1909)
Characiformes	<i>Creagrutus planquettei</i> (Gry & Renno, 1989)
Characiformes	<i>Curimatopsis crypticus</i> (Vari, 1982)
Characiformes	<i>Cynopotamus essequibensis</i> (Eigenmann, 1912)
Characiformes	<i>Cyphocharax aff. spilurus</i>
Characiformes	<i>Cyphocharax gouldingi</i> (Vari, 1992)
Characiformes	<i>Cyphocharax helleri</i> (Eigenmann & Eigenmann, 1889)
Characiformes	<i>Cyphocharax spilurus</i> (Gnther, 1864)
Characiformes	<i>Erythrinus erythrinus</i> (Bloch & Schneider, 1801)
Characiformes	<i>Gasteropelecus sternicla</i> (Linnaeus, 1758)
Characiformes	<i>Hemibrycon surinamensis</i> (Gry, 1962)
Characiformes	<i>Hemigrammus boesemani</i> (Gry, 1959)
Characiformes	<i>Hemigrammus guyanensis</i> (Gry, 1959)
Characiformes	<i>Hemigrammus ocellifer</i> (Steindachner, 1882)
Characiformes	<i>Hemigrammus ora</i> (Zarske, Le Bail & Gry, 2006)
Characiformes	<i>Hemigrammus rodwayi</i> (Durbin, 1909)
Characiformes	<i>Hemigrammus unilineatus</i> (Gill, 1858)
Characiformes	<i>Hemiodus quadrimaculatus</i> (Pellegrin, 1909)
Characiformes	<i>Hoplerythrinus unitaeniatus</i> (Spix & Agassiz, 1829)
Characiformes	<i>Hoplias aimara</i> (Valenciennes, 1847)
Characiformes	<i>Hoplias malabaricus</i> (Bloch, 1794)
Characiformes	<i>Hyphessobrycon borealis</i> (Zarske, Le Bail & Gry, 2006)
Characiformes	<i>Hyphessobrycon copelandi</i> (Durbin, 1908)

Table III.S1 continued

Characiformes	<i>Hyphessobrycon roseus</i> (Gry, 1960)
Characiformes	<i>Hyphessobrycon simulatus</i> (Gry, 1960)
Characiformes	<i>Hypomasticus despaxi</i> (Puyo, 1943)
Characiformes	<i>Jupiaba abramoides</i> (Eigenmann, 1909)
Characiformes	<i>Jupiaba keithi</i> (Gry, Planquette & Le Bail, 1996)
Characiformes	<i>Jupiaba meunieri</i> (Gry, Planquette & Le Bail, 1996)
Characiformes	<i>Leporinus acutidens</i> (Valenciennes, 1837)
Characiformes	<i>Leporinus friderici</i> (Bloch, 1794)
Characiformes	<i>Leporinus gossei</i> (Gry, Planquette & Le Bail, 1991)
Characiformes	<i>Leporinus granti</i> (Eigenmann, 1912)
Characiformes	<i>Leporinus lebaili</i> (Gry & Planquette, 1983)
Characiformes	<i>Leporinus nijsseni</i> (Garavello, 1990)
Characiformes	<i>Leporinus pellegrini</i> (Steindachner, 1910)
Characiformes	<i>Melanocharacidium cf. blennioides</i>
Characiformes	<i>Melanocharacidium dispilomma</i> (Buckup, 1993)
Characiformes	<i>Microcharacidium eleotrioides</i> (Gry, 1960)
Characiformes	<i>Moenkhausia aff. grandisquamis</i>
Characiformes	<i>Moenkhausia aff. intermedia</i>
Characiformes	<i>Moenkhausia chrysargyrea</i> (Günther, 1864)
Characiformes	<i>Moenkhausia collettii</i> (Steindachner, 1882)
Characiformes	<i>Moenkhausia georgiae</i> (Gry, 1965)
Characiformes	<i>Moenkhausia hemigrammoides</i> (Gry, 1965)
Characiformes	<i>Moenkhausia moisae</i> (Gry, Planquette & Le Bail, 1995)
Characiformes	<i>Moenkhausia oligolepis</i> (Günther, 1864)
Characiformes	<i>Moenkhausia surinamensis</i> (Gry, 1965)
Characiformes	<i>Myloplus ternetzi</i> (Norman, 1929)
Characiformes	<i>Nannostomus beckfordi</i> (Günther, 1872)
Characiformes	<i>Nannostomus bifasciatus</i> (Hoedeman, 1954)
Characiformes	<i>Parodon guyanensis</i> (Gry, 1959)
Characiformes	<i>Phenacogaster wayampi</i> (Le Bail & Lucena, 2010)
Characiformes	<i>Phenacogaster wayana</i> (Le Bail & Lucena, 2010)
Characiformes	<i>Poptella brevispina</i> (Reis, 1989)
Characiformes	<i>Pristella maxillaris</i> (Ulrey, 1894)
Characiformes	<i>Pyrrhulina filamentosa</i> (Valenciennes, 1847)
Characiformes	<i>Roeboexodon guyanensis</i> (Puyo, 1948)
Characiformes	<i>Serrapinnus gracilis</i> (Gry, 1960)
Characiformes	<i>Steindachnerina varii</i> (Gry, Planquette & Le Bail, 1991)
Characiformes	<i>Thayeria ifati</i> (Gry, 1959)
Cichliformes	<i>Aequidens tetramerus</i> (Heckel, 1840)

Table III.S1 continued

Cichliformes	<i>Apistogramma gossei</i> (Kullander, 1982)
Cichliformes	<i>Cichlasoma bimaculatum</i> (Linnaeus, 1758)
Cichliformes	<i>Cleithracara maronii</i> (Steindachner, 1881)
Cichliformes	<i>Crenicichla albopunctata</i> (Pellegrin, 1904)
Cichliformes	<i>Crenicichla johanna</i> (Heckel, 1840)
Cichliformes	<i>Crenicichla saxatilis</i> (Linnaeus, 1758)
Cichliformes	<i>Geophagus camopiensis</i> (Pellegrin, 1903)
Cichliformes	<i>Guianacara geayi</i> (Pellegrin, 1902)
Cichliformes	<i>Guianacara owroewefi</i> (Kullander & Nijssen, 1989)
Cichliformes	<i>Krobia aff. guianensis sp1</i>
Cichliformes	<i>Krobia aff. guianensis sp2</i>
Cichliformes	<i>Krobia itanyi</i> (Puyo, 1943)
Cichliformes	<i>Nannacara aureocephalus</i> (Allgayer, 1983)
Cichliformes	<i>Satanoperca rhynchitis</i> (Kullander, 2012)
Cyprinodontiformes	<i>Anablepsoides holmiae</i> (Eigenmann, 1909)
Cyprinodontiformes	<i>Anablepsoides igneus</i> (Huber, 1979)
Cyprinodontiformes	<i>Anablepsoides lungi</i> (Berkenkamp, 1984)
Cyprinodontiformes	<i>Laimosemion agilae</i> (Hoedeman, 1954)
Cyprinodontiformes	<i>Laimosemion cladophorus</i> (Huber, 1991)
Cyprinodontiformes	<i>Laimosemion geayi</i> (Vaillant, 1899)
Cyprinodontiformes	<i>Laimosemion xiphidius</i> (Huber, 1979)
Cyprinodontiformes	<i>Micropoecilia bifurca</i> (Eigenmann, 1909)
Gobiiformes	<i>Eleotris pisonis</i> (Gmelin, 1789)
Gymnotiformes	<i>Apteronotus aff. albifrons</i>
Gymnotiformes	<i>Brachyhypopomus beebei</i> (Schultz, 1944)
Gymnotiformes	<i>Eigenmannia virescens</i> (Valenciennes, 1836)
Gymnotiformes	<i>Electrophorus electricus</i> (Linnaeus, 1766)
Gymnotiformes	<i>Gymnotus carapo</i> (Linnaeus, 1758)
Gymnotiformes	<i>Gymnotus coropinae</i> (Hoedeman, 1962)
Gymnotiformes	<i>Hypopomus artedi</i> (Kaup, 1856)
Gymnotiformes	<i>Hypopygus lepturus</i> (Hoedeman, 1962)
Gymnotiformes	<i>Japigny kirschbaum</i> (Meunier, Jgu & Keith, 2011)
Gymnotiformes	<i>Sternopygus macrurus</i> (Bloch & Schneider, 1801)
Incertae sedis in Ovalentaria	<i>Polycentrus schomburgkii</i> (Mller & Troschel, 1849)
Myliobatiformes	<i>Potamotrygon hystrix</i> (Mller & Henle, 1841)
Siluriformes	<i>Ancistrus aff. temminckii</i> (Valenciennes, 1840)
Siluriformes	<i>Ancistrus cf. leucostictus</i> (Gnther, 1864)
Siluriformes	<i>Batrochoglanis raninus</i> (Valenciennes, 1840)
Siluriformes	<i>Callichthys callichthys</i> (Linnaeus, 1758)
Siluriformes	<i>Cetopsidium orientale</i> (Vari, Ferraris & Keith, 2003)
Siluriformes	<i>Chasmocranus brevior</i> (Eigenmann, 1912)

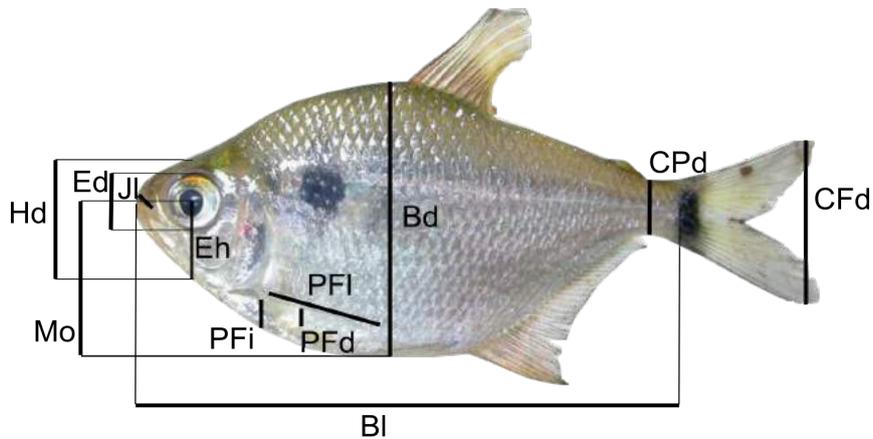
Table III.S1 continued

Siluriformes	<i>Chasmocranus longior</i> (Eigenmann, 1912)
Siluriformes	<i>Corydoras aeneus</i> (Gill, 1858)
Siluriformes	<i>Corydoras amapaensis</i> (Nijssen, 1972)
Siluriformes	<i>Corydoras geoffroy</i> (Lacepde, 1803)
Siluriformes	<i>Corydoras guianensis</i> (Nijssen, 1970)
Siluriformes	<i>Corydoras solox</i> (Nijssen & Isbrcker, 1983)
Siluriformes	<i>Corydoras spilurus</i> (Norman, 1926)
Siluriformes	<i>Cteniloricaria platystoma</i> (Gnther, 1868)
Siluriformes	<i>Farlowella reticulata</i> (Boeseman, 1971)
Siluriformes	<i>Farlowella rugosa</i> (Boeseman, 1971)
Siluriformes	<i>Glanidium leopardum</i> (Hoedeman, 1961)
Siluriformes	<i>Harttia fowleri</i> (Pellegrin, 1908)
Siluriformes	<i>Harttia guianensis</i> (Rapp Py-Daniel & Oliveira, 2001)
Siluriformes	<i>Harttiella longicauda</i> (Covain & Fish-Muller, 2012)
Siluriformes	<i>Helogenes marmoratus</i> (Gnther, 1863)
Siluriformes	<i>Heptapterus bleekeri</i> (Boeseman, 1953)
Siluriformes	<i>Heptapterus tenuis</i> (Mees, 1986)
Siluriformes	<i>Hypostomus gymnorhynchus</i> (Norman, 1926)
Siluriformes	<i>Imparfinis pijpersi</i> (Hoedeman, 1961)
Siluriformes	<i>Ituglanis amazonicus</i> (Steindachner, 1882)
Siluriformes	<i>Ituglanis nebulosus</i> (de Pinna & Keith, 2003)
Siluriformes	<i>Lithoxus boujardi</i> (Muller & Isbrcker, 1990)
Siluriformes	<i>Lithoxus planquettei</i> (Boeseman, 1982)
Siluriformes	<i>Lithoxus stocki</i> (Nijssen & Isbrcker, 1990)
Siluriformes	<i>Loricaria aff. parnahybae</i>
Siluriformes	<i>Loricaria cataphracta</i> (Linnaeus, 1758)
Siluriformes	<i>Megalechis thoracata</i> (Valenciennes, 1840)
Siluriformes	<i>Ochmacanthus reinhardtii</i> (Steindachner, 1882)
Siluriformes	<i>Pimelodella cristata</i> (Mller & Troschel, 1849)
Siluriformes	<i>Pimelodella geryi</i> (Hoedeman, 1961)
Siluriformes	<i>Pimelodella macturki</i> (Eigenmann, 1912)
Siluriformes	<i>Pimelodella procera</i> (Mees, 1983)
Siluriformes	<i>Pseudancistrus brevispinis</i> (Heitmans, Nijssen & Isbrcker, 1983)
Siluriformes	<i>Rhamdia quelen</i> (Quoy & Gaimard, 1824)
Siluriformes	<i>Rineloricaria platyura</i> (Mller & Troschel, 1849)
Siluriformes	<i>Tatia brunnea</i> (Mees, 1974)
Siluriformes	<i>Tatia intermedia</i> (Steindachner, 1877)
Siluriformes	<i>Trachelyopterus galeatus</i> (Linnaeus, 1766)
Synbranchiformes	<i>Synbranchus marmoratus</i> (Bloch, 1795)

**Table III.S2:** Physical characteristics and pairwise geodesic distance among the selected sites for French Guiana and continental France (mean  $\pm$  SD)

Characteristics	French Guiana	Continental France
Distance from the source (km)	4.02 $\pm$ 4.22	7.96 $\pm$ 3.73
Altitude (m)	176.53 $\pm$ 171.01	234.64 $\pm$ 27.81
Slope (‰)	5.05 $\pm$ 2.90	4.29 $\pm$ 3.08
Pairwise distance between sites (km)	117.90 $\pm$ 73.26	98.97 $\pm$ 57.22

**Figure III.S1:** Morphological measurements measured on each fish species



**Table III.S3:** Morphological measurements (a) and functional traits (b) measured on each fish species

## (a) Morphological measurements

<b>Symbole</b>	<b>Name</b>	<b>Definition</b>
Bl	Body length	Standard Length
Hd	Head depth	Head depth along the vertical axis of the eye
CPd	Caudal peduncle depth	Minimal caudal peduncle depth
CFd	Caudal fin depth	Maximum depth of caudal fin
Ed	Eye diameter	Vertical diameter of the eye
Jl	Maxillary Jaw Length	Length from snout to the corner of the mouth
Bbl	Barbel maximum Length	Length of the longest barbel
PFl	Pectoral fin length	Length of the longest ray of the pectoral fin
PFi	Pectoral fin position	Vertical distance between the upper insertion of the pectoral fin to the bottom of the body
Bd	Body depth	Maximum body depth
Eh	Eye position	Vertical distance between the centre of the eye to the bottom of the body
Mo	Oral gape position	Vertical distance from the top of the mouth to the bottom of the body
CFs	Caudal fin surface	Total fin surface
PFs	Pectoral fin surface	Total fin surface
MaxLength	Maximum length	Maximum adult length (obtained from FishBase (Froese & Pauly, 2015))

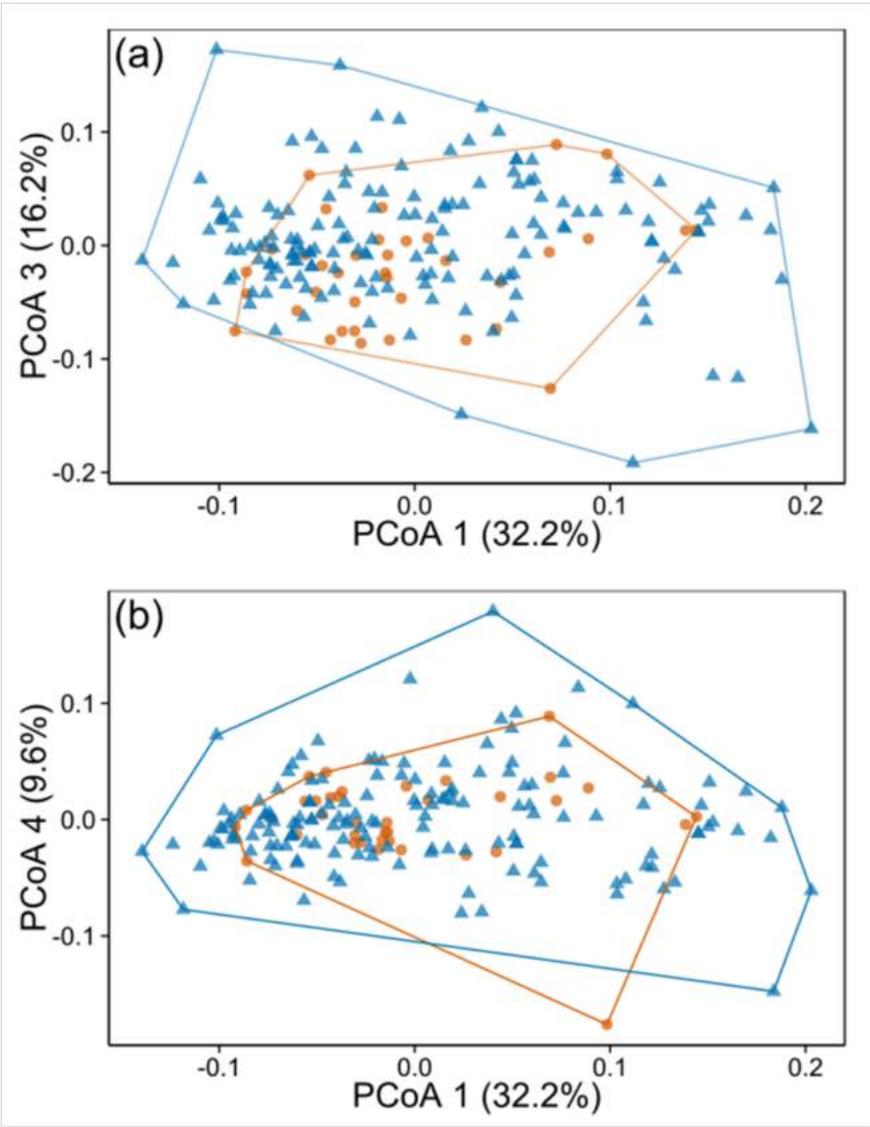
Table III.S3 continued

(b) Functional traits

Component	Functional traits	Measure	Relevance
	Maximum length	$\text{Log}(\text{MaxLength})$	Metabolism, trophic impacts, locomotion ability, nutrient cycling
Prey detection	Eye size	$\frac{Ed}{Hd}$	Visual acuity (Boyle & Horn, 2006)
	Barbel length	$\frac{Bl}{Bd}$	Detection of hidden preys
Prey capture	Oral gape position	$\frac{Mo}{Hd}$	Feeding position in the water column (Dumay <i>et al.</i> , 2004; Lefcheck <i>et al.</i> , 2014)
Position in the water column	Maxillary length	$\frac{Jl}{Hd}$	Size and strength of jaw
	Eye position	$\frac{Eh}{Hd}$	Position of fish and/or of its prey in the water column (Winemiller, 1991)
	Body elongation	$\frac{Bl}{Bd}$	Position of fish and/or of its prey in the water column (Winemiller, 1991)
Swimming	Body lateral shape	$\frac{Hd}{Bl}$	Relative depth of the head compared to the body
	Pectoral fin position	$\frac{Pfi}{Bd}$	Pectoral fin use for maneuverability (Dumay <i>et al.</i> , 2004)
	Pectoral fin shape	$\frac{Pfl^2}{Pfl^s}$	Pectoral fin use for propulsion (Fulton <i>et al.</i> , 2001)
	Pectoral fin size	$\frac{Pfl^s}{Bl}$	Pectoral fin use for propulsion (Fulton <i>et al.</i> , 2001)
	Caudal peduncle throttling	$\frac{CFd}{CPd}$	Caudal propulsion efficiency through reduction of drag (Webb, 1984)
	Caudal fin aspect ratio	$\frac{CFd^2}{CFs}$	Caudal fin use for propulsion and/or direction (Gatz, 1968; Webb, 1984)
	Fin surface ratio	$\frac{PFs}{CFs}$	Fin use for swimming
	Relative fin surface	$\frac{(PFs+CFs)}{(Bl \times Bd)}$	Fin total surface compared to body lateral surface

**Table III.S4:** Spearman's correlations between the functional traits and the four PCoA components

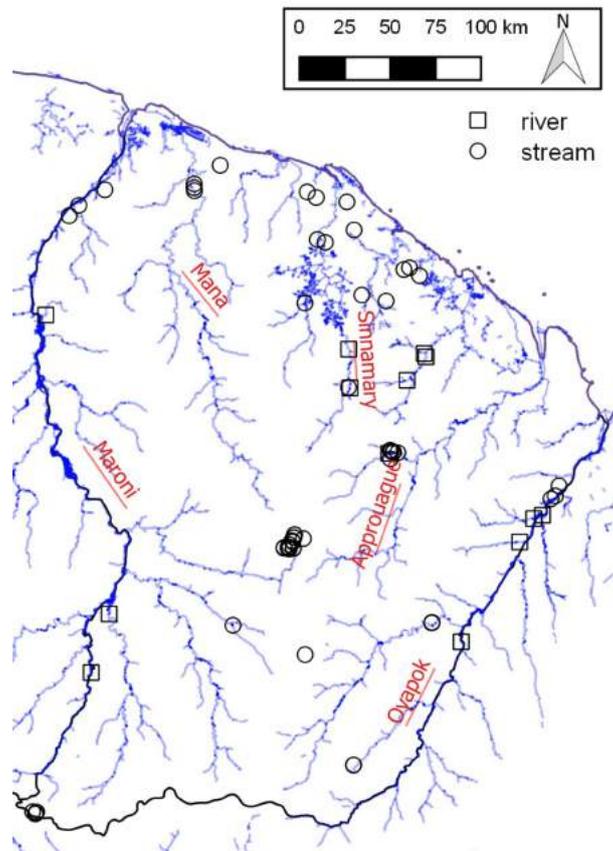
<b>Functional traits</b>	<b>Axis 1</b>	<b>Axis 2</b>	<b>Axis 3</b>	<b>Axis 4</b>
$\text{Log}(MaxLength)$	0.23	-0.49	-0.45	-0.066
$\frac{Ed}{Hd}$	-0.59	0.38	0.090	0.26
$\frac{Bbl}{Bl}$	0.49	0.057	-0.011	0.20
$\frac{Mo}{Hd}$	-0.70	-0.17	-0.031	-0.11
$\frac{Jl}{Hd}$	-0.15	-0.43	0.00	0.10
$\frac{Eh}{Hd}$	0.52	-0.14	-0.26	0.22
$\frac{Bl}{Bd}$	0.40	-0.21	-0.10	0.29
$\frac{Hd}{Bd}$	0.55	0.20	0.43	-0.05
$\frac{PFl}{Bd}$	0.072	-0.79	0.18	0.33
$\frac{PFl^2}{PFs}$	-0.34	0.60	-0.28	-0.31
$\frac{PFl}{Bl}$	0.059	0.35	0.34	-0.042
$\frac{CFd}{CPd}$	-0.41	0.59	-0.27	-0.21
$\frac{CFd^2}{CFs}$	-0.60	0.44	-0.50	-0.31
$\frac{PFs}{CFs}$	0.65	-0.038	0.36	0.27
$\frac{(PFs+CFs)}{(Bl \times Bd)}$	0.33	0.027	0.63	0.50



**Figure III.S2:** Ordination of fish species along the first and the third principal components (representing 48.4% of the total inertia) (a) and along the first and the fourth principal components (representing 41.8% of the total inertia) (b) based on the functional distance matrix computed with Gowers distance on the 15 morpho-anatomic measures. Convex hulls for continental France (●) and French Guiana (▲) are represented

## III.2 Inclusion of phylogenetic diversity for Guianese assemblages

After showing that the processes structuring fish assemblages differed between different regions, I focused on the Guianese freshwater fish assemblages and I included the phylogenetic diversity facet to the analyses to go further into the processes that structure these assemblages, based on the framework developed in the Introduction (Figure I.6). These analyses are based on the fish inventories on 75 stream sites and 13 river sites in non-impacted or lowly disturbed areas (Figure III.1). The first step was thus to build the phylogenetic relationships between Guianese species.



**Figure III.1:** Map of French Guiana showing the location of stream (open circle) and river (open square) study sites

### Molecular material

We sampled tissues from 703 specimens belonging to 230 species (1 to 3 specimens per species), and stored them in 96% ethanol. We extracted DNA from 0.05–0.25 cm<sup>2</sup> of tissue using salt-extraction protocol (Aljanabi & Martinez, 1997). We performed DNA amplification in a final volume of 25  $\mu$ L including 1U of GoTaq<sup>®</sup> (Promega), 5X buffer (Promega), 10  $\mu$ M of dNTP, 20  $\mu$ M of each primer (12S: teleo\_R 5'-CTCCGGTACTTACCATG-3' and V05F.898 5'-AACTCGTGCCAGCCACC-3', Valentini *et al.*, 2016; COI: FishF1 5'-TCAACCAACCACAAAGACATTGGCAC-3' and FishR1 5'-TAGACTTCTGGGTGGCCAAAGAATCA-3', Ward *et al.*, 2005) and 1  $\mu$ L of

DNA template. For 12S, the PCR mixture was denatured at 95°C for 10 min, followed by 35 cycles of 30 s at 95°C, 30 s at 55°C and 1 min at 72°C, and followed by a final elongation at 72°C for 7 min, in a room dedicated to amplified DNA, with negative air pressure and physically separated from the DNA extraction rooms. For COI, the PCR mixture was denatured at 94°C for 2 min, followed by 35 cycles of 30 s at 94°C, 40 s at 52°C and 1 min at 72°C, and followed by a final elongation at 72°C for 10 min. A 600 bp product of the 12S rRNA gene and a 655 bp product of the COI gene was obtained and sequenced using Sanger sequencing method by Genoscreen biotechnology company. Sequences were analysed using GENEIOUS version 9.0<sup>2</sup>, Kearse *et al.*, 2012).

We also added to our data the sequences available on GenBank, corresponding to 472 sequences (128 for the 12S and 344 for the COI) for 142 species. In total, we had 811 sequences for the COI and 621 sequences for the 12S. For each species, individuals with the two markers were kept. The remaining individuals with a single marker were combined to produce pseudo-individuals combining the two markers. This procedure permitted to consider 460 individuals (290 real individuals and 170 pseudo-individuals). We also kept species for which no real or pseudo-individuals were individuals (only one marker was available for them, representing 160 individuals with one of the two markers). We used three outgroup species to root our tree: the pouched lamprey (*Geotria australis*), the smalltooth sawfish (*Pristis pectinata*) and the bull shark (*Carcharhinus leucas*) that all occur in nearby tropical marine waters (Froese & Pauly, 2015) (Table III.1).

### Phylogenetic inferences

COI sequences were then aligned using TranslatorX (Abascal *et al.*, 2010), with the multiple alignment build with MAFFT version 7.222 (Kato & Standley, 2013) and automatic detection of reading frame with vertebrate mitochondrial genetic code. 12S rRNA sequences were aligned with MAFFT v7.22 using structural alignment methods (Q-INS-i iterative refinement method), with sequences firstly aligned by Orders (outgroup species sequences were also aligned together). The Order multiple sequence alignments were then merge into one alignment using the “merge” option of MAFFT. The 12S and COI alignments were trimmed using Gblocks version 0.91b (Castresana, 2000), with half of allowed gap positions and default values for other parameters. The two alignments were concatenated, and partition scheme (with first, second and third codon for COI and 12S) and corresponding substitution models were obtained using PartitionFinder version 2.1.1 (Lanfear *et al.*, 2016). The best partition scheme was to keep all markers as one, with the GTR+I+G model of substitution.

Phylogenetic inferences were conducted using maximum likelihood with RAxML version 8.2.4 (Stamatakis, 2014) with the “autoMRE” option for automatic determination of bootstrap replicates. The analysis was constrained using a taxonomic tree determined at the Order level. From the resulting tree, all individuals

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<sup>2</sup><https://www.geneious.com/>

**Table III.1:** Accession number of the supplementary sequences for the COI and 12S markers retrieved from GenBank used in the analyses.

<b>Genus species</b>	<b>GenBank accession number</b>	
	COI	12S
<i>Acanthodoras cataphractus</i>	EU490847	AY264120
<i>Acaronia nassa</i>	AY263862	
<i>Ageneiosus ucayalensis</i>	EU490849	AY264122
<i>Anchovia clupeioides</i>		EU552651
<i>Anchoviella lepidentostole</i>	JQ365218	EU552653
<i>Anostomus ternetzi</i>		KF415311
<i>Apteronotus albifrons</i>	JQ667494	AB054132
<i>Astronotus ocellatus</i>	AY263859	AP009127
<i>Astyanax bimaculatus</i>	FJ439410	
<i>Bagre bagre</i>	GU702396	DQ990572
<i>Batrochoglanis raninus</i>	EU179809	EU179829
	JX899730	
<i>Brachyhypopomus brevirostris</i>	KF533324	
	KF533325	
<i>Brachyhypopomus pinnicaudatus</i>	GU701847	
	GU701848	
	GU701851	
<i>Brachyplatystoma filamentosum</i>	FJ418760	JF898675
	FJ978041	JF898676
<i>Brachyplatystoma rousseauxii</i>	FJ418759	JF898672
<i>Brachyplatystoma vaillantii</i>	HM453213	JF898666
<i>Bunocephalus coracoideus</i>	DQ508071	AP012006
<i>Bunocephalus verrucosus</i>	EU490846	
<i>Callichthys callichthys</i>	EU359408	HM114376
	GU702211	HM114377
	JN988766	HM114378
	JN988767	HM114379
<i>Caranx hippos</i>	FJ347905	
	FJ347906	
	JQ841097	
	JQ842407	
	KF461147	
<i>Caranx latus</i>	JQ365253	
	JQ365254	
	JQ635255	
	JQ840438	
	JQ841100	

Table III.1 continued

<i>Cathorops spixii</i>	JQ365264	
	JQ365265	
	JQ365266	
	JQ365267	
<i>Centropomus parallelus</i>	JQ365272	
	JQ365273	
	JQ365274	
	JQ365275	
	JX033999	
<i>Centropomus undecimalis</i>	EU752068	
	JQ841102	
	JQ841103	
	JQ842409	
<i>Chaetodipterus faber</i>	FJ583016	EF616894
<i>Chalceus macrolepidotus</i>	JF800931	AB054130
<i>Cichla monoculus</i>	JN988798	
	JN988799	
<i>Colomesus psittacus</i>	AP011910	AP011910
<i>Crenicichla alta</i>	AY263860	
<i>Cynoscion acoupa</i>	JQ365312	
<i>Doras carinatus</i>		AY264089
<i>Doras micropoeus</i>		AY264090
<i>Dormitator maculatus</i>	JQ935861	KF415347
<i>Epinephelus itajara</i>	JF42152	
	JN021300	
	JQ841167	
	JQ841887	
	JN026710	
<i>Geophagus surinamensis</i>		
<i>Guyanancistrus longispinis</i>		JN855720
<i>Harttia fowleri</i>	JF292255	
<i>Harttia guianensis</i>	JF292265	EU310447
<i>Hartiella intermedia</i>	JF292281	
	JF292284	
	JF292285	
	JF292286	
	JF292287	
<i>Hartiella lucifer</i>	JF292288	
	JF292289	
	JF292290	

Table III.1 continued

<i>Hartiella parva</i>	JF292274	
	JF292275	
	JF292276	
<i>Hartiella pilosa</i>	JF292271	
	JF292272	
	JF292273	
<i>Helogenes marmoratus</i>		AP012014
<i>Hemiancistrus medians</i>	JF746999	JN855719
<i>Hemiodontichthys acipenserinus</i>		EU310448
<i>Hemisorubim platyrhynchos</i>	GU570708	JF898664
	GU570709	
<i>Heros efasciatus</i>	DQ119218	GQ258749
<i>Hoplias malabaricus</i>	GU702203	AP011992
<i>Hyphessobrycon eques</i>	FJ749057	FJ748942
<i>Hypopygus lepturus</i>	KF533334	
<i>Hypostomus gymnorhynchus</i>		JN855752
<i>Hypostomus plecostomus</i>	JN026849	
	JN026850	
	JN026851	
	KF410695	
	KJ720692	
<i>Hypselecara temporalis</i>	DQ119219	AP009506
<i>Kryptolebias marmoratus</i>	FN544255	AY946280
	JQ840547	AF364957
<i>Kryptolebias sepia</i>		AY946277
<i>Laimosemion agilae</i>		AF092310
<i>Laimosemion geayi</i>		AY578720
		AY578721
		AY57876
		AY946279
<i>Lepidosiren paradoxa</i>		AF302934
		Z48715
<i>Lithodoras dorsalis</i>		AY264076
<i>Lutjanus jocu</i>	KF633372	
	KF633373	
	KF633374	
	KF633375	
	KF633376	
<i>Lycengraulis grossidens</i>	AP011563	AP011563

Table III.1 continued

<i>Macrodon ancylodon</i>	JQ365417	
	JQ365418	
	JQ365419	
	JX124804	
	JX124805	
<i>Megalechis thoracata</i>	GU701890	HM114370
	GU701965	HM114371 HM114372
<i>Megalops atlanticus</i>	AP004808	AP004808
<i>Metaloricaria paucidens</i>		EU310455
<i>Moenkhausia oligolepis</i>	EU177017	
<i>Mugil cephalus</i>	AP002930	AP002930
	AP002931	AP002931
	HQ149715	KF374974
	JQ060532	KF374976
<i>Mugil incilis</i>	JX185183	KF374978
<i>Mugil liza</i>	JX185142	KF374978
	JX185143	KF374979
<i>Nebris microps</i>	JQ365453	
	JQ365454	
	JQ365455	
	JQ365456	
	JQ365457	
<i>Notarius grandicassis</i>	JX124821	DQ990540
<i>Odontognathus mucronatus</i>	JX124826	
	JX124827	
	JX124828	
	JX124829	
	JX124830	
<i>Oligoplites saliens</i>	JQ365467	
	JQ365468	
	JQ365469	
	JQ365470	
	JQ365471	
<i>Osteoglossum bicirrhosum</i>	AB043025	AB043025
	FJ418754	AF417343
<i>Panaqolus koko</i>	JF747003	
<i>Pellona flavipinnis</i>	AP009619	AP009619
<i>Pimelabditus moli</i>	GU181205	JF489897
	GU181206	JF489898
	GU181207	JF489899

Table III.1 continued

<i>Pimelodus blochii</i>	FJ978039	JF898713 JF898714 JF898715 JF898716
<i>Plagioscion auratus</i>	FJ418762	
<i>Plagioscion squamosissimus</i>	GU701562	FJ549363
<i>Platystacus cotylephorus</i>	DQ508086	
<i>Poecilia reticulata</i>	GU702152	AB898687
	JN028265	KJ013505
	JN028266	KJ460033
<i>Poecilia vivipara</i>	GU701904	
	GU701907	
	GU701908	
	GU701911	
<i>Potamotrygon orbignyi</i>	JN184071	AF448006
<i>Pseudoplatystoma fasciatum</i>	GU570710	FJ549367
<i>Pseudoplatystoma tigrinum</i>	GU570877	JF898658
<i>Pygocentrus nattereri</i>		AP012000
<i>Rhamdia quelen</i>	EU179804	EU179824
<i>Sciades herzbergii</i>		DQ990568
<i>Sciades parkeri</i>		DQ990567
<i>Sciades passany</i>		DQ990570
<i>Sciades proops</i>		DQ990566
<i>Sphoeroides testudineus</i>	AP011916	AP011916
<i>Sternopygus macrurus</i>	GU701495	
<i>Syacium gunteri</i>	JX887477	
<i>Symphurus plagusia</i>		AF488497
<i>Synbranchus marmoratus</i>	AP004439	AP004439 GU701491
<i>Tetragonopterus chalceus</i>	FJ749080	FJ748963
<i>Tomeurus gracilis</i>		EF017507
<i>Trachelyopterus galeatus</i>	GU701484	JX899742
	GU701490	
<i>Trachinotus falcatus</i>	JQ365600	
	JQ365601	
	JQ365602	
	JQ365602	
	JQ841416	
<i>Carcharhinus leucas</i> (outgroup)	KF646785	KF646786
<i>Geotria australis</i> (outgroup)	KT185629	KT185629
	NC_029404	NC_029404

Table III.1 continued

<i>Pristis pectinata</i> (outgroup)	KP400584	KP400584
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from the same species were collapsed to form only one tip. For some species, individuals did not form monophyletic groups and were closely related to individuals from other species. For these, the most recent common ancestor to all the individuals was used to transform all descending branches into polytomy and only one tip per species was kept. The polytomies were then randomly converted into dichotomies with branch length of 0 for further analyses. The resulting tree was composed of 303 species and was then dated using a semi-parametric method based on penalized likelihood (Sanderson, 2002). The resulting phylogenetic tree is presented in Figure III.2.

We then matched assemblage data, functional traits data and the phylogenetic tree to only keep the species common to the three datasets for subsequent analyses (195 species in total). We did this separately for streams and rivers, as our goal was not to compare streams and rivers and the difference in sampling protocol can affect the results.

### Functional space

We computed functional distance matrix between species with Gowers distance based on the morphological ratios and the fish size as previously. We used the mean ratio values for species with replicates, but we did not take into account intraspecific variability because 40% of the species did not have replicate data. We used a PCoA on the functional distance matrix to obtain the coordinates of the species in the functional space (Villéger *et al.*, 2008; Leitão *et al.*, 2016). We kept the first four principal components to describe the functional space according to Maire *et al.* (2015), representing 80.6% of the inertia (33.5%, 22.6%, 14.5% and 10.0% for the first four axes of the PCoA respectively).

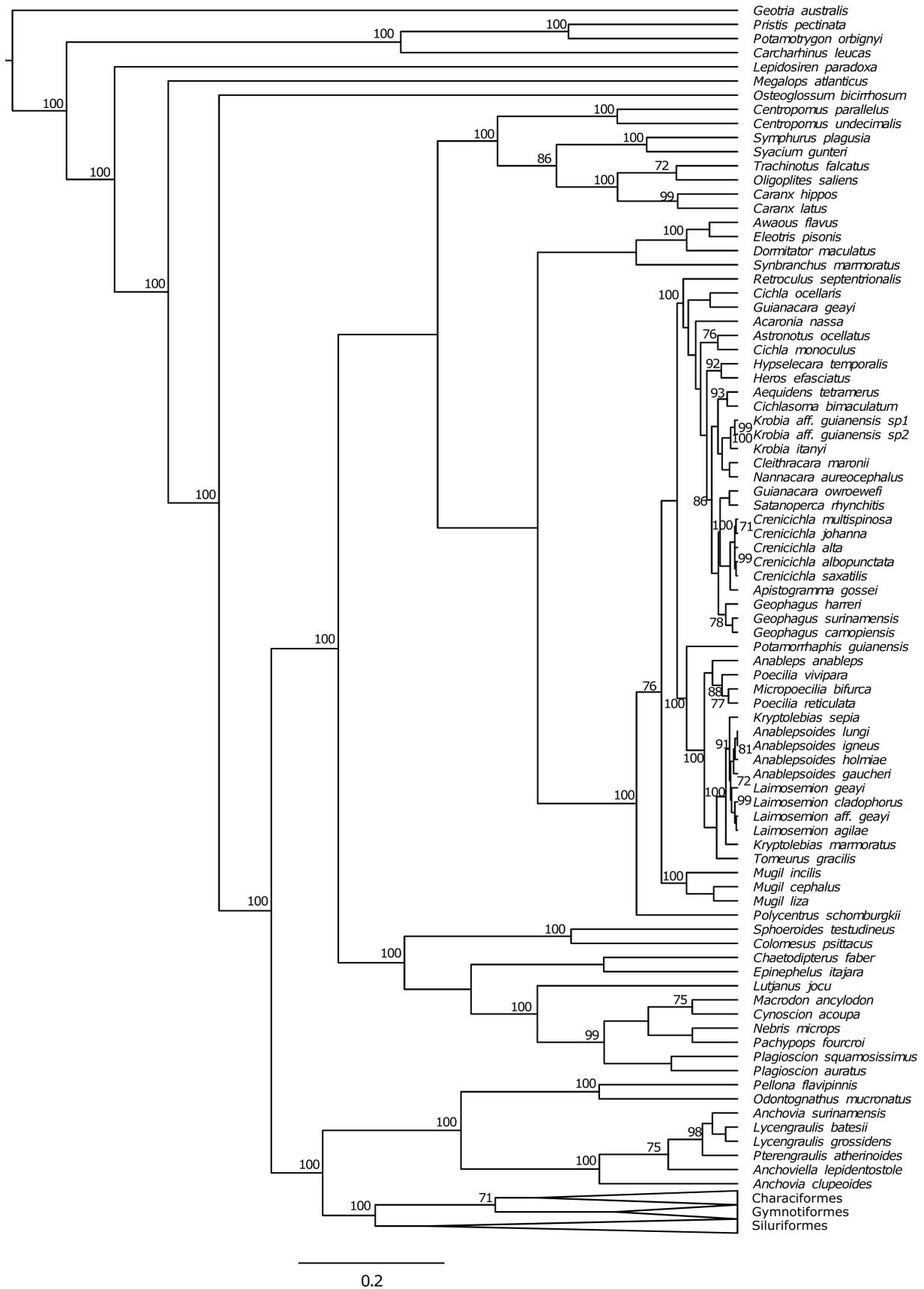
### Phylogenetic signal in functional traits

We measured the degree of phylogenetic signal at two levels. First we compared overall functional distance between species (obtained in the *Functional traits measurement* section) to the phylogenetic distance between species using Mantel test (Legendre & Legendre, 2012). We then test the degree of phylogenetic signal in each trait with the K statistic and (Blomberg *et al.*, 2003). K values of 1 indicate that the trait distribution on the phylogeny perfectly matches a Brownian motion expectation of trait evolution on the phylogeny. K values less than 1 indicate greater convergence in values than expected under a Brownian trait evolution model, and K values greater than 1 indicates a higher degree of phylogenetic signal than Brownian motion. K values significances were assessed by comparing their values to the distribution of values obtained by shuffling the species across the tips of the phylogeny 1,000 times.

**Table III.2:** PartitionFinder results to select the different scheme tested and their corresponding log likelihood (LnL), the number of estimated parameters and its AICc

Partitioning scheme	Model	lnL	Parameters	AICc
COI_1+COI_2+COI_3+12S	GTR+I+G	-79435.66	1227	3174837.31
COI_1+COI_2+COI_3, 12S	GTR+I+G, GTR+I+G	-79260.04	1238	3228760.07
COI_1+COI_2+12S, COI_3	GTR+I+G, GTR+G	-78129.58	1237	3221545.15
COI_1+COI_2, COI_3+12S	GTR+I+G, GTR+I+G	-78969.21	1238	3228178.42
COI_1+COI_2, COI_3, 12S	GTR+I+G, GTR+G, GTR+I+G	-77973	1248	3275946
COI_1+COI_3+12S, COI_2	GTR+I+G, GTR+I+G	-79186.71	1238	3228613.42
COI_1+COI_3, COI_2+12S	GTR+I+G, GTR+I+G	-79102.29	1238	3228444.58
COI_1+COI_3, COI_2, 12S	GTR+I+G, GTR+I+G, GTR+I+G	-78918.28	1249	3282834.56
COI_1+12S, COI_2+COI_3	GTR+I+G, GTR+I+G	-78934.8	1238	3228109.61
COI_1, COI_2, COI_3, 12S	GTR+I+G, GTR+I+G	-79161.69	1238	3228563.37
COI_1, COI_2, COI_3 + 12S	GTR+I+G, GTR+I+G, GTR+I+G	-78799.59	1249	3282597.18
COI_1+12S, COI_2, COI_3	GTR+I+G, GTR+I+G, GTR+G	-77968.39	1248	3275936.79
COI_1, COI_2, 12S + COI_3	GTR+I+G, GTR+I+G, GTR+G	-78017.19	1248	3276034.38
COI_1, COI_2, COI_3+12S	GTR+I+G, GTR+I+G, GTR+I+G	-78829.4	1249	3282656.79
COI_1, COI_2, COI_3, 12S	GTR+I+G, GTR+I+G, GTR+G + GTR+I+G	-77833.18	1259	3330864.37

Overall, functional distances between species were correlated with phylogenetic distance (Mantel  $r = .49$ ,  $p = .001$ ). Looking trait by trait, K values were all less than 1, meaning that the traits are less conserved than expected under a Brownian motion model. However, all K values are more conserved than predicted by random association between phylogeny and traits. These results thus indicated a moderate trait conservatism.



**Figure III.2:** Maximum likelihood phylogenetic tree of Guianese freshwater fish species based on 12S and COI. Only bootstrap values greater than 70 are indicated

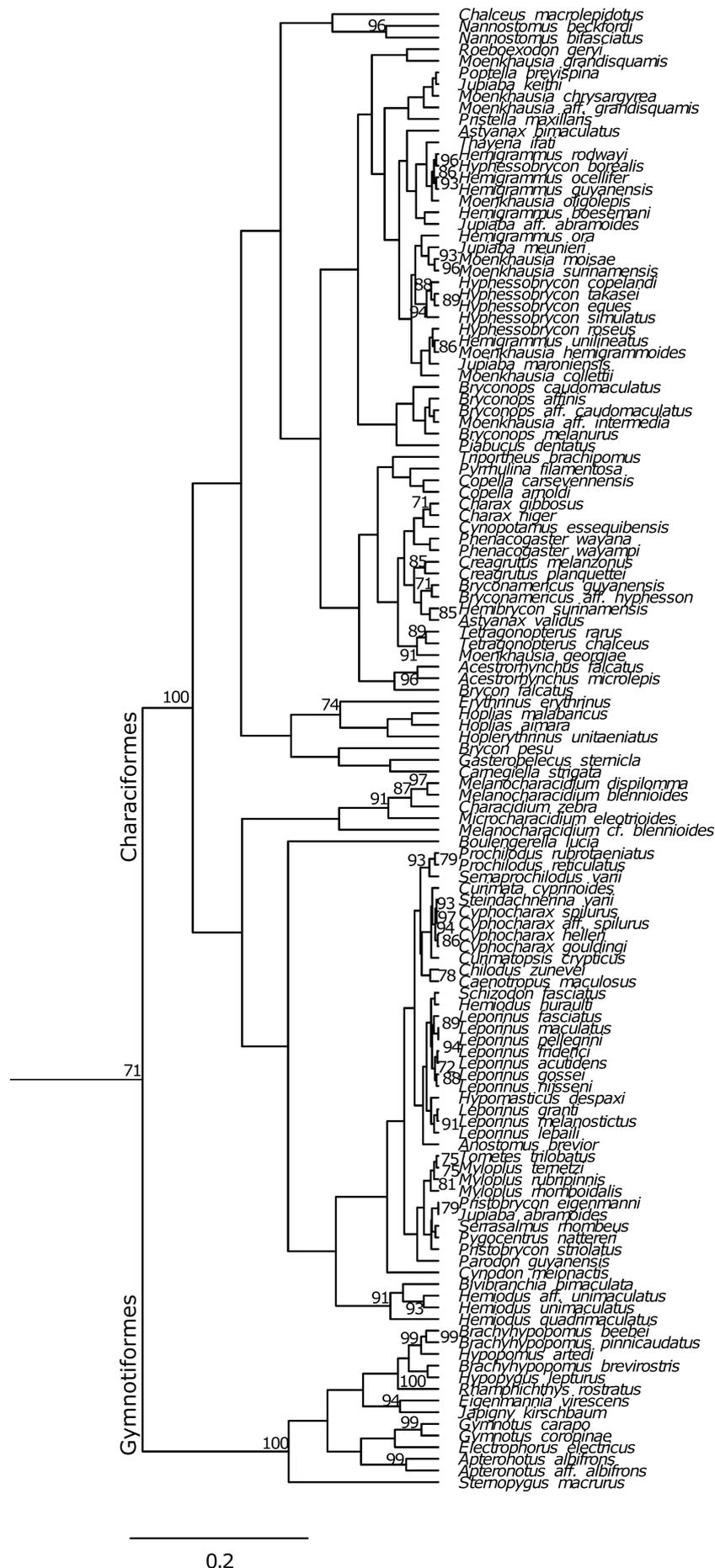


Figure III.2 continued: Characiformes and Gymnotiformes clades

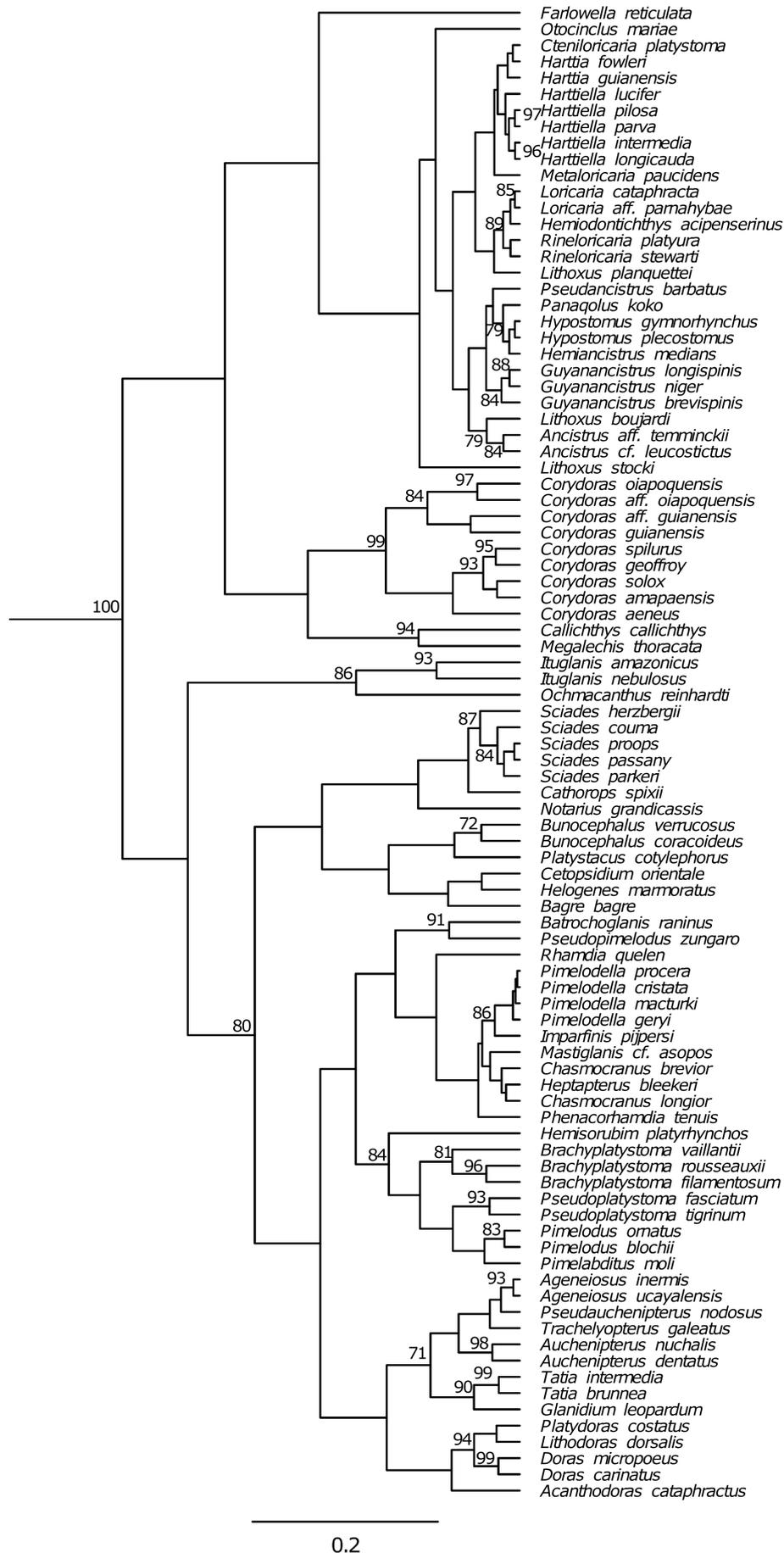


Figure III.2 continued: Siluriformes clade

**Within-assemblage scale:  $\alpha$ -diversity measures**

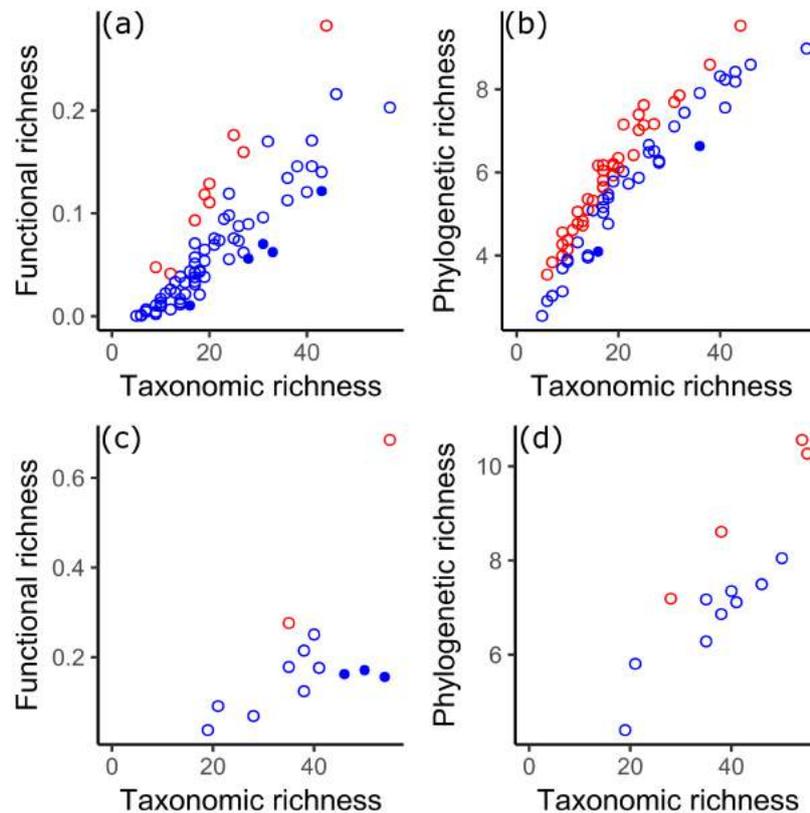
At the within-assemblage scale, we used species richness as a measure of taxonomic  $\alpha$ -diversity. For functional diversity, we calculated functional richness as the relative volume occupied in the functional space by the species present in one local assemblage (Cornwell *et al.*, 2006; Villéger *et al.*, 2008). We used Faith's PD as a measure of phylogenetic richness (Faith, 1992). These two metrics are correlated with species richness, but as our goal was not to compare them between sites, we did not use other metrics that are less correlated with species richness [e.g. dispersion measure (Webb, 2000; Villéger *et al.*, 2008)].

To assess if the observed functional and phylogenetic richness differed from random values given the taxonomic richness, we used two null models (one for each diversity facets) that shuffle species identity in the functional trait matrix or in the phylogenetic tree. We simulated 999 random matrices/trees and we calculated functional and phylogenetic richness. We used these null distributions to compute a standardized effect size (SES) for each assemblage as follows:  $SES = \frac{obs - \overline{null}}{sd(null)}$ . Negative SES values mean that the observed functional or phylogenetic diversity value is lower than expected with the observed taxonomic richness, whereas positive SES value designate higher observed diversity than expected. In addition, we calculated a p-value for each assemblage by ranking the observed value in the simulated values and dividing it rank by 1,000 (1 observed value + 999 simulated values).

For stream, taxonomic richness and functional richness were highly correlated (Spearman's correlation:  $\rho = 0.90$ ,  $p < .001$ ; Figure III.3a) as well as taxonomic richness and phylogenetic richness ( $\rho = 0.95$ ,  $p < .001$ ; Figure III.3b). Phylogenetic and functional richness were also correlated ( $\rho = 0.94$ ,  $p < .001$ ). However, when controlling for taxonomic richness, very few assemblages showed lower functional or phylogenetic richness than expected (Figure III.3a and b). Only one site (crique Apa) showed significant lower functional and phylogenetic richness than expected.

For river, taxonomic richness and functional richness were not significantly correlated ( $\rho = 0.50$ ,  $p = .10$ ; Figure III.3c). Taxonomic richness and phylogenetic richness were highly correlated ( $\rho = 0.82$ ,  $p < .001$ ; Figure III.3d). Phylogenetic and functional richness were not correlated ( $\rho = 0.22$ ,  $p = .47$ ). When controlling for taxonomic richness, only 3 assemblages showed lower functional richness than expected.

These results showed that functional and phylogenetic diversity of local assemblages do not differ from random expectation, suggesting that neutral processes shaped local diversity of fish assemblages in French Guiana, both in rivers and in streams. For streams, we did not observed the presence of strong environmental filtering or limiting similarity. Limiting similarity is a process that first affects species



**Figure III.3:** Taxonomic, functional and phylogenetic  $\alpha$ -diversities of assemblages located in streams (a,b) and in rivers (c,d). Red (blue) dots indicates that the observed functional or phylogenetic diversity is higher (lower) than expected under null models. Filled dots indicates that the observed diversity is significantly different from values obtained from null model

abundance, that can lead to competitive exclusion, thus affecting species occurrence. Abundances thus can better detect this type of process that affects abundances (Freilich & Connolly, 2015). Environmental filtering was not a dominant process for local assembly. Environmental filtering occurs when some environmental characteristics select species. However if local environment is heterogeneous and do not constrain species to have particular trait, local assemblages will not be different from randomly assembled assemblages (Hubbell, 2011). We already reported that local richness was mainly determined by the reach position, and thus by the size of the stream in the network and to a lesser extent by the local environment (see Chapter II). This might be reinforce the hypothesis of neutral assembly processes for stream assemblages. For river, we could have expected that environmental filtering will mainly structure assemblages as we sampled only river bank and thus the species associated to this habitat. Two explanations can be hypothesized. First, as for streams, neutral processes structured local assemblages, meaning that the environment has a low effect. Secondly, a sampling and subsequent analysis effect can explain these patterns. The species caught in the rivers are species that are only inhabiting river banks (or that go to river banks for a time during the day). This pool of river species is the one used to construct random assemblages for assessing difference between observed and simulated diversity measures. The random assemblages are thus

composed of species only catchable in river banks. Environmental filters might thus act on the river species pool and not on the species within this pool, and will not be detectable without information about the other species present in the river. This would be a major improvement to understand local assembly processes in rivers.

### **Between-assemblage scale: $\beta$ -diversity measures**

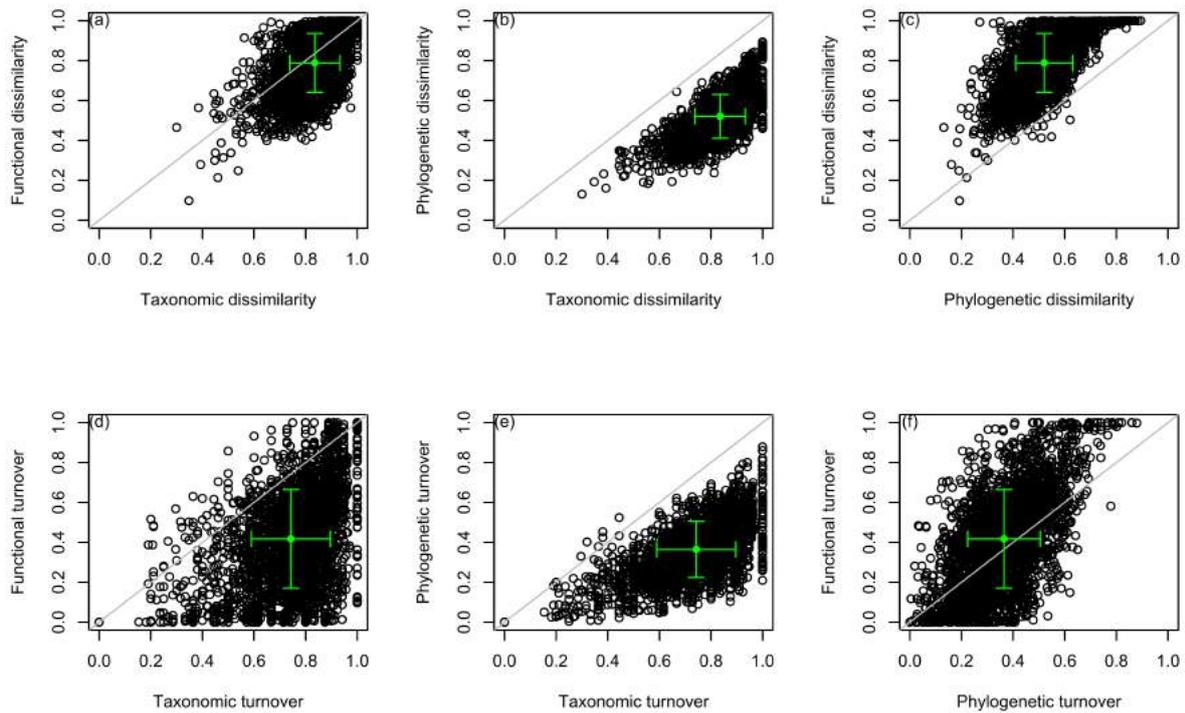
For  $\beta$ -diversity, we used Jaccard's dissimilarity index to compute pairwise taxonomic  $\beta$ -diversity (Jaccard, 1912). We also computed its turnover component (species replacement between two assemblages) according to Baselga (2012).

We used the same framework for functional and phylogenetic  $\beta$ -diversities. For functional  $\beta$ -diversity, we used convex hull volumes of assemblage and their intersection (Villéger *et al.*, 2011a). Functional dissimilarity will be equal to 1 when volumes do not overlap and 0 when volumes are identical. For phylogenetic  $\beta$ -diversity, we used the fraction of branch length shared (or not shared) between two assemblages (UniFrac index, Lozupone *et al.*, 2006). Phylogenetic dissimilarity will be equal to 1 when no branch are shared between the assemblages and 0 all branches are shared. We also extracted the turnover component of functional and phylogenetic  $\beta$ -diversities (Villéger *et al.*, 2013; Leprieur *et al.*, 2012).

We then compared the observed values of  $\beta$ -diversities to three different null models. First we compared the observed taxonomic  $\beta$ -diversity to values obtained from 999 randomly assembled assemblages by constraining both species richness within assemblages and species occurrence ('trial-swap' algorithm, Miklós & Podani, 2004). A second set of null models shuffled species identity in the trait matrix or in the phylogenetic tree without altering assemblage matrices (with 999 simulations), thus maintaining the observed taxonomic  $\beta$ -diversity. For each null models, we calculated SES and p values by assemblages as before and looked if overall, SES values departed from 0. Only results for taxonomic and phylogenetic will be presented here, as the functional null model presented computation errors that need to be examined in details.

For stream, the three  $\beta$ -diversity metrics were highly correlated (Pearson's correlation: taxonomic-phylogenetic:  $r = .78$ ,  $p < .001$ , Figure III.4a; taxonomic-functional:  $r = .68$ ,  $p < .001$ , Figure III.4b; phylogenetic-functional:  $r = .70$ ,  $p < .001$ , Figure III.4c). Taxonomic  $\beta$ -diversity was higher than phylogenetic  $\beta$ -diversity (Wilcoxon signed-rank test:  $z = 61.27$ ,  $p < .001$ ) and higher than functional  $\beta$ -diversity ( $z = 10.69$ ,  $p < .001$ ). Phylogenetic  $\beta$ -diversity was lower than functional  $\beta$ -diversity ( $z = -54.23$ ,  $p < .001$ ).

Turnover metrics were also correlated (taxonomic-phylogenetic:  $r = .64$ ,  $p < .001$ , Figure III.4d; taxonomic-functional:  $r = .45$ ,  $p < .001$ , Figure III.4e; phylogenetic-functional:  $r = .60$ ,  $p < .001$ , Figure III.4f). Taxonomic turnover was higher than phylogenetic turnover ( $z = 58.58$ ,  $p < .001$ ) and higher than functional turnover ( $z = 46.86$ ,  $p < .001$ ). Phylogenetic turnover was lower than functional



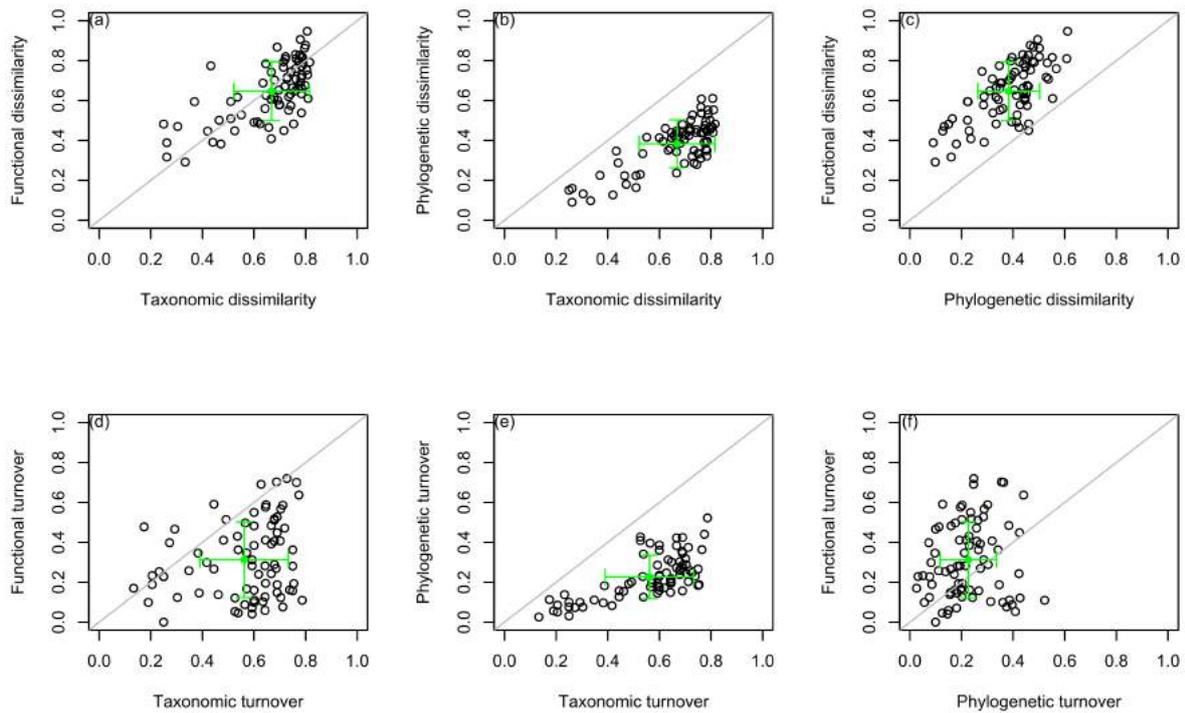
**Figure III.4:** Taxonomic, functional and phylogenetic  $\beta$ -diversities of assemblages located in streams. Green dots and bars represent medians and SD respectively. The line 1:1 is indicated

turnover ( $z = -8.38$ ,  $p < .001$ ).

Null models revealed that a low number of pairwise comparisons significantly differed from null values for taxonomic and phylogenetic diversities [taxonomic  $\beta$ -diversity: 33/2740/2 ( $p < 0.025$ /not different/ $p > 0.975$ ); taxonomic turnover: 33/2740/2; phylogenetic  $\beta$ -diversity: 36/2538/201; phylogenetic turnover: 41/2511/223], but overall SES values were different and higher than 0 (taxonomic  $\beta$ -diversity: median = 0.12,  $z = 5.14$ ,  $p < .001$ ; taxonomic turnover: median = 0.14,  $z = 6.13$ ,  $p < .001$ ; phylogenetic  $\beta$ -diversity: median = 0.29,  $z = 14.48$ ,  $p < .001$ ; phylogenetic turnover: median = 0.37,  $z = 16.94$ ,  $p < .001$ ).

For rivers, the three  $\beta$ -diversity metrics were correlated ( $r = .70$ ,  $p < .001$ , Figure III.4a; taxonomic-functional:  $r = .57$ ,  $p < .001$ , III.4b; phylogenetic-functional:  $r = .70$ ,  $p < .001$ , III.4c). Taxonomic  $\beta$ -diversity was higher than phylogenetic  $\beta$ -diversity ( $z = 8.91$ ,  $p < .001$ ) but not different from functional  $\beta$ -diversity ( $z = 1.28$ ,  $p = .20$ ). Phylogenetic  $\beta$ -diversity was lower than functional  $\beta$ -diversity ( $z = -8.94$ ,  $p < .001$ ).

Turnover metrics were also correlated (taxonomic-phylogenetic:  $r = .64$ ,  $p < .001$ , Figure III.4d; taxonomic-functional:  $r = .45$ ,  $p < .001$ , Figure III.4e; phylogenetic-functional:  $r = .60$ ,  $p < .001$ , Figure III.4f). Taxonomic turnover was higher than phylogenetic turnover ( $z = 9.13$ ,  $p < .001$ ) and higher than functional turnover ( $z = 7.19$ ,  $p < .001$ ). Phylogenetic turnover was lower than functional turnover ( $z = -2.51$ ,  $p = .01$ ).



**Figure III.5:** Taxonomic, functional and phylogenetic  $\beta$ -diversities of assemblages located in rivers. Green dots and bars represent medians and SD respectively. The line 1:1 is indicated

Null models revealed that less than half of pairwise comparisons differed from null values for taxonomic diversity (taxonomic  $\beta$ -diversity: 17/47/14; taxonomic turnover: 17/47/14) and overall, SES values did not differ from 0 (taxonomic  $\beta$ -diversity: median = 0.52,  $z = 0.71$ ,  $p = .47$ ; taxonomic turnover: median = 0.52,  $z = 0.70$ ,  $p = .49$ ). All observed phylogenetic diversity values were lower than expected. Overall SES values were different and lower than 0 only for phylogenetic turnover (phylogenetic  $\beta$ -diversity: median = 0.06,  $z = -0.06$ ,  $p = .95$ ; phylogenetic turnover: median = -0.61,  $z = -4.60$ ,  $p < .001$ ).

These results showed that dispersal limitation globally structured fish assemblages in French Guiana, based on the relationship between functional and taxonomic turnover (higher taxonomic turnover than functional turnover). This still needs to be confirmed with the appropriate null models. However, relationships between taxonomic and phylogenetic turnover highlighted that streams and rivers differed in how dispersal limitation structures fish assemblages. For streams, taxonomic and phylogenetic  $\beta$ -diversity were on average higher than expected, suggesting that stream fish assemblage have been isolated and have evolved more independently than rivers assemblages. For these rivers assemblages, the phylogenetic turnover was lower than expected, suggesting that their isolation was more recent than for stream assemblages. These differences might roots in the past history of Guianese fish assemblage and the evolution of drainage basins. The connections between drainage basins between hot periods permitted the exchange of fauna between basins

(Boujard & Tito de Morais, 1992; Dias *et al.*, 2014), which can be facilitated for species inhabiting rivers than species from the upstream part of the hydrological network (Cardoso & Montoya-Burgos, 2009), both in the travel distance and in the physiological requirement (dispersal capacity, salinity and turbidity tolerances). For stream species, reaching other stream sections from the same drainage basin or from other basins needs high dispersal capacity and the ability to tolerate different conditions.

These hypotheses of strong dispersal limitation and low environmental filtering need however to be tested using spatial and environmental information. Indeed, looking at how  $\beta$ -diversities change with distance can help to confirm the processes that structure fish assemblages. Taxonomic  $\beta$ -diversity should increase with geographical distance and environmental distance. If the main process structuring assemblages is dispersal limitation, environmental distance between these assemblages should have a lower effect than spatial distance (environmental distance will be low between the assemblages). On the opposite, strong effect of environmental distance is expected if environmental filtering occurs.

In conclusion, freshwater fish assemblages of French Guiana are mainly structured by dispersal limitation between drainage basins or between tributaries within each drainage basin. The environment seems to have a slight impact on fish assemblages structure in non impacted stream or rivers. We could expect that, as anthropogenic disturbances greatly change local environmental conditions (high turbidity and modification of stream characteristics, Dedieu *et al.*, 2014), the processes structuring both local assemblages and their structures will also change (e.g. an increase of environmental filtering). Testing this however necessitates to increase our knowledge of local fish assemblages (both in space and the species present), which can only be made currently via the development of more exhaustive sampling methods.



# Chapter IV: Toward a new sampling protocol: a molecular approach with eDNA metabarcoding

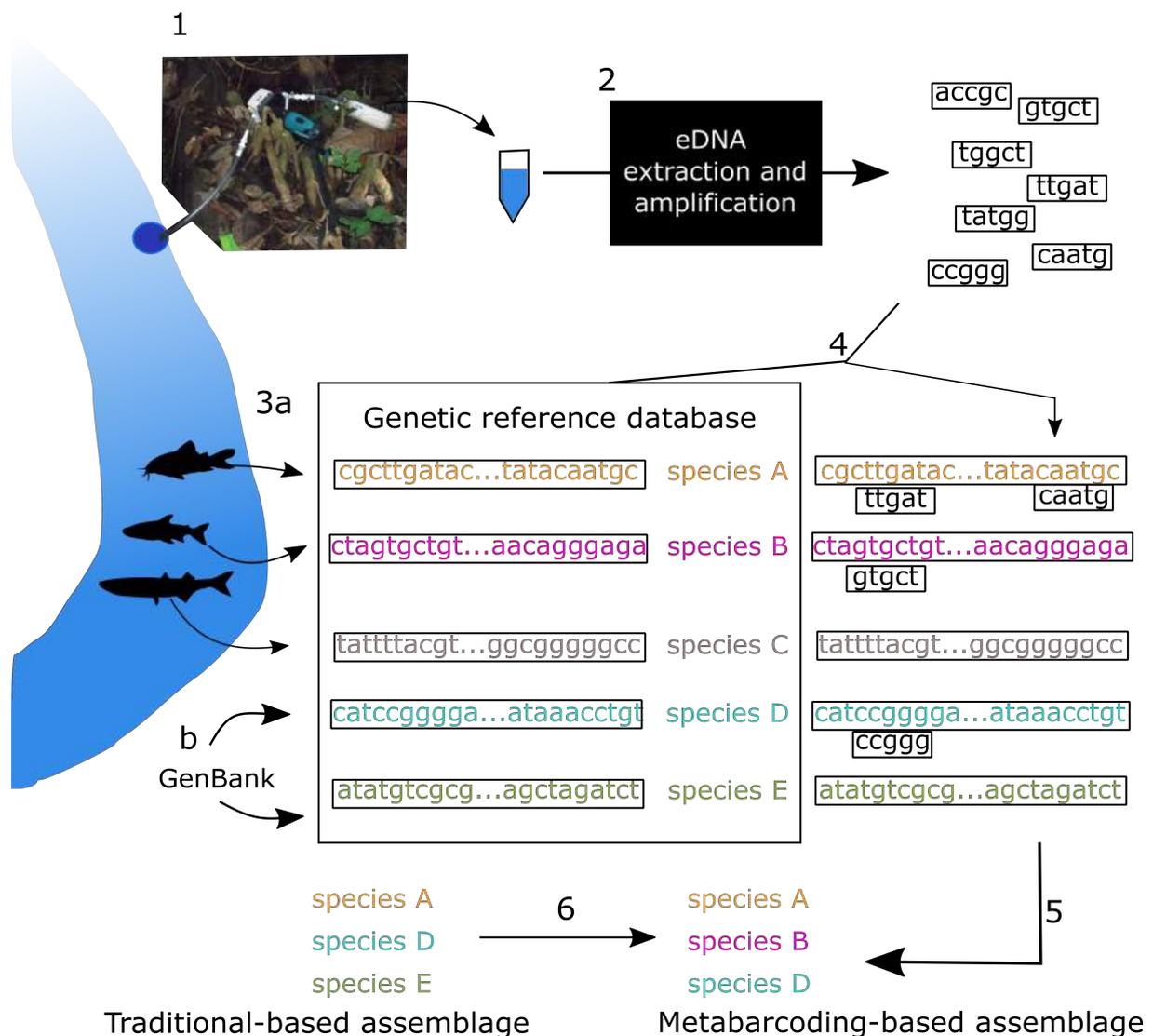
## IV.1 Summary

Extending the previous templates to others part of the hydrographic network of French Guiana or testing the impact of anthropogenic disturbances on the processes structuring fish assemblages depend on our capacity to exhaustively describe local assemblages. However, there is currently no method available to do so in French Guiana. Indeed, the rotenone sampling used to build the fish assemblage database of the previous chapters has been regulated by European authorities since 2008 (European laws 2008/296/CE and 2008/317/CE) and is now banned. Moreover, its strong deleterious impact on the fauna makes it a relatively bad tool for biodiversity conservation projects. Nets sampling for large rivers suffers from the same shortcoming: gill-nets cause a substantial fish mortality. Nets sampling is also selective toward species and only efficient for the species living near the bank and having a specific size (defined by the mesh size of the nets) are sampled. Alternatives to these destructive sampling protocols are needed to pursue ecological and evolutionary studies that comply with laws on animal welfare.

Among the sampling methods for freshwater fish fauna, electrofishing, which is widely used and has a low impact on fish, has been proposed as an alternative to rotenone sampling in small Guianese streams. However, the conditions for its application (high conductivity and low water depth) were limited to a small number of streams (Allard, 2014). Direct underwater observations of fish assemblages, although not impacting freshwater fauna and being used both in streams and rivers can require high sampling effort depending on the water turbidity and the presence of diurnal and nocturnal or cryptic species. The development of a sampling method with a relatively low sampling effort, applicable to both streams and rivers and that provide a reliable image of fish assemblages is still pending.

Such an alternative might rise through the development of DNA metabarcoding (Figure IV.1). DNA metabarcoding designates the DNA-based identification of multiple species from a mix of specimens or from the DNA present in an environmental sample (soil, water, air). In the latter case, the method is qualified as environmental DNA metabarcoding (eDNA metabarcoding). The eDNA metabarcoding approach has demonstrated its efficiency in temperate streams, where it often gave more complete image of local assemblages than traditional methods. However, its applications to tropical freshwaters are scarce (Simpfendorfer *et al.*, 2016; Bellemain *et al.*, 2016; Lopes *et al.*, 2017) and tests on highly diverse assemblages such as the Guianese freshwater fish are needed.

A first lock to apply eDNA metabarcoding to Guianese water is the lack of genetic



**Figure IV.1:** Summary of the eDNA metabarcoding protocol used to sample Guianese freshwater streams and assess fish assemblages. 1: For each site, we filtered about 60 liters of water (30 minutes filtering time) in a rapid flow area. 2: DNA extraction, amplification and curation of the samples and reads analyses were handled by SPYGEN (a private company specialized on the use of eDNA for species detection). 3: We built a genetic reference database to assign eDNA. Sequences were mainly obtained by sequencing individuals caught during fieldwork sessions and complemented by samples provided by HYDRECO lab (a). Reference database was complemented with a few additional sequences available from GenBank public repository (b). 4: Assignment of eDNA reads to the genetic reference database. Several protocols have been proposed (Blast, Altschul *et al.*, 1990; Edgar, 2010; OBITools, Boyer *et al.*, 2016; phylogenetic placement, Munch *et al.*, 2008). Here we used ecotag to identify the species present in the sample. 5: The site species list obtained by eDNA metabarcoding. 6: The list of species produced by metabarcoding was compared to the list of species obtained with other sampling methods (nets and rotenone) or from external databases. (Photo: K. Cilleros)

resources available in public repositories (e.g. GenBank). We thus first built a genetic reference database for the 12S eDNA metabarcoding fragment on 181 freshwater fish species of French Guiana using our tissue collection. We then sampled 39 sites for eDNA, in both streams and rivers in 2014 and 2015 and compared the identity of species detected using traditional sampling (rotenone for streams and nets for rivers) and those detected using eDNA metabarcoding. We also compared the diversity patterns ( $\alpha$ - and  $\beta$ -diversity) derived from traditional and eDNA sampling.

First, we compared the eDNA to the Genbank public reference database to search for false detection of species belonging from others biogeographic realms and confirmed that only Amazonian species were detected. We then used our custom reference database to compare fish assemblages derived from eDNA and traditional samples using gillnets (for rivers) and rotenone (for streams). Among the 216 species detected by at least one method, 119 species were detected by both methods, 13 were exclusively detected by eDNA metabarcoding and 84 only by traditional methods. This last number was mainly due to species uninformed in the reference database (54), and to a lower extent by species that cannot be discriminated using the 12S eDNA fragment (23). The remaining 8 species account for those informed but not detected. At the drainage basin scale, only five species out of the 132 detected by the eDNA metabarcoding were detected outside their known distribution. In river sites, we were able to detect species inhabiting streams and rivers, and in stream sites, we only detected stream species. This can be interpreted as a transport of eDNA with the water flow. Locally, the congruence between the two sampling methods was relatively low (about 27% of species in common) and highly variable between streams and rivers. We however found that richness estimates were correlated as well as species relative occurrence. Moreover, the distinction of assemblages by drainage basin was more marked when using eDNA metabarcoding, whereas the sampling method mostly affected assemblage composition variation.

This work is currently under review in *Molecular Ecology Resources*.

## IV.2 Manuscript C

### Unlocking biodiversity and conservation studies in high diversity environments using environmental DNA (eDNA): a test with Guianese freshwater fishes

*Molecular Ecology Resources*, under review

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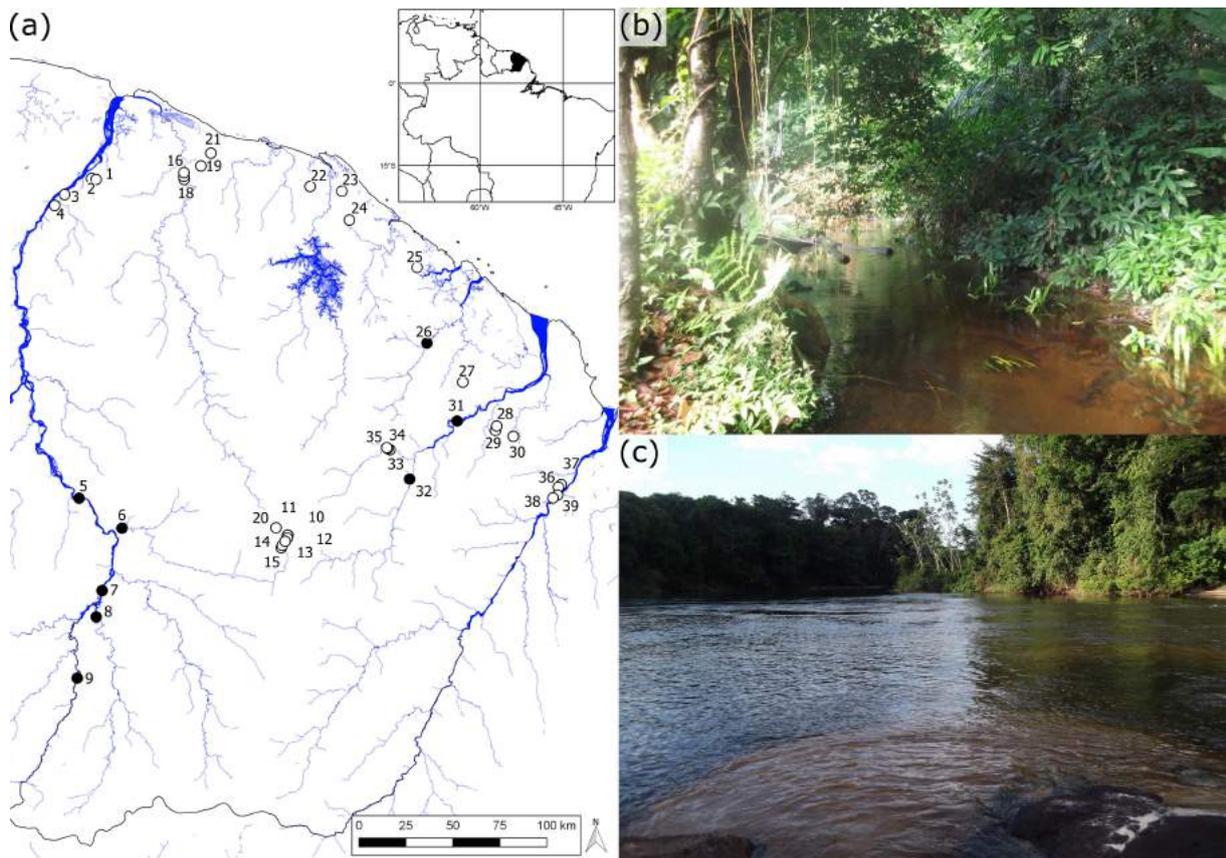
## INTRODUCTION

Evaluating the distribution or the occurrence of organisms is a crucial step in biodiversity science. Achieving those tasks can be difficult when assemblages are species rich and/or when the organisms cannot be directly observed (Murphy & Willis, 1996). This is particularly true for fish in tropical freshwater ecosystems, where local assemblages count dozens of species and their observation is limited by water turbidity, depth, and current velocity. Hence, fish are often sampled using nets, electricity and even toxicants (Allard, 2014; Murphy & Willis, 1996; Portt *et al.*, 2006). These traditional methods are selective towards species (Gunzburger, 2007), and some of these methods, such as gill nets and toxicants, are destructive for the fauna (Dalu *et al.*, 2015; Snyder, 2003). Their use for scientific purposes is highly debated and the development of alternative non-destructive methods is urgently needed to comply with ethics and laws on animal welfare and biodiversity conservation (Ellender *et al.*, 2012; Hickey & Closs, 2006; Thomsen & Willerslev, 2015). With advances in sequencing technologies, the use of environmental DNA (eDNA), total DNA present in environment samples, like faeces, soil, air or water, has gained huge consideration to study biodiversity in the last years (Taberlet *et al.*, 2012; Valentini *et al.*, 2009). To date, eDNA usefulness and efficiency has been assessed under controlled and natural conditions in temperate freshwaters where eDNA provided a realistic picture of fish species assemblages (Jerde *et al.*, 2011; Thomsen *et al.*, 2012; Civade *et al.*, 2016; Hänfling *et al.*, 2016; Valentini *et al.*, 2016). Environmental DNA hence offers the opportunity to reduce sampling effort (Jerde *et al.*, 2011; Olds *et al.*, 2016), and to save time and money compared to traditional fish sampling methods under temperate environments (Fukumoto *et al.*, 2015; Jerde *et al.*, 2011).

The situation markedly differs in the tropics that host higher species richness than temperate areas. For instance, French Guiana counts as much fish species as Western Europe (380 species) while its surface accounts for less than 1% of Western Europe (Le Bail *et al.*, 2012; Melki, 2016). Moreover our ability to make fish inventories in tropical rivers is limited because the frequent low conductivity of rainforest streams makes electrofishing ineffective, and the turbidity of large rivers limits direct underwater observations (Allard, 2014). These limitations led to use a combination of netting methods (gillnets, cast nets, hand nets), sometimes combined with toxicants (e.g. rotenone), to get a picture of the fauna in both rivers and streams (Allard *et al.*, 2016; Araújo *et al.*, 2009). Despite the use of multiple sampling methods, such inventories often remain incomplete, especially in large rivers, because most species have a low density and hence are difficult to detect (Hercos *et al.*, 2013). As a consequence, gathering data on entire fish assemblages in the tropics are almost impossible without sacrificing a substantial part of the fauna, and/or strongly disturbing the environment. This obviously acts as a barrier to scientific advances on ecosystem structure and functioning, and on associated biodiversity conservation and management issues. Developing fish eDNA in tropical freshwaters is therefore more than a way to reduce sampling effort, as it would open avenues for tropical biodiversity research and conservation.

Nevertheless, there is no guarantee that eDNA will be as efficient in tropical rivers as in temperate ones. The higher temperature of tropical waters do not affects eDNA degradation (Robson *et al.*, 2016), but the stronger solar radiation and the water acidity and turbidity might speed up its degradation or impact its detection rate and thus restrict its efficiency (Barnes *et al.*, 2014; Matheson *et al.*, 2014; Pilliod *et al.*, 2014). Field studies using eDNA in tropical environment are growing (Bellemain *et al.*, 2016; Lopes *et al.*, 2017; Simpfendorfer *et al.*, 2016) but most of them focused on a single species (but see Lopes *et al.*, 2017). Those studies detected the targeted species in fewer locations than expected. This bias was more pronounced in large rivers, leaving uncertainties on the method efficiency in large tropical rivers and/or on the current spatial distribution of the species determined by traditional sampling methods (Bellemain *et al.*, 2016; Simpfendorfer *et al.*, 2016). To our knowledge, Lopes *et al.* (2017) was the only study using group-specific primers to identify simultaneously several taxa in a tropical freshwater environmental sample (hereafter called eDNA metabarcoding; Taberlet *et al.*, 2012). This study focussed on a small fraction of South American anuran species (11 frogs from Brazilian streams) and succeeded to detect those species, even at low densities. Such metabarcoding approach therefore deserves to be tested on entire and more diverse assemblages, but this implies to develop reference molecular databases made of known species sequences to be able to assign species to eDNA sequences (Ardura *et al.*, 2013; Pochon *et al.*, 2015). Such reference databases are currently lacking for most tropical freshwater species.

Here, we tested the efficiency of using eDNA with metabarcoding approach to freshwater fish diversity. We aimed to get a picture of freshwater fish assemblages in 39 rivers and streams of French Guiana. We first developed a reference database



**Figure IV.2:** Location of the 39 studied sites (a). Stream (e.g. Apa, site 1; b) and river sites (Machicou, site 32; c) are marked with open and full circles, respectively. Numbers are as in Table IV.1

gathering 191 species (out of the 380 known freshwater fish species; de Mérona *et al.*, 2012; Le Bail *et al.*, 2012), to assign eDNA sequences to species. Then, we compared the fish species assemblage detected by metabarcoding to the local fish fauna known from these sites. Here, we used a hierarchical framework and tested if metabarcoding results were consistent with the fauna known from the river drainage basin, the hydrologic unit (stream vs river) and the local site. Finally, we measured the congruence between the diversity patterns (richness, occurrence,  $\beta$ -diversity) estimated using metabarcoding and those derived from traditional sampling methods. The Guianese fish assemblages partly differ between river drainages due to biogeographic reasons (Le Bail *et al.*, 2012). They also partly differ between upstream (streams, with low species richness) and downstream areas (rivers, higher species richness), as some species only inhabit streams whereas others live only in rivers (de Mérona *et al.*, 2012). We thus tested how the biogeographical and habitat patterns derived from eDNA metabarcoding and traditional methods fit with the theoretical knowledge on Guianese fish assemblages.

## **METHODS**

### **Study area and sample location**

We sampled 39 watercourse sections of French Guiana for fish using both traditional methods (fish nets and toxicants) and eDNA metabarcoding (see Table IV.1 and Fig. IV.2a). The sites included both small streams (less than 10 m wide and 1 m deep, 31 sites; Fig. IV.2b) and rivers (more than 20 m wide and 1 m deep, 8 sites; Fig. IV.2c). These sites were located within all the 8 major river drainages of French Guiana, upstream from the main human settlements and away from major human disturbances (mining or deforestation). This insured that fish assemblages were not modified by human activities during the sampling period (2010 to 2015). Traditional and metabarcoding sampling occurred on the same year for rivers (2015). Streams were sampled from 2010 to 2013 using traditional methods, and metabarcoding samples were done in 2014 (Table IV.1). For streams, traditional sampling was not possible in 2014 because we were not allowed to use rotenone since late 2013. Nevertheless, local fish assemblages should not change across years because stream fish have low dispersal abilities (Cilleros *et al.*, 2016) and are strongly dependent on the physical structure of their habitat (Allard *et al.*, 2016; Brosse *et al.*, 2013). Moreover, all the samples were collected during the dry season (September-November) to ensure fish assemblage differences between methods are not due to seasonal changes in habitat use or migrations. In each site, we converted fish species abundance data into occurrence data to avoid potential bias due to differences in sampling techniques and potential differences in sampling efficiency between sites.

**Table IV.1:** List of studied sites with their river drainage membership and the type of watercourse (stream or river). See Figure 1 for site locations. Coordinates are given in the WGS84 coordinate reference system

Site number	Site name	Drainage basin	Watercourse type	Latitude	Longitude	Date of traditional sampling	Date of eDNA sampling
1	Apa	Maroni	Stream	5.3432	-54.0869	2012	2014
2	Crique des Cascades	Maroni	Stream	5.3476	-54.1053	2012	2014
3	Crique Bastien	Maroni	Stream	5.27003	-54.23433	2012	2014
4	Crique Penta	Maroni	Stream	5.21758	-54.28227	2012	2014
5	Papaichton	Maroni	River	3.80456	-54.16561	2015	2015
6	Saut Sonnelle	Maroni	River	3.66042	-53.95992	2015	2015
7	Twenke	Maroni	River	3.35922	-54.05492	2015	2015
8	Pikin Tabiki	Maroni	River	3.23267	-54.08319	2015	2015
9	Apsik Icholi	Maroni	River	2.93669	-54.174	2015	2015
10	Crique Nouvelle France 6	Maroni	Stream	3.62707	-53.16569	2011	2014
11	Crique Nouvelle France 5	Maroni	Stream	3.62707	-53.16692	2011	2014
12	Crique Nouvelle France 4	Maroni	Stream	3.61284	-53.16884	2011	2014
13	Crique Nouvelle France 3	Maroni	Stream	3.59726	-53.17847	2013	2014
14	Crique Nouvelle France 2	Maroni	Stream	3.57697	-53.19268	2011	2014
15	Crique Nouvelle France 1	Maroni	Stream	3.56573	-53.1975	2011	2014
16	Crique Voltalia 4	Mana	Stream	5.37639	-53.663	2011	2014
17	Crique Voltalia 3	Mana	Stream	5.35789	-53.66336	2011	2014
18	Crique Voltalia 2	Mana	Stream	5.3427	-53.66158	2011	2014
19	Crique Petit laussat Aval	Mana	Stream	5.40887	-53.58121	2012	2014
20	Crique l'Est	Mana	Stream	3.66264	-53.22197	2012	2014
21	Crique Organabo	Organobo	Stream	5.46971	-53.534	2012	2014

Table IV.1 continued

22	Crique Toussaint	Sinnamary	Stream	5.30908	-53.05873	2010	2014
23	Crique Paracou	Sinnamary	Stream	5.28692	-52.90708	2012	2014
24	Crique Eau Claire	Kourou	Stream	5.14711	-52.87149	2010	2014
25	Crique Humus	Kourou	Stream	4.91867	-52.54702	2012	2014
26	Saut Bief	Comte	River	4.55343	-52.49948	2015	2015
27	Crique Petit Approuague	Comte	Stream	4.36481	-52.32803	2011	2014
28	Crique Kapiri 6	Approuague	Stream	4.15347	-52.16586	2011	2014
29	Crique Kapiri 1	Approuague	Stream	4.13208	-52.17138	2012	2014
30	Crique Kapiri 2	Approuague	Stream	4.10373	-52.08655	2012	2014
31	Athanase	Approuague	River	4.17772	-52.35567	2015	2014
32	Machicou	Approuague	River	3.89741	-52.58253	2015	2014
33	Crique parare 2	Approuague	Stream	4.03861	-52.67697	2010	2014
34	Crique parare 5	Approuague	Stream	4.04399	-52.68746	2010	2014
35	Crique parare 8	Approuague	Stream	4.04848	-52.69214	2010	2014
36	Crique SM	Oyapock	Stream	3.8587	-51.87103	2012	2014
37	Crique Marie	Oyapock	Stream	3.87064	-51.85653	2012	2014
38	Crique Pied Saut	Oyapock	Stream	3.80605	-51.89595	2012	2014
39	Crique Minette	Oyapock	Stream	3.82033	-51.875	2012	2014

### **Traditional methods**

We adapted the traditional method protocol to the type of watercourse. For small streams, we conducted fish surveys under several different research projects between 2010 and 2013 (CNRS-Nouragues, PAG-DEAL-HYDRECO, CEBA-DIADEMA projects). We standardized the sampling protocol for all sites (Allard *et al.*, 2016). At each site, we isolated a river section located upstream of a confluence using two fine mesh (4 mm) stop nets. The length of each section was proportional to stream width and was on average  $33.67 \pm 12.13$  m. We collected fish after releasing a small quantity of rotenone (PREDATOX<sup>®</sup>: a 6.6% emulsifiable solution of rotenone extracted from *Derris elliptica* by Saphyr, Antibes, France) a few meters upstream of the first net. The rotenone is a nonselective piscicide traditionally used by Amazonian tribes, but also often used by scientists to sample fish in tropical streams (Allard, 2014; Murphy & Willis, 1996) and control fish populations (Finlayson *et al.*, 2000). All stunned fish were collected and at the end of each sampling session, we searched for fishes lying on the bottom or hidden in the leaves and debris. No rotenone sample was collected after 2013, since the use of rotenone was banned, according to European laws 2008/296/EC and 2008/317/EC, making therefore impossible to collect entire fish assemblages using this method since this date. There is still no alternative to rotenone, since electrofishing is not efficient due to the low conductivity of Guianese streams and attempts to make inventories using others methods (snorkelling, hand nets, cast nets, traps) yielded incomplete fish inventories (Allard, 2014). For rivers, we conducted fish surveys in November 2015. At each site, we placed 20 50-meters long gill-nets with different mesh sizes (15, 20, 25, 30 and 35 mm) on river banks and removed them after an overnight sampling to collect fishes. In addition, we achieved cast net and fine meshed hand net samples close to the banks to complement inventories for small species not captured using gill nets. For both methods, we identified each individual to the species level and we collected tissues for each species to build up the reference DNA database.

### **Reference DNA database**

We sampled tissues from 503 specimens belonging to 231 species (1 to 3 specimens per species), and stored them in 96% ethanol. We extracted DNA from 0.05–0.25 cm<sup>2</sup> of tissue using salt-extraction protocol (Aljanabi & Martinez, 1997). We performed DNA amplification in a final volume of 25 µL including 1U of GoTaq<sup>®</sup> (Promega), 5X buffer (Promega), 10 µM of dNTP, 20 µM of each primer (teleo\_R 5'-CTTCCGGTACACTTACCATG-3' and V05F.898 5'-AAACTCGTGCCAGCCACC-3', Valentini *et al.*, 2016) and 1µL of DNA template. The PCR mixture was denatured at 95°C for 10 min, followed by 35 cycles of 30 s at 95°C, 30 s at 55°C and 1 min at 72°C, and followed by a final elongation at 72°C for 7 min, in a room dedicated to amplified DNA, with negative air pressure and physically separated from the DNA extraction rooms. A 600 bp product of the 12S rRNA gene were obtained and sequenced using Sanger sequencing method by Genoscreen biotechnology

company, except for 27 species for which no DNA fragment was amplified. Sequences were analysed using GENEIOUS version 9.0<sup>1</sup>, Kearsse *et al.*, 2012).

### **eDNA metabarcoding sampling and analysis**

We collected eDNA samples in November 2014 for streams and in October 2015 for rivers, based on the (Valentini *et al.*, 2016) protocol for running waters. For each sample, we used a filtration kit made of a sterile filtration capsule (Enviroheck HV 1 µm; Pall Corporation, Ann Arbor, MI, USA), a peristaltic pump (Vampir Sampler; Bürkle GmbH, Bad Bellingen, Germany) and sterile single use tubing. All the materials were handled with sterile gloves. The peristaltic pump, although not in contact with the water, was sterilized using sodium hypochlorite between each site to avoid contamination. At each site, we placed the input part of tubing in a high flow part of the watercourse located in the middle of the stream or river channel. Sampling was achieved in rapids hydromorphologic units to ensure an optimal homogenisation of the water through the water column. The operator always remained downstream from the filtration area, and stayed on the bank (for small streams) or on emerging rocks (for larger streams and rivers). Water was pumped about 20 cm below the surface and each filtration lasted 30 minutes at 1.67 L.min<sup>-1</sup>. Each sample hence results from the filtration of c.a. 50 litres of water. For sites located along the same river course, we did sampling from downstream to upstream to avoid contamination by eDNA transport by the boat (for rivers) or on our clothes. For the same reason, we took eDNA samples just upstream from the nets. At the end of the filtration, we emptied the filtration capsule of water, filled it with 150 mL of preservation buffer (Tris-HCl 0.1 M, EDTA 0.1 M, NaCl 0.01 M, and N-lauroyl sarcosine 1%, pH 7.58) and stored it in the dark in sterile individual plastic bags. Samples were then stored at room temperature before DNA extraction. Extraction was achieved at the end of the field session, 1 to 3 weeks after sampling. For DNA extraction, filtration capsules were left at 56°C for 2 h, agitated manually for 5 minutes and then emptied into three 50 mL tubes. In total, approximately 120 mL were retrieved in three tubes that were centrifuged for 15 minutes at 15,000 g. Supernatant was removed with a sterile pipette, leaving 15 mL of liquid at the bottom of the tube. Subsequently, 33 mL of ethanol and 1.5 mL of 3M sodium acetate were added to each 50 mL tube. The three tubes were centrifuged at 15,000 g for 15 min at 6°C and the supernatant was discarded. After this step, 360 µL of ATL Buffer of the DNeasy Blood & Tissue Extraction Kit (Qiagen) were added to the first tube, the tube was vortexed and the supernatant was transferred to the second tube (Tréguier *et al.*, 2014). This operation was repeated for all tubes. The supernatant of the third tube was finally transferred to a 2 mL tube and the DNA extraction was performed following the manufacturer's instructions. Two negative extraction controls were also performed. They were amplified and sequenced in the same way and in parallel to the samples to monitor possible contaminations.

We performed DNA amplifications were performed in a final volume of 25 µL

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<sup>1</sup><https://www.geneious.com/>

including 1 U of AmpliTaq Gold DNA Polymerase (Applied Biosystems, Foster City, CA), 10 mM of Tris-HCl, 50 mM of KCl, 2.5 mM of MgCl<sub>2</sub>, 0.2 mM of each dNTP, 0.2 μM of teleo primers (teleo\_R 5'-CTTCCGGTACACTTACCATG-3' and teleo\_F 5'-ACACCGCCCGTCACTCT-3', Valentini *et al.*, 2016) and 3 μL of DNA template. We added also to the mixture 4 μM of human blocking primer for teleo primers and 0.2 μg/μL of bovine serum albumin (BSA, Roche Diagnostic, Basel, Switzerland). The teleo primers were 5'-labeled with a seven-nucleotide tag unique to each sample (with at least three differences between any pair of tags) allowing the assignment of each sequence to the corresponding sample during sequence analysis. Tags for forward and reverse primers were identical for each sample. The teleo primer was preferred to Mifish primer (Miya *et al.*, 2015), because Valentini *et al.* (2016) demonstrated that Mifish provide slightly higher taxonomic resolution than teleo while being twice longer. Teleo primers were therefore preferred to optimize the robustness of the amplification. PCR was conducted for 12 replicates with the same protocol described in the 'Reference DNA database' section increasing the number of PCR cycles to 50.

We also amplified two extraction negative controls and three PCR controls and sequenced them in parallel with the 39 samples. We pooled the purified PCR products in equal volumes, to achieve an expected sequencing depth of 400 000 reads per sample. Library preparation and sequencing were performed at Fasteris facilities (Geneva, Switzerland). Four libraries were prepared using the Metafast protocol<sup>2</sup>. For streams samples, we carried out the paired-end sequencing (2x125 bp) in an Illumina MiSeq sequencer (Illumina, San Diego, CA, USA) using the Paired-end MiSeq Reagent Kit V2 (Illumina, San Diego, CA, USA) following the manufacturer's instructions. For rivers samples, we ran the libraries on an Illumina HiSeq 2500 (2x125 bp) (Illumina, San Diego, CA, USA) using the HiSeq SBS Kit v4 (Illumina, San Diego, CA, USA) following the manufacturer's instructions. We analyzed sequence reads using the programs implemented in the OBITools package<sup>3</sup> (Boyer *et al.*, 2016) following the protocol described in Valentini *et al.* (2016). We performed the taxonomic assignment of Molecular Operational Taxonomic Units (MOTUs) using the program *ecotag* with the local reference database built for this study and with GenBank nucleotide database. The program works in two steps: first, it looks for the reference sequence that has the highest similarity with the query sequence. This value of similarity is then used as a threshold to look for sequences in the reference database for which the similarity with the first match reference sequence is equal or lower than the threshold. The query sequence is then assigned to the most recent common ancestor of the first matched sequence and the second. MOTUs with similarity lower than 98% with the reference database (local or GenBank) were discarded. To take into account tag jumping (Schnell *et al.*, 2015), we discarded all sequences with a frequency of occurrence below 0.0003 per taxon per sample and per sequencing run. These thresholds were empirically determined to clear all reads from the extraction and PCR negative controls included in our global data production procedure

<sup>2</sup><http://www.fasteris.com/dna/?q=content/metafast-protocol-amplicon-metagenomic-analysis>

<sup>3</sup><https://git.metabarcoding.org/obitools/obitools/wikis/home>

(De Barba *et al.*, 2014).

### Statistical analyses

First, we compared the species detected by eDNA metabarcoding to the list of species known at three different spatial scales: the river drainage scale, the watercourse type and the site scale. At the drainage scale, we used an updated version of Le Bail *et al.* (2012) and Melki (2016) checklists to verify if the species detected using eDNA metabarcoding belong to the species assemblage known to inhabit the considered river drainage. At the watercourse type scale, we merged all available ecological information on the species using Keith *et al.* (2000); Le Bail *et al.* (2000); Planquette *et al.* (1996) to determine if each species inhabits only streams, only rivers or both streams and rivers. At the site scale, we compared the list of species, genera and families obtained by metabarcoding to the list of species, genera and families collected by traditional sampling. Then, we calculated the percentage of species detected in each site by each method as the number of species detected by one method divided by the total number of species detected at this site. We also calculated the percentage of species detected only by metabarcoding or only by traditional methods by separating species detected only by one method from species detected by both methods.

Afterwards, we tested the congruence of diversity patterns given by the two methods. We calculated the species richness at each site for both methods and tested the correlation between them using Pearson's  $r$  correlation coefficient. We calculated species relative occurrence as the percentage of sites where a species was detected and we tested if the occurrence patterns were correlated using Kendall's  $\tau$  rank correlation coefficient.

Finally, we used  $\beta$ -diversity between sites to characterize spatial patterns of fish assemblage diversity. We calculated  $\beta$ -diversity using the turnover component of the Jaccard's dissimilarity index (Baselga, 2012) and used non-metric multidimensional scaling (NMDS) to visualize  $\beta$ -diversity patterns in two dimension plots. To test the congruence of produced ordinations between the two methods, we used Procrustes analyses with 999 permutations. We measured the goodness-of-fit between ordinations using  $m$  statistic, which varies from 0 (perfect congruence) to 1 (no congruence). We ran this procedure after assigning metabarcoding reads to species. We also tested the congruence between methods using the MOTUs without species assignment matrices to determine if raw metabarcoding data can be used to describe spatial patterns in fish assemblages. We used nonparametric multivariate analysis to test if fish assemblages differed in their composition and their variability i) between streams and rivers, and ii) between river drainages. In this last analysis, in addition to the complete dataset, we also used a reduced dataset comprising only the streams. The difference of species composition between river drainages based on river fish fauna (excluding streams) was not tested because the number of river sites ( $n = 8$ ) was not sufficient to provide a meaningful representation of each river drainage.

We tested differences in composition between groups with a permutational multivariate analysis of variance (PERMANOVA, Anderson, 2001) and differences in assemblage variation within a group with an analysis of homogeneity of multivariate dispersion (PERMADISP, Anderson, 2006). Significance was assessed with 999 permutations.

All statistical analyses were performed using R.3.3.2 (R Core Team, 2015) and the package 'vegan' version 2.4-1 (Oksanen *et al.*, 2016), 'betapart' version 1.3 (Baselga *et al.*, 2013) and 'dunn.test' version 1.3.4 (Dinno, 2017). Significance threshold was fixed at  $p < .05$ .

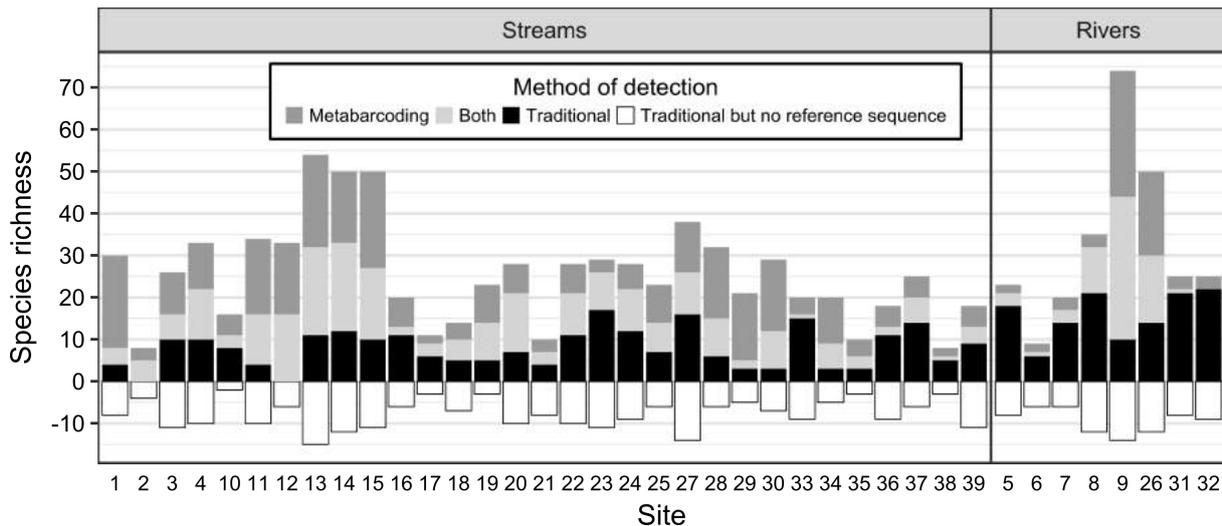
## RESULTS

With traditional methods, we collected 7,029 individuals that represented 203 species (Table IV.1) from 12 orders. In almost all sites, cryptic fish were collected, including highly cryptic Siluriformes and litter bank fishes such as small killifishes. Several species of Gymnotiformes inhabiting small Guianese streams, although known to be resistant to rotenone, were also caught.

Among the 503 specimens sequenced to build-up the 12S mitochondrial gene reference database, 191 unique sequences were obtained. They were assigned to species using the 12S metabarcoding fragment as in Valentini *et al.* (2016) (see Materials and Methods). When a sequence matched several reference taxa, they were assigned to the lowest taxonomic level (genera, family or order) grouping all the matched reference taxa (Table IV.S1). 181 unique sequences were identified to the species level, six to the genus (*Bryconops*, *Gymnotus*, *Hemigrammus*, *Leporinus*, *Moenkhausia*, *Pimelodella*), three to the family (Characidae, Hypopomidae, Prochilodontidae) and one to the order (Characiformes; Table IV.S1).

For the eDNA metabarcoding analysis, a total of 17,775,665 reads were obtained, among which 2,015,596 were assigned when using GenBank database as reference (11%, see Table IV.S2). The assigned taxa corresponded to fish species present in French Guiana or in neighbouring Amazonian regions (1,050,515 reads), but reads were also assigned to other diverse taxa (Anura, Mammalia, Aves, Insecta and Reptilia; Table IV.S3). Among the reads assigned to fish taxa, 904,782 (86%) were assigned to 28 taxonomic units occurring in French Guiana (22 species and 4 genera; Table IV.S3). The 14% remaining reads were assigned to species closely related to Guianese ones (all those genera are present in French Guiana). At the site scale, the number of reads assigned to GenBank data ranged from 0 to 296,917 (median: 19,715 reads).

Using our custom reference database, 8,109,492 reads were assigned (46%), with a number of reads assigned at each site ranging from 18,895 to 870,235 (median: 112,997 reads, Table IV.S2). The reads assigned to the reference database were distributed among 148 taxonomic units: 132 species (7,521,532 reads), nine genera (550,264 reads), two sub-families (638 reads), four families (29,503 reads) and one order (495 reads). When comparing the complete lists of species detected with the two methods (*i.e.* removing from the analysis all individuals and assigned reads



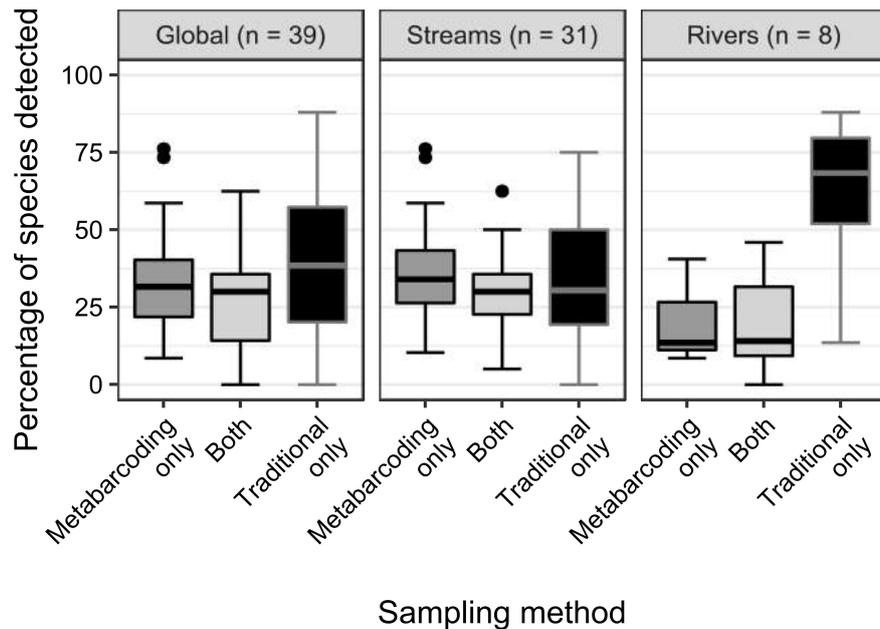
**Figure IV.3:** Species richness per site detected with traditional and eDNA metabarcoding methods. Species caught only with traditional methods are in black (species informed in the reference database) and white (absent from the reference database), detected only with metabarcoding in dark grey and by both metabarcoding and traditional methods in light grey

determined at the genus level or higher), we found that 119 species were detected by the two methods, 13 only by metabarcoding and 84 only by traditional methods (Table IV.1). Within these 84 species, 53 had no reference sequence. Among the 31 species detected by traditional methods and referenced in our reference sequence database, 23 were not assigned to a species but to higher taxonomic level (from genus to order), and only 8 were not detected by eDNA while referenced in our reference sequence database.

At the river drainage scale, five species (*i.e.* 3.79% of the 132 species detected by metabarcoding) were detected outside their known Guianese river drainages distribution. These detections represented 12 out of a total of 680 fish occurrences (*i.e.* 1.76% of fish occurrences). Moreover, three out of the five species were detected no more than twice in river drainages out of their known distribution (*Corydoras aeneus* detected at one site in the Oyapock River drainage, *Hemiodus quadrimaculatus* once in the Maroni, *Ancistrus aff. temminckii* once in the Kourou and once in the Sinnamary and *Krobia itanyi* in two sites in the Approuague). The remaining species, *Krobia aff. guianensis sp1*, was detected six times in the Approuague River drainage.

When focusing on the watercourse type, 27 species (20.45% of the 132 species) detected in rivers have not been caught in rivers and are only known from streams. For streams, only three detected species (2.27% of the 132 species) have not been caught in streams (*Crenicichla multispinosa*, *Mastiglanis cf. asopos* and *Plagioscion squamosissimus*).

At the site scale, the number of species detected was lower with metabarcoding than with traditional methods in 29 out of the 39 sampled sites and the number of species common to both methods was low (mean  $\pm$  SD: 20.74  $\pm$  10.90 percent of species in common). When we excluded the species not informed in the reference



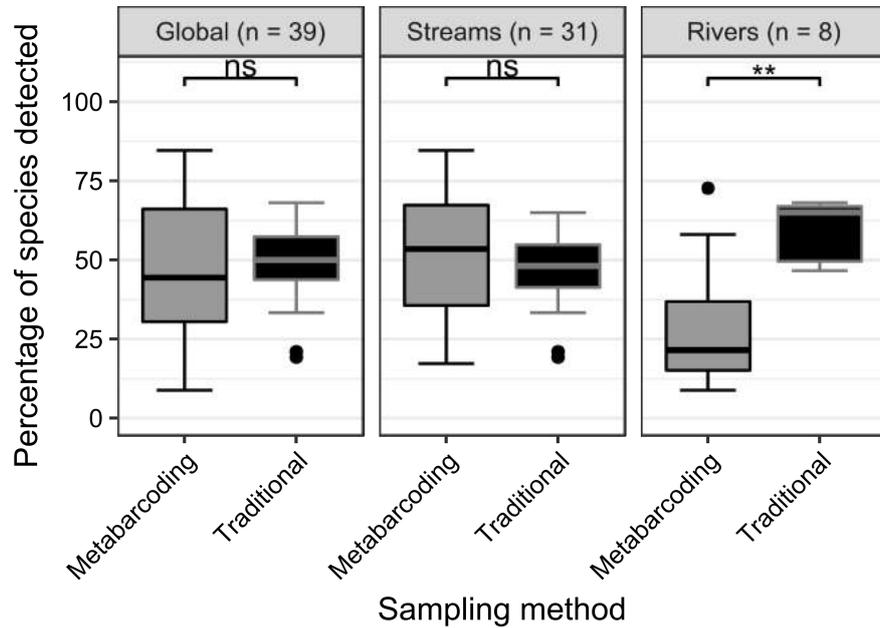
**Figure IV.4:** Percentage of species detected with traditional and eDNA metabarcoding methods. Only species informed in the reference database were considered. Species caught only with traditional methods are in black, those detected only with metabarcoding in dark grey and those detected by both metabarcoding and traditional methods (shared) in light grey

database, eDNA metabarcoding gave a lower number of species than traditional methods in 20 sites (Figure IV.3). For three sites the richness estimation was equal between methods and a higher species richness was detected by metabarcoding in 16 sites.

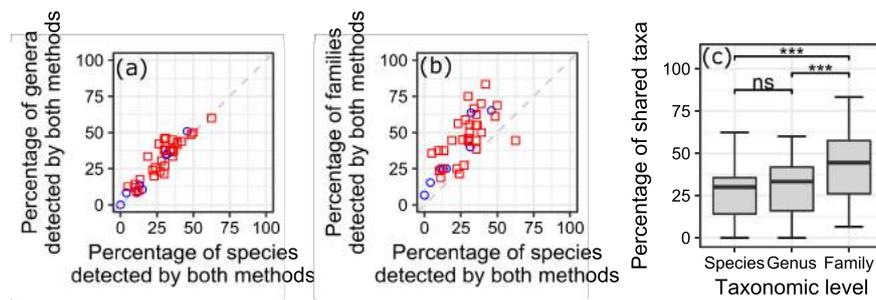
The two methods showed low congruence in detected species identity, with an average of 27.18% ( $\pm 13.92$ ) of detected species in common, and each method detected an equal amount of species (Kruskal-Wallis rank sum test:  $\chi^2 = 5.56$ ,  $p = .062$ ; Figure IV.4). Distinguishing between streams and rivers showed that the latter held true for streams ( $\chi^2 = 2.89$ ,  $p = .24$ ) but not for rivers ( $\chi^2 = 9.77$ ,  $p = .0076$ ), where traditional methods detected most of the species.

The total percentage of species detected by each method (shared between the two methods and only metabarcoding vs. shared and only traditional) did not differ between the two methods (Wilcoxon rank-sum test:  $z = -1.03$ ,  $p = .30$ , Figure IV.5). Such a trend was verified for streams ( $z = 0.61$ ,  $p = .61$ , Figure IV.5), but in the rivers the traditional methods detected a greater percentage of species than metabarcoding ( $z = -2.68$ ,  $p = .0073$ , Figure IV.5).

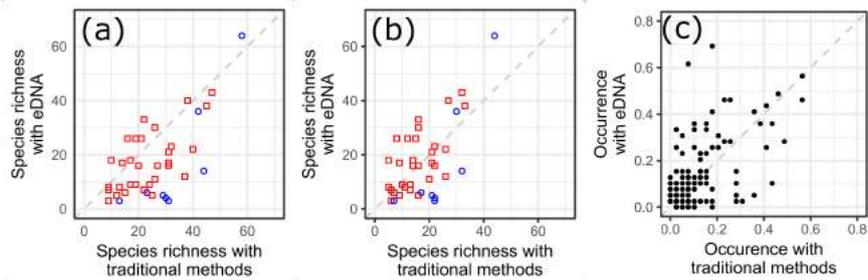
At higher taxonomic resolution scales, the results were similar using the genus taxonomic level (Figure IV.S1a, Fig. IV.S2a and IV.S3a). The congruence in detected genus identity was low between the two methods (29.90  $\pm$  14.94 percent of genera in common) and a higher percentage of genera was detected by traditional methods when considering all the sites together or only rivers (Wilcoxon test: global:  $z = -3.07$ ,  $p = .0022$ ; streams:  $z = -1.84$ ,  $p = .066$ ; rivers:  $z = -2.70$ ,  $p = .0035$ ). The percentage of genera detected by both methods (shared genera) was similar to that obtained at the



**Figure IV.5:** Percentage of species detected at each site with metabarcoding (dark grey) or traditional methods (black). Difference between eDNA and traditional methods were tested using Wilcoxon rank-sum test, ns:  $p > .05$ ; \*\*  $p < .01$



**Figure IV.6:** Percentage of genera (a) and families (b) detected by both eDNA and traditional methods compared to the percentage of species detected by both methods. The line 1:1 is represented as the dashed line on all plots and sites are classified according to the watercourse type ( $\square$  for streams and  $\circ$  for rivers). (c) Percentage of detected taxa by both metabarcoding and traditional methods according to the taxonomic level used (species, genus and family). Differences between taxonomic levels were tested using Dunns test for stochastic dominance, ns:  $p > .05$ ; \*\*\*  $p < .001$



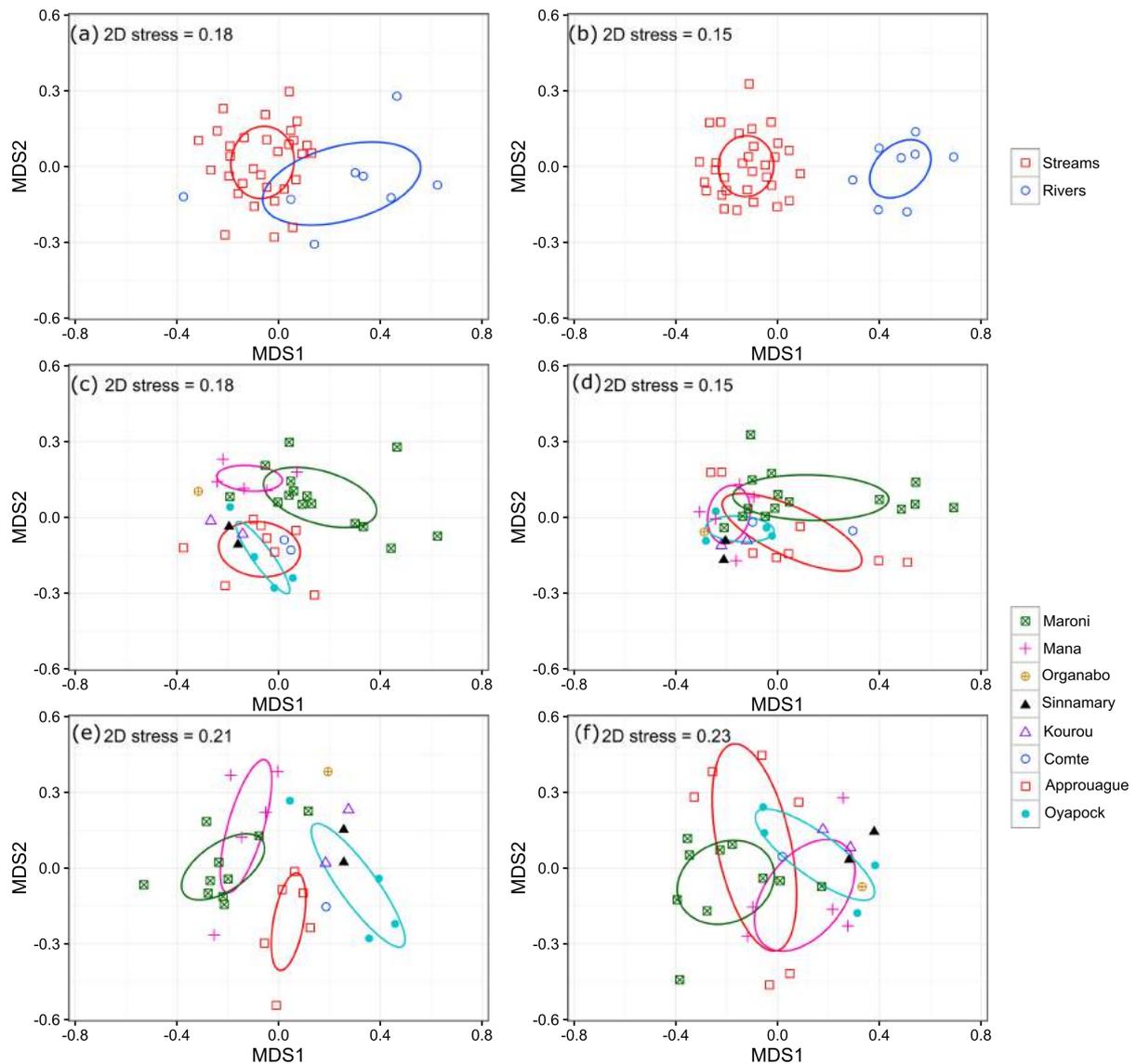
**Figure IV.7:** Relationship between species richness (a, b) and species occurrences (c) obtained with traditional methods and metabarcoding when using (a) the entire data and (b) after removing the species not informed in the reference database. Species occurrences are expressed as the percentage of sites where a species was detected. The line 1:1 is represented as the dashed line on all plots. For a and b, sites are classified according to the watercourse type (□ for streams and ○ for rivers)

species taxonomic level (Dunn's test for stochastic dominance:  $z = 0.86$ ,  $p = .19$ ; Figure IV.6a and c). At the family level, the congruence between the two methods increased (44.53–18.25 percent of genera in common; Figure IV.S1B, Fig. IV.S2b and IV.S3b) and the traditional methods still detected more families when considering all the sites together and when considering only rivers (Wilcoxon test: global:  $z = -2.47$ ,  $p = .013$ ; streams:  $z = -1.15$ ,  $p = .25$ ; rivers:  $z = -2.58$ ,  $p = .0099$ ). The percentage of families detected by the two methods was higher than the percentage of species (Dunn's test:  $z = 4.19$ ,  $p < .001$ ; IV.6b and c) and of genus (Dunn's test:  $z = 3.33$ ,  $p < .001$ ; Figure IV.6c) shared between methods.

Considering the assemblage descriptors, the species richness estimated by the two methods were significantly correlated ( $r = .64$ ,  $p = .001$ , Figure IV.7a and b). Species occurrences were also correlated, with the most widespread species detected in a greater number of sites with metabarcoding ( $r = .39$ ,  $p < .001$ , Figure IV.7c). Ordinations of sites based on  $\beta$ -diversity showed a significant but weak concordance between the two methods (Procrustes analysis:  $m^2 = .57$ ,  $p = .001$ ). When analyzing rivers and streams separately, concordances between the two methods were also significant, but weak (rivers:  $m^2 = .42$ ,  $p = .029$ ; streams:  $m^2 = .70$ ,  $p = .001$ ). When using complete MOTUs data, the concordance between traditional and metabarcoding methods was still significant but lower (Procrustes analysis:  $m^2 = .78$ ,  $p = .001$ ).

NMDS ordinations of all sites showed a distinction between the river and the stream sites for both methods (Figure IV.8a and b). Assemblage variability within group (*i.e.* stream or river) did not differ for both methods (PERMADISP: eDNA metabarcoding:  $F_{1,37} = 2.08$ ,  $p = .16$ ; traditional:  $F_{1,37} = 2.29$ ,  $p = .14$ ). Species composition however differed between rivers and streams, with stronger differences when using traditional methods (PERMANOVA: eDNA metabarcoding:  $F_{1,37} = 7.07$ ,  $p = .001$ ; traditional:  $F_{1,37} = 12.12$ ,  $p = .001$ ).

When testing the effect of river drainage membership, assemblages assessed from eDNA differed between river drainages (PERMANOVA:  $F_{1,37} = 3.06$ ,  $p = .001$ ) but their variability within river drainage did not differ (PERMADISP:  $F_{1,37} = 1.75$ ,  $p = .13$ ).



**Figure IV.8:** Non metric multidimensional scaling (NMDS) ordinations of (a, c) entire dataset for metabarcoding, (b, d) entire dataset for traditional methods, (e) only stream assemblages detected with metabarcoding and (f) only stream assemblage detected with traditional methods. For a and b, sites are classified according to the watercourse type (□ for streams and ○ for rivers) and for c, d, e and f, sites are classified according to the river drainage (see legend for river drainage membership). Ellipses represent standard deviation for each group

When using traditional methods either assemblage variability (PERMADISP:  $F_{1,37} = 8.95$ ,  $p = .001$ ) or species composition differed between river drainages. But those differences were weaker compared to the results obtained with metabarcoding (PERMANOVA:  $F_{1,37} = 1.56$ ,  $p = .011$ ). Nevertheless, metabarcoding provided a better discrimination of the stream fauna between river drainages than traditional methods (Figure IV.8c and d).

For streams, assemblages detected using metabarcoding did not vary significantly within river drainage (Figure IV.8e, PERMADISP:  $F_{7,23} = 1.64$ ,  $p = .20$ ), but their composition differed between river drainages (PERMANOVA:  $F_{7,23} = 3.85$ ,  $p = .001$ ). The variability within river drainage of fish assemblages and their composition based on traditional methods differed significantly between river drainages (Figure IV.8f, PERMADISP:  $F_{7,23} = 7.11$ ,  $p = .001$ ; PERMANOVA:  $F_{7,23} = 1.64$ ,  $p = .004$ ).

## DISCUSSION

This first study of freshwater fish diversity using metabarcoding approach in tropical freshwater showed contrasting results compared to temperate areas, where it provided an exhaustive picture of the fish assemblages (Civade *et al.*, 2016; Valentini *et al.*, 2016). The fish inventories we made using eDNA metabarcoding over the Guianese territory revealed that the local assemblage inventories were not consistent between methods. Fifty percent of the fish fauna was detected by both traditional methods and eDNA metabarcoding in all but one of the considered sites but this discrepancy might be explained by several reasons explained below.

Firstly, the incompleteness of fish assemblages inventories in tropical streams and rivers using traditional methods do not provide an exhaustive picture of the fauna (Mojica *et al.*, 2014). Indeed, most traditional methods are known as species selective (Murphy & Willis, 1996) and investigate the fauna from selected habitats and/or for a restricted reach of the river or stream that does not encompass all available habitats (Allard *et al.*, 2016), and thus some species might remain undetected locally. In contrast, eDNA metabarcoding provide a way to detect fish independently from the species local habitat (Olds *et al.*, 2016), and integrates fish data over larger scales (from a few hundred meters to several kilometres) than the local sampling habitat (Civade *et al.*, 2016; Deiner & Altermatt, 2014; Deiner *et al.*, 2016; Fukumoto *et al.*, 2015). Our inventories are nevertheless incomplete as a substantial part of the fauna known to occur in each river, previously detected using traditional methods, was not detected using metabarcoding.

Secondly, imperfect detection (Mojica *et al.*, 2014; Willoughby *et al.*, 2016) or erroneous assignments of reads to species probably explain incompleteness in eDNA-based inventories. The incompleteness of the reference database (ca. 25% of the species caught are not informed in the reference database, representing  $24.31 \pm 7.23\%$  of species at each site) might for instance explain the grouping of some reads in higher taxonomic units (genera or family). In other words, slight differences between reference sequences of the same rank, especially genus, can result in the assignment

of reads to one unique unit (Ardura *et al.*, 2013; Pochon *et al.*, 2015). That was probably the case for some genera with closely related species from a morphological point of view (and probably also from a molecular point of view, Brown *et al.*, 2015; Flynn *et al.*, 2015, such as *Bryconops*, *Leporinus* or *Pimelodella* genera. Those genera were indeed represented by a high number of reads in our results, but species discrimination was not possible and those genera were excluded from our analyses.

Gathering more molecular data on species is therefore a crucial step to develop a precise method based on eDNA to inventory species rich ecosystems (Valentini *et al.*, 2009). Public repositories are for the moment lacking information on the species occurring in these ecosystems. Here, using GenBank to assign our eDNA sequences yielded few Guianese fish taxa assignments, and some of the eDNA sequences also matched to non-fish taxa. Although the teleost primers were designed to amplify Teleostei DNA, they may also amplify non-target taxa without the need of mismatches on the primers (Valentini *et al.*, 2016). We here extended reference databases of Neotropical fauna using 12S rRNA molecular marker for 114 new species. Although these species account for only 5% of the 4035 Neotropical freshwater fish species reported in Lévêque *et al.* (2007), they nevertheless account for a wide range of genera (18.6% of the 705 Neotropical freshwater fish genera) and families (60.8% of the 74 Neotropical freshwater fish families). As the species considered represent most of the major fish orders of the Neotropics, such reference database can thus be used in future metabarcoding fish inventories using family taxonomic resolution throughout the Neotropics. Indeed, using family taxonomic resolution, both metabarcoding and traditional methods detected a high proportion of families in common. In contrast, using genera besides of species did not improve the congruence between traditional and metabarcoding results because more than half of the genera considered are monospecific in French Guiana (see Table IV.S1). Metabarcoding fish inventories at finer taxonomic resolution (genus or species) over larger spatial areas (Guiana shield, Amazon River drainage or entire Neotropics) will nevertheless need complements on the reference database. We hence appeal forthcoming studies to complement our reference data on more species.

Despite the imperfect local species detection, the fish assemblages detected using metabarcoding were consistent with the fauna known to occur at higher spatial scales since only 5 out of the 132 species detected using metabarcoding were detected outside of their spatial distribution range. Three of these species probably account for the actual presence of the species in the considered river drainage, while they were never detected using traditional methods. Those species are indeed known either from adjacent river drainages (*Kribia aff. guianensis sp1* or *Satanoperca rhynchitis*; Le Bail *et al.*, 2000, 2012 or for having a large distribution in the Neotropics that encapsulates French Guiana. This is the case for *Corydoras aeneus* (Froese & Pauly, 2015) which presence in the Oyapock River drainage is therefore probable. Those last three species were probably erroneously assigned to closely related species due to the incompleteness of our reference database. For instance, *Ancistrus aff. temminckii* was detected out of its known range, in areas colonised by the closely related

*Ancistrus aff. hoplogenyis*. *A. aff. hoplogenyis* was not informed in our reference database, so sequences of *A. aff. hoplogenyis* were hence probably wrongly assigned to *A. aff. temminckii*, the most similar species in the reference database. Identically, *Hemiodus quadrimaculatus* was detected in the Maroni River drainage, instead of *Hemiodus huraulti*, a closely related species, that was not informed in the reference database.

Within river drainages, we adequately differentiated small stream fauna from large rivers using metabarcoding. Only three out of the 86 species detected by metabarcoding in small streams were only detected in rivers using traditional methods, but two (*Crenicichla multispinosa* and *Mastiglanis cf. asopos*) are known to occur in small streams (Keith *et al.*, 2000; Planquette *et al.*, 1996), although not found using traditional methods in our study. Only one occurrence of *Plagoscion squamosissimus* in a small stream is unexpected. In rivers, metabarcoding allowed to detect both stream and river fish fauna, due to the upstream-downstream drift of eDNA throughout the watercourse, as shown by Civade *et al.* (2016). Our river samples therefore encapsulate the fish fauna from the river together with the fish belonging to the nearby streams, making eDNA using metabarcoding approach less efficient than traditional methods to discriminate the fauna from large rivers to that of the nearby streams.

In contrast, the small stream fauna from distinct river drainages was better discriminated using metabarcoding than using traditional methods. This might be due to the integrative characteristic of eDNA using metabarcoding approach (Civade *et al.*, 2016) that, although incomplete, provides a more representative species list of the river drainage biodiversity than using traditional methods which provide a spatially localized inventory (dozens of metres in small rivers and hundreds of metres in large rivers). Traditional methods make sampling efficient only in a part of available habitats. For instance, large rivers are traditionally sampled using gill nets that are inefficient to capture small sized species known to occur both in streams and in the rapids of large rivers (Keith *et al.*, 2000; Le Bail *et al.*, 2000; Melki, 2016; Planquette *et al.*, 1996). Moreover, nets are hardly usable and their efficiency is limited in burden areas or rapids and hence they are unable to detect the species living only in those habitats. The same habitat types are sampled in all the sites Allard *et al.* (2016), making inter-drainage discrepancies less marked and hence under-estimating the faunistic distinctiveness between river drainages. Those two distinct perceptions of the fauna might explain why despite a significant correlation in fish richness and occurrence between metabarcoding and traditional methods, the species detected by the two methods markedly differ.

## CONCLUSION

Given the limitations of both traditional methods and metabarcoding, those two approaches provided distinct perceptions of fish assemblages. Therefore, each method captured a distinct fraction of the real fish assemblage. While traditional

methods provided an almost complete, but spatially limited picture of the fish species diversity (Allard, 2014), the metabarcoding provided a partial, but more spatially extended inventory. This makes the two approaches complementary to get a realistic picture of the biodiversity at the reach scale.

The results obtained using metabarcoding differed from those obtained in temperate environments, where metabarcoding provided an exhaustive picture and even an overrepresentation of the fish diversity (Hänfling *et al.*, 2016; Olds *et al.*, 2016; Valentini *et al.*, 2016). Protocols improvement in sampling, like increasing the sampling effort per site (sampling time length of sampling or number of filtration) might increase species detection (Hänfling *et al.*, 2016). On the other hand, the lack of reference sequences and bias in detection for abundant species or in amplification for abundant sequences (Adams *et al.*, 2013; Kelly *et al.*, 2014) might also decreased the number of species detected using eDNA, and those methodological limitations should be quantified in the future.

Despite those pitfalls and limitations, the eDNA metabarcoding is a promising approach for fish biodiversity assessment in tropical areas. Given the rare erroneous species detection, the significant correlations between fish diversity and occurrences for both traditional methods and eDNA metabarcoding, as well as the higher capacity of metabarcoding than traditional methods to discriminate between river drainages, it appears that metabarcoding can be used as a rough, but rapid biodiversity assessment method in the Neotropics. Turning eDNA metabarcoding into a more exhaustive inventory tool will need to expand the reference database and optimize field and laboratory protocols (Rees *et al.*, 2015; Roussel *et al.*, 2015). The eDNA metabarcoding method nevertheless deserves to be used since now as a complementary tool to traditional methods pending future developments of this methodology to make it more exhaustive.

The development of metabarcoding is challenging because it would overcome the current destructive inventory tools (e.g. rotenone, gillnets) that are currently banned from most countries for both ethical and legal reasons. For instance, in Europe, the use of rotenone has been regulated since 2008 (European laws 2008/296/CE and 2008/317/CE). Although few exceptional authorizations have been obtained to conduct scientific studies, its use has now completely been banned. Developing a new non-destructive sampling method would unlock the current situation where scientists and environmental managers can no longer achieve complete species inventories. The implementation of eDNA-based methods would therefore permit to continue collecting information on fish assemblages in tropical freshwaters, which is of particular importance given the current rise of anthropogenic disturbances, and associated decline of aquatic biodiversity on Neotropical ecosystems (Allard *et al.*, 2016; Hammond *et al.*, 2007; Winemiller *et al.*, 2016).

## **ACKNOWLEDGMENTS**

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## **CONFLICT OF INTEREST**

P.T. is inventors of a patent on 'teleo' primers and on the use of the amplified fragment for identifying amphibians and fish species from environmental samples. This patent only restricts commercial applications and has no impact on the use of this method by academic researchers. A.V. and T.D. are research scientists in a private company, specialized on the use of eDNA for species detection.

**Table IV.S1:** List of species found using eDNA metabarcoding and traditional methods. Occurrences are expressed as the percentage of sites where the species was found. For each species, we indicated the taxonomic unit at which the 12S rRNA metabarcoding fragment was identified. “-” indicates species absent from the reference database

<b>Species</b>	<b>Taxonomic detection level</b>	<b>Occurrence with eDNA metabarcoding</b>	<b>Occurrence with traditional methods</b>
<i>Acestrorhynchus falcatus</i> (Bloch, 1794)	Species	46.15	56.41
<i>Acestrorhynchus microlepis</i> (Müller & Troschel, 1844)	-	-	20.51
<i>Acnodon oligacanthus</i> (Müller & Troschel, 1844)	Species	2.56	2.56
<i>Aequidens tetramerus</i> (Heckel, 1840)	Species	25.64	10.25
<i>Ageneiosus inermis</i> (Linnaeus, 1766)	Species	2.56	15.38
<i>Ageneiosus ucayalenis</i> (Castelnau, 1855)	Species	0	5.13
<i>Anablepsoides holmiae</i> (Eigenmann, 1909)	Species	0	2.56
<i>Anablepsoides igneus</i> (Huber, 1991)	Species	28.21	25.64
<i>Anablepsoides lungi</i> (Berkenkamp, 1984)	Species	0	10.26
<i>Anchovia surinamensis</i> (Bleeker, 1865)	Species	0	2.56
<i>Ancistrus aff. hoplogenyis</i> (Günther, 1864)	-	-	10.26
<i>Ancistrus aff. temminckii</i> (Valenciennes, 1840)	Species	33.33	2.56
<i>Ancistrus cf. leucostictus</i> (Günther, 1864)	Species	28.21	23.08
<i>Anostomus brevior</i> (Géry, 1963)	Species	5.13	5.13
<i>Aphyocharacidium melandetum</i> (Eigenmann, 1912)	-	-	2.56
<i>Apistogramma gossei</i> (Kullander, 1982)	Species	12.82	5.13
<i>Apteronotus albifrons</i> (Linnaeus, 1766)	-	-	2.56
<i>Astyanax bimaculatus</i> (Linnaeus, 1758)	Species	0	7.69
<i>Astyanax validus</i> (Géry, Planquette & Le Bail, 1991)	Species	10.26	10.26
<i>Auchenipterus dentatus</i> (Valenciennes, 1840)	-	-	2.56

Table IV.S1 continued

<i>Auchenipterus nuchalis</i> (Spix & Agassiz, 1829)	Species	0	7.69
<i>Batrochoglanis raninus</i> (Valenciennes, 1840)	Species	35.90	43.59
<i>Bivibranchia bimaculata</i> (Vari, 1985)	Species	2.56	10.26
<i>Brachyhypopomus beebei</i> (Schultz, 1944)	Species	2.56	5.13
<i>Brachyplatystoma vaillantii</i> (Valenciennes, 1840)	Species	0	2.56
<i>Brycon falcatus</i> (Müller & Troschel, 1844)	Species	2.56	7.69
<i>Brycon pesu</i> (Müller & Troschel, 1845)	Species	2.56	7.69
<i>Bryconamericus aff. hyphesson</i>	Species	0	2.56
<i>Bryconamericus guyanensis</i> (Zarske, Le Bail & Géry, 2010)	Species	43.59	41.03
<i>Bryconops aff. caudomaculatus</i> (Günther, 1864)	Genus	Bryconops	10.26
<i>Bryconops affinis</i> (Günther, 1864)	Genus	Bryconops	46.15
<i>Bryconops caudomaculatus</i> (Günther, 1864)	Genus	Bryconops	17.95
<i>Bryconops melanurus</i> (Bloch, 1794)	Genus	Bryconops	20.51
<i>Caenotropus maculosus</i> (Eigenmann, 1912)	Species	2.56	7.69
<i>Callichthys callichthys</i> (Linnaeus, 1758)	Species	25.64	2.56
<i>Cetopsidium orientale</i> (Vari, Ferraris & Keith, 2003)	Species	15.38	12.82
<i>Chalceus macrolepidotus</i> (Cuvier, 1818)	Species	2.56	7.69
<i>Characidium zebra</i> (Eigenmann, 1909)	Species	28.21	48.72
<i>Charax gibbosus</i> (Linnaeus, 1758)	Species	2.56	10.26
<i>Chasmocranus brevior</i> (Eigenmann, 1912)	Species	10.26	7.69
<i>Chasmocranus longior</i> (Eigenmann, 1912)	Species	41.03	17.95
<i>Chilodus zunevei</i> (Puyo, 1946)	Species	5.13	5.13
<i>Cichla ocellaris</i> (Bloch & Schneider, 1801)	Species	5.13	5.13
<i>Cichlasoma bimaculatum</i> (Linnaeus, 1758)	Species	5.13	0
<i>Cleithracara maronii</i> (Steindachner, 1881)	Species	23.08	12.82
<i>Copella arnoldi</i> (Regan, 1912)	Species	2.56	0

Table IV.S1 continued

<i>Copella carsevennensis</i> (Regan, 1912)	-	-	48.72
<i>Corydoras aeneus</i> (Gill, 1858)	Species	7.69	0
<i>Corydoras aff. guianensis</i>	Species	5.13	5.13
<i>Corydoras amapaensis</i> (Nijssen, 1972)	Species	0	7.69
<i>Corydoras geoffroy</i> (Lacepède, 1803)	Species	12.82	10.26
<i>Corydoras solox</i> (Nijssen & Isbrücker, 1983)	Species	5.13	0
<i>Corydoras spilurus</i> (Norman, 1926)	Species	0	2.56
<i>Creagrutus melanzonus</i> (Eigenmann, 1909)	Species	5.13	0
<i>Creagrutus planquettei</i> (Géry & Renno, 1989)	Species	2.56	2.56
<i>Crenicichla albopunctata</i> (Pellegrin, 1904)	Species	12.82	15.38
<i>Crenicichla johanna</i> (Heckel, 1840)	Species	0	5.13
<i>Crenicichla multispinosa</i> (Pellegrin, 1903)	Species	5.13	2.56
<i>Crenicichla saxatilis</i> (Linnaeus, 1758)	Species	15.38	28.21
<i>Cteniloricaria platystoma</i> (Günther, 1868)	Species	2.56	0
<i>Curculionichthys sp</i>	Species	7.69	2.56
<i>Curimata cyprinoides</i> (Linnaeus, 1766)	Species	2.56	7.69
<i>Curimatopsis crypticus</i> (Vari, 1982)	Species	2.56	2.56
<i>Cynodon meionactis</i> (Géry, Le Bail & Keith, 1999)	Species	2.56	7.69
<i>Cynopotamus essequibensis</i> (Eigenmann, 1912)	-	-	10.26
<i>Cyphocharax aff. spilurus</i>	-	-	2.56
<i>Cyphocharax helleri</i> (Steindachner, 1910)	Species	20.51	12.82
<i>Cyphocharax spilurus</i> (Günther, 1864)	Species	10.26	15.38
<i>Doras carinatus</i> (Linnaeus, 1776)	Species	5.13	7.69
<i>Doras micropoeus</i> (Eigenmann, 1912)	Species	2.56	7.69
<i>Eigenmannia virescens</i> (Valenciennes, 1836)	Species	41.03	35.90
<i>Electrophorus electricus</i> (Linnaeus, 1766)	Species	23.08	5.13

Table IV.S1 continued

<i>Eleotris pisonis</i> (Gmelin, 1789)	Species	2.56	2.56
<i>Erythrinus erythrinus</i> (Bloch & Schneider, 1801)	Species	25.64	20.51
<i>Farlowella reticulata</i> (Boeseman, 1971)	Species	15.38	12.82
<i>Farlowella rugosa</i> (Boeseman, 1971)	-	-	2.56
<i>Gasteropelecus sternicla</i> (Linnaeus, 1758)	-	-	15.38
<i>Geophagus camopienis</i> (Pellegrin, 1903)	Species	0	2.56
<i>Geophagus harreri</i> (Gosse, 1976)	Species	10.26	7.69
<i>Geophagus surinamensis</i> (Bloch, 1791)	Species	7.69	10.26
<i>Glanidium leopardum</i> (Hoedeman, 1961)	Species	12.82	7.69
<i>Guianacara geayi</i> (Pellegrin, 1902)	Species	2.56	10.26
<i>Guianacara owroewefi</i> (Kullander & Nijssen, 1989)	Species	15.38	7.69
<i>Guyanancistrus brevispinis</i> (Heitmans, Nijssen & Isbrücker, 1983)	Species	10.26	5.13
<i>Gymnotus carapo</i> (Linnaeus, 1758)	Genus	Gymnotus	69.23
<i>Gymnotus coropinae</i> (Hoedeman, 1962)	Genus	Gymnotus	46.15
<i>Harttia guianensis</i> (Rapp Py-Daniel & Oliveira, 2001)	-	-	12.82
<i>Harttiella lucifer</i> (Covain & Fisch-Muller, 2012)	-	-	5.13
<i>Helogenes marmoratus</i> (Günther, 1863)	-	-	61.54
<i>Hemiancistrus medians</i> (Kner, 1854)	-	-	2.56
<i>Hemibrycon surinamensis</i> (Géry, 1962)	Species	15.38	15.38
<i>Hemigrammus boesemani</i> (Géry, 1959)	Species	0	5.13
<i>Hemigrammus guyanensis</i> (Géry, 1959)	Genus	Hemigrammus	2.56
<i>Hemigrammus ocellifer</i> (Steindachner, 1882)	Genus	Hemigrammus	23.08
<i>Hemigrammus ora</i> (Zarske, Le Bail & Géry, 2006)	Species	0	2.56
<i>Hemigrammus rodwayi</i> (Durbin, 1909)	Species	0	17.95
<i>Hemigrammus unilineatus</i> (Gill, 1858)	Species	5.13	35.90
<i>Hemiodus aff. unimaculatus</i>	Species	2.56	17.95

Table IV.S1 continued

<i>Hemiodus huraulti</i> (Géry, 1964)	Order	Characiformes	5.13
<i>Hemiodus quadrimaculatus</i> (Pellegrin, 1909)	Species	2.56	7.69
<i>Hemiodus unimaculatus</i> (Bloch, 1794)	Species	2.56	7.69
<i>Heptapterus bleekeri</i> (Boeseman, 1953)	Species	2.56	0
<i>Hoplerythrinus unitaeniatus</i> (Spix & Agassiz, 1829)	Species	30.77	5.13
<i>Hoplias aimara</i> (Valenciennes, 1847)	Species	46.15	25.64
<i>Hoplias malabaricus</i> (Bloch, 1794)	Species	69.23	17.95
<i>Hyphessobrycon borealis</i> (Zarske, Le Bail & Géry, 2006)	-	-	33.33
<i>Hyphessobrycon copelandi</i> (Durbin, 1908)	Species	0	2.56
<i>Hyphessobrycon roseus</i> (Géry, 1960)	Species	0	5.13
<i>Hyphessobrycon simulatus</i> (Géry, 1960)	Species	0	7.69
<i>Hypomasticus despaxi</i> (Puyo, 1943)	Species	10.26	7.69
<i>Hypopomus artedi</i> (Kaup, 1856)	Species	46.15	23.08
<i>Hypopygus lepturus</i> (Hoedeman, 1962)	Family	Hypopomidae	5.13
<i>Hypostomus gymnorhynchus</i> (Norman, 1926)	Species	10.26	12.82
<i>Hypostomus plecostomus</i> (Linnaeus, 1758)	-	-	2.56
<i>Imparfinis pijpersi</i> (Hoedeman, 1961)	Species	2.56	0
<i>Ituglanis amazonicus</i> (Steindachner, 1882)	Species	35.90	10.26
<i>Ituglanis nebulosus</i> (de Pinna & Keith, 2003)	Species	25.64	17.95
<i>Japigny kirschbaum</i> (Meunier, Jégu & Keith, 2011)	Species	12.82	7.69
<i>Jupiaba abramoides</i> (Eigenmann, 1909)	-	-	38.46
<i>Jupiaba keithi</i> (Géry, Planquette & Le Bail, 1996)	Species	7.69	7.69
<i>Jupiaba maroniensis</i> (Géry, Planquette & Le Bail, 1996)	Species	0	2.56
<i>Jupiaba meunieri</i> (Géry, Planquette & Le Bail, 1996)	-	-	2.56
<i>Krobia aff. guianensis sp1</i>	Species	33.33	10.26
<i>Krobia aff. guianensis sp2</i>	Species	7.69	17.95

Table IV.S1 continued

<i>Krobia itanyi</i> (Puyo, 1943)	Species	33.33	15.38
<i>Laimosemion aff. geayi</i> (Mol et al., 2012)	Species	2.56	0
<i>Laimosemion agilae</i> (Hoedeman, 1954)	Species	2.56	30.77
<i>Laimosemion cf. geayi</i>	Species	0	2.56
<i>Laimosemion cladophorus</i> (Huber, 1991)	Species	0	2.56
<i>Laimosemion geayi</i> (Vaillant, 1899)	-	-	17.95
<i>Laimosemion xiphidius</i> (Huber, 1979)	-	-	23.08
<i>Leporinus acutidens</i> (Valenciennes, 1837)	Genus	Leporinus	5.13
<i>Leporinus fasciatus</i> (Bloch, 1794)	Species	7.69	10.26
<i>Leporinus friderici</i> (Bloch, 1794)	Species	12.82	12.82
<i>Leporinus gossei</i> (Géry, Planquette & Le Bail, 1991)	Genus	Leporinus	2.56
<i>Leporinus granti</i> (Eigenmann, 1912)	Genus	Leporinus	12.82
<i>Leporinus leballi</i> (Géry & Planquette, 1983)	Species	10.26	10.26
<i>Leporinus maculatus</i> (Müller & Isbrücker, 1993)	Genus	Leporinus	2.56
<i>Leporinus melanostictus</i> (Norman, 1926)	Genus	Leporinus	2.56
<i>Leporinus nijsseni</i> (Garavello, 1990)	Genus	Leporinus	2.56
<i>Lithoxus boujardi</i> (Muller & Isbrücker, 1993)	Species	2.56	2.56
<i>Lithoxus planquettei</i> (Boeseman, 1982)	Species	10.26	28.21
<i>Lithoxus stocki</i> (Nijssen & Isbrücker, 1990)	Species	0	5.13
<i>Loricaria cataphracta</i> (Linnaeus, 1758)	Species	0	7.69
<i>Lycengraulis batesii</i> (Günther, 1868)	Species	0	5.13
<i>Mastiglanis cf. asopos</i> (Bockmann, 1994)	Species	2.56	2.56
<i>Megalechis thoracata</i> (Valenciennes, 1840)	-	-	7.69
<i>Melanocharacidium blennioides</i> (Eigenmann, 1909)	Species	2.56	5.13
<i>Melanocharacidium dispilomma</i> (Buckup, 1993)	Species	2.56	5.13
<i>Metaloricaria paucidens</i> (Isbrücker, 1975)	Species	2.56	5.13

Table IV.S1 continued

<i>Metynnis lippincottianus</i> (Cope, 1870)	-	-	2.56
<i>Microcharacidium eleotrioides</i> (Géry, 1960)	Species	35.90	15.38
<i>Moenkhausia aff. grandisquamis</i>	Genus	Moenkhausia	7.69
<i>Moenkhausia aff. intermedia</i>	Species	5.13	2.56
<i>Moenkhausia chrysargyrea</i> (Günther, 1864)	Genus	Moenkhausia	38.46
<i>Moenkhausia collettii</i> (Steindachner, 1882)	Species	5.13	28.21
<i>Moenkhausia georgiae</i> (Géry, 1965)	Species	12.82	7.69
<i>Moenkhausia grandisquamis</i> (Müller & Troschel, 1845)	Genus	Moenkhausia	12.82
<i>Moenkhausia hemigrammoides</i> (Géry, 1965)	Species	0	10.26
<i>Moenkhausia moisae</i> (Géry, Planquette & Le Bail, 1995)	Genus	Moenkhausia	23.08
<i>Moenkhausia oligolepis</i> (Günther, 1864)	Species	25.64	41.03
<i>Moenkhausia surinamensis</i> (Géry, 1965)	Genus	Moenkhausia	15.38
<i>Myloplus rhomboidalis</i> (Cuvier, 1818)	Species	2.56	7.69
<i>Myloplus rubripinnis</i> (Müller & Troschel, 1844)	-	-	12.82
<i>Myloplus ternetzi</i> (Norman, 1929)	Species	30.77	20.51
<i>Nannacara aureocephalus</i> (Allgayer, 1983)	Species	35.90	38.46
<i>Nannostomus beckfordi</i> (Günther, 1872)	Species	2.56	5.13
<i>Nannostomus bifasciatus</i> (Hoedeman, 1954)	Species	12.82	15.38
<i>Ochmacanthus cf. alternus</i> (Myers, 1927)	-	-	2.56
<i>Ochmacanthus reinhardtii</i> (Steindachner, 1882)	Species	0	2.56
<i>Otocinclus mariae</i> (Fowler, 1940)	Species	5.13	2.56
<i>Pachypops fourcroyi</i> (Lacepède, 1802)	Species	2.56	7.69
<i>Parodon guyanensis</i> (Géry, 1959)	Species	2.56	5.13
<i>Phenacogaster wayana</i> (Le Bail & Lucena, 2010)	Species	0	10.26
<i>Phenacorhamdia tenuis</i> (Mees, 1986)	Species	7.69	7.69
<i>Piabucus dentatus</i> (Koelreuter, 1763)	Species	0	5.13

Table IV.S1 continued

<i>Pimelabditus moli</i> (Parisi & Lundberg, 2009)	Species	2.56	2.56
<i>Pimelodella cristata</i> (Müller & Troschel, 1849)	Genus	Pimelodella	33.33
<i>Pimelodella geryi</i> (Hoedman, 1961)	Genus	Pimelodella	10.26
<i>Pimelodella procera</i> (Mees, 1983)	Species	7.69	10.26
<i>Pimelodus blochii</i> (Valenciennes, 1840)	-	-	2.56
<i>Pimelodus ornatus</i> (Kner, 1858)	Species	2.56	7.69
<i>Plagioscion auratus</i> (Castelnau, 1855)	Species	10.26	7.69
<i>Platydoras costatus</i> (Linnaeus, 1758)	Species	2.56	2.56
<i>Polycentrus schomburgkii</i> (Müller & Troschel, 1849)	Species	15.38	5.13
<i>Poptella brevispina</i> (Reis, 1989)	Species	10.26	43.59
<i>Potamorrhaphis guianensis</i> (Jardine, 1843)	Species	0	5.13
<i>Potamotrygon orbignyi</i> (Castelnau, 1855)	Species	7.69	5.13
<i>Pristella maxillaris</i> (Ulrey, 1894)	Species	0	5.13
<i>Pristobrycon striolatus</i> (Steindachner, 1908)	Species	2.56	2.56
<i>Prochilodus rubrotaeniatus</i> (Jardine, 1841)	Species	2.56	7.69
<i>Pseudancistrus barbatus</i> (Valenciennes, 1840)	Species	7.69	7.69
<i>Pseudoplatystoma fasciatum</i> (Linnaeus, 1766)	Species	7.69	0
<i>Pterengraulis atherinoides</i> (Linnaeus, 1766)	Species	2.56	0
<i>Pyrrhulina filamentosa</i> (Valenciennes, 1847)	Species	56.41	56.41
<i>Rhamdia quelen</i> (Quoy & Gaimard, 1824)	Species	2.56	28.21
<i>Rhamphichthys rostratus</i> (Linnaeus, 1766)	Species	0	5.13
<i>Rineloricaria nsp1 aff. stewarti</i>	Species	23.08	15.38
<i>Rineloricaria platyura</i> (Müller & Troschel, 1849)	Species	2.56	0
<i>Roeboexodon geryi</i> (Myers, 1960)	Species	2.56	10.26
<i>Satanoperca rhynchitis</i> (Kullander, 2012)	Species	7.69	5.13
<i>Schizodon fasciatus</i> (Spix & Agassiz, 1829)	Order	Characiformes	10.26

Table IV.S1 continued

<i>Semaprochilodus varii</i> (Castro, 1988)	Family	Prochilodontidae	5.13
<i>Serrapinnus gracilis</i> (Géry, 1690)	Family	Characidae	5.13
<i>Serrasalmus eigenmanni</i> (Norman, 1929 )	Species	2.56	12.82
<i>Serrasalmus rhombeus</i> (Linnaeus, 1766)	-	-	12.82
<i>Steindachnerina varii</i> (Géry, Planquette & Le Bail, 1991)	Species	12.82	0
<i>Sternopygus macrurus</i> (Bloch & Schneider, 1801)	Species	48.72	46.15
<i>Synbranchus marmoratus</i> (Bloch, 1795)	Species	61.54	7.69
<i>Tatia brunnea</i> (Mees, 1974)	Species	15.38	2.56
<i>Tatia intermedia</i> (Steindachner, 1877)	Species	10.26	7.69
<i>Tetragonopterus chalceus</i> (Spix & Agassiz, 1829)	Species	2.56	12.82
<i>Tetragonopterus rarus</i> (Zarske, Géry & Isbrücker, 2004)	Species	10.26	2.56
<i>Thayeria ifati</i> (Géry, 1959)	Species	2.56	10.26
<i>Trachelyopterus galeatus</i> (Linnaeus, 1766)	Species	2.56	5.13
<i>Triportheus brachipomus</i> (Valenciennes, 1850)	-	-	12.82

**Table III.S2** Number of reads obtained from the NGS runs per sample before and after bioinformatic filtering

Site name	Watershed	Watercourse type	Number of reads per sample
Control			7,870
			357,436
			2,039
			23,943
			1,033,352
Crique parare 5	Approuague	Stream	209,312
Crique parare 8	Approuague	Stream	461,007
Crique parare 2	Approuague	Stream	44,967
Pikin Tabiki	Maroni	River	1,565,695
Saut Sonnelle	Maroni	River	852,552
Papaichton	Maroni	River	1,235,661
Saut Bief	Comte	River	677,714
Machicou	Approuague	River	1,954,347
Athanase	Approuague	River	2,031,480
Twenke	Maroni	River	1,346,169
Apsik Icholi	Maroni	River	442,803
Crique Marie	Oyapock	Stream	803,477
Crique SM	Oyapock	Stream	441,709
Crique Minette	Oyapock	Stream	470,174
Crique Pied Saut	Oyapock	Stream	451,058
Crique Kapiri 2	Approuague	Stream	98,842
Crique Kapiri 1	Approuague	Stream	96,132
Crique Kapiri 6	Approuague	Stream	145,703
Crique Petit Approuague	Comte	Stream	145,206
Crique Humus	Kourou	Stream	134,347
Crique Eau Claire	Kourou	Stream	124,136
Crique Paracou	Sinnamary	Stream	116,440
Crique Toussaint	Sinnamary	Stream	107,175
Crique Organobo	Organobo	Stream	261,145
Crique Petit laussat Aval	Mana	Stream	304,807
Crique Voltalia 2	Mana	Stream	259,417
Crique Voltalia 3	Mana	Stream	382,708
Crique Voltalia 4	Mana	Stream	250,053
Crique des Cascades	Maroni	Stream	878,385
Apa	Maroni	Stream	126,689
Crique Bastien	Maroni	Stream	106,933
Crique Penta	Maroni	Stream	91,667
Crique Nouvelle France 1	Maroni	Stream	144,262
Crique Nouvelle France 2	Maroni	Stream	123,139
Crique Nouvelle France 6	Maroni	Stream	175,609
Crique Nouvelle France 5	Maroni	Stream	161,476
Crique Nouvelle France 4	Maroni	Stream	125,194
Crique Nouvelle France 3	Maroni	Stream	303,219
Crique à l'Est	Mana	Stream	124,856
<b>Total</b>			<b>17,775,665</b>
Percentage of the initial number of reads			100

Site name	Number of reads per sample with an occurrence > 10 and a length > 20 bp	Number of non internal reads per sample (obiclean)
Control	6,867	6,775
	161,434	142,345
	0	0
	23,095	21,764
	255,980	226,523
Crique parare 5	193,738	191,662
Crique parare 8	444,353	433,617
Crique parare 2	43,272	43,046
Pikin Tabiki	1,297,118	1,163,918
Saut Sonnelle	427,764	383,321
Papaichton	744,182	645,537
Saut Bief	595,816	572,171
Machicou	1,584,607	1,392,239
Athanase	1,246,918	1,118,966
Twenke	924,817	833,361
Apsik Icholi	382,995	377,733
Crique Marie	781,847	763,115
Crique SM	430,751	422,454
Crique Minette	458,874	446,353
Crique Pied Saut	441,135	430,122
Crique Kapiri 2	88,579	88,187
Crique Kapiri 1	87,573	86,935
Crique Kapiri 6	132,328	130,847
Crique Petit Approuague	130,677	130,004
Crique Humus	122,502	121,979
Crique Eau Claire	113,437	112,633
Crique Paracou	107,621	106,219
Crique Toussaint	97,072	96,506
Crique Organabo	245,412	238,741
Crique Petit laussat Aval	283,906	276,842
Crique Voltalia 2	244,628	233,249
Crique Voltalia 3	368,328	355,762
Crique Voltalia 4	230,714	224,726
Crique des Cascades	843,037	819,217
Apa	113,730	113,217
Crique Bastien	96,177	95,055
Crique Penta	82,231	81,858
Crique Nouvelle France 1	130,206	129,229
Crique Nouvelle France 2	109,505	109,139
Crique Nouvelle France 6	159,009	157,235
Crique Nouvelle France 5	144,998	143,798
Crique Nouvelle France 4	112,408	111,985
Crique Nouvelle France 3	273,455	272,158
Crique à l'Est	112,323	110,986
<b>Total</b>	<b>14,428,043</b>	<b>13,564,122</b>
Percentage of the initial number of reads	81.2	76.3

Site name	Number of reads with an identity $\geq$ 0.98 with the reference database	
	GenBank	Guyana fish
Control	6,775	0
	142,281	0
	0	0
	21,516	0
	224,975	30
Crique parare 5	18,980	129,546
Crique parare 8	0	288,050
Crique parare 2	1,406	42,587
Pikin Tabiki	235,833	870,235
Saut Sonnelle	135,301	245,896
Papaichton	221,837	252,025
Saut Bief	95,681	356,512
Machicou	32,703	599,182
Athanase	296,928	281,453
Twenke	136,037	488,319
Apsik Icholi	64,888	278,497
Crique Marie	85,177	601,195
Crique SM	11,685	349,292
Crique Minette	16	396,781
Crique Pied Saut	10,413	296,396
Crique Kapiri 2	15,149	56,315
Crique Kapiri 1	13,123	52,373
Crique Kapiri 6	11,318	83,442
Crique Petit Approuague	23,125	76,364
Crique Humus	20,259	75,113
Crique Eau Claire	18,110	61,752
Crique Paracou	20,609	53,752
Crique Toussaint	14,083	56,092
Crique Organabo	108,878	38,537
Crique Petit laussat Aval	22,765	135,767
Crique Voltalia 2	7,634	136,534
Crique Voltalia 3	43,364	151,289
Crique Voltalia 4	39,632	142,318
Crique des Cascades	88,671	607,259
Apa	13,796	75,131
Crique Bastien	22,312	47,675
Crique Penta	10,616	51,866
Crique Nouvelle France 1	15,907	112,348
Crique Nouvelle France 2	17,583	92,319
Crique Nouvelle France 6	31,661	71,505
Crique Nouvelle France 5	22,421	96,060
Crique Nouvelle France 4	17,991	75,643
Crique Nouvelle France 3	36,508	213,130
Crique à l'Est	33,268	70,149
<b>Total</b>	<b>2,015,668</b>	<b>8,108,699</b>
Percentage of the initial number of reads	11.3	45.6

Site name	Number of reads with an with a frequency of occurrence > 0.0003 per taxon per sample and per sequencing run	
	GenBank	Guyana fish
Control	6,775	0
	142,281	0
	0	0
	21,516	0
	224,975	0
Crique parare 5	18,980	129,546
Crique parare 8	0	288,050
Crique parare 2	1,406	42,587
Pikin Tabiki	235,833	870,235
Saut Sonnelle	135,301	245,896
Papaichton	221,837	252,025
Saut Bief	95,681	356,512
Machicou	32,689	599,182
Athanase	296,917	281,453
Twenke	136,037	488,319
Apsik Icholi	64,877	278,497
Crique Marie	85,177	601,068
Crique SM	11,685	349,260
Crique Minette	16	396,781
Crique Pied Saut	10,413	296,396
Crique Kapiri 2	15,149	56,315
Crique Kapiri 1	13,123	52,373
Crique Kapiri 6	11,318	83,442
Crique Petit Approuague	23,125	76,330
Crique Humus	20,259	75,113
Crique Eau Claire	18,110	61,752
Crique Paracou	20,609	53,752
Crique Toussaint	14,083	56,092
Crique Organabo	108,878	38,537
Crique Petit laussat Aval	22,765	135,767
Crique Voltalia 2	7,634	136,534
Crique Voltalia 3	43,364	151,289
Crique Voltalia 4	39,632	142,318
Crique des Cascades	88,671	607,259
Apa	13,796	75,131
Crique Bastien	22,293	47,675
Crique Penta	10,616	51,866
Crique Nouvelle France 1	15,890	112,348
Crique Nouvelle France 2	17,583	92,305
Crique Nouvelle France 6	31,661	71,505
Crique Nouvelle France 5	22,421	96,060
Crique Nouvelle France 4	17,991	75,643
Crique Nouvelle France 3	36,508	213,130
Crique à l'Est	33,268	70,149
<b>Total</b>	<b>2,015,596</b>	<b>8,108,492</b>
Percentage of the initial number of reads	11.3	45.6

**Table III.S3** Results from assignment using GenBank nucleotide database

Taxonomic group	Order	Family	Genus	Taxon†
Amphibia	Anura	Bufonidae	Atelopus	Atelopus
			Atelopus flavescens	
			Rhinella	Rhinella
			Rhinella dapsilis	
		Dendrobatidae	Anomaloglossus	Anomaloglossus baeobatrachus
		Hylidae	Hyla	Hyla helenae
			Hypsiboas	Hypsiboas boans
				Hypsiboas geographicus
			Hylinae	
			Osteocephalus	Osteocephalus cannatellai
		Osteocephalus oophagus		
Osteocephalus taurinus				
Leptodactylidae	Lithodytes	Lithodytes lineatus		
Ranidae	Rana	Rana palmipes		
Aves	Galliformes	Cracidae	Crax	Crax rubra
	Passeriformes	Tyrannidae	Mionectes	Mionectes oleagineus
Bacteria	Thermales	Thermaceae	Meiothermus	Meiothermus silvanus DSM 9946
"Fish"	Characiformes	Anostomidae	Leporinus	<b>Leporinus affinis</b>
		Chilodontidae	Chilodus	<b>Chilodus punctatus</b>
		Erythrinidae	Hoplias	<b>Hoplias malabaricus</b>
		Prochilodontidae	Prochilodus	<b>Prochilodus</b>
	Clupeiformes	Engraulidae	Pterengraulis	<b>Pterengraulis atherinoides</b>
	Gymnotiformes	Gymnotidae	Electrophorus	<b>Electrophorus electricus</b>
			Gymnotus	<b>Gymnotus carapo</b>
		Sternopygidae	Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>
	Myliobatiformes	Potamotrygonidae	Potamotrygon	<i>Potamotrygon motoro</i>
	Siluriformes	Auchenipteridae	Auchenipterus	<i>Auchenipterus demerarae</i>
			Tatia	<b>Tatia intermedia</b>
		Doradidae	Doras	<b>Doras micropoeus</b>
		Loricariidae	Ancistrus	<b>Ancistrus</b>
				<i>Ancistrus sp. 2a Parente, 2015</i>
			Cteniloricaria	<b>Cteniloricaria platystoma</b>
			Farlowella	<b>Farlowella reticulata</b>
			Harttia	<b>Harttia guianensis</b>
			Hartiella	<b>Hartiella lucifer</b>
			Hemiancistrus	<b>Hemiancistrus medians</b>
			Hypostomus	<b>Hypostomus gymnorhynchus</b>
			Lithoxus	<b>Lithoxus planquettei</b>
			Metaloricaria	<b>Metaloricaria paucidens</b>
			Panaqolus	<b>Panaqolus koko</b>
			Pseudancistrus	<b>Pseudancistrus</b>
		<b>Pseudancistrus barbatus</b>		
	Rineloricaria	<b>Rineloricaria</b>		
	Pimelodidae	Pimelabditus	<b>Pimelabditus moli</b>	
Pimelodus		<b>Pimelodus ornatus</b>		

Taxonomic group	Order	Family	Genus	Taxon†
"Fish"	Siluriformes	Pimelodidae	Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>
	Perciformes	Polycentridae	Polycentrus	<b>Polycentrus schomburgkii</b>
Insecta	Odonata	Libellulidae		Libellulidae
Mammalia	Didelphimorphia	Didelphidae	Caluromys	Caluromys philander
	Primates	Cebidae	Cebus	Cebus albifrons
		Hominidae	Homo	Homo
				Homo sapiens
				Homininae
Sauria	Squamata	Iguanidae	Polychrus	Polychrus marmoratus
	Testudines	Geoemydidae	Rhinoclemmys	Rhinoclemmys
<b>Total</b>				

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site		
		Crique parare 5	Crique parare 8	Crique parare 2
Atelopus	Atelopus	12	0	0
	Atelopus flavescens	21	0	0
Rhinella	Rhinella	0	0	0
	Rhinella dapsilis	0	0	0
Anomaloglossus	Anomaloglossus baeobatrachus	0	0	0
Hyla	Hyla helenae	0	0	0
Hypsiboas	Hypsiboas boans	0	0	0
	Hypsiboas geographicus	0	0	0
	Hylinae	0	0	0
Osteocephalus	Osteocephalus cannatellai	0	0	0
	Osteocephalus oophagus	0	0	0
	Osteocephalus taurinus	0	0	0
Lithodytes	Lithodytes lineatus	0	0	0
Rana	Rana palmipes	0	0	0
Crax	Crax rubra	0	0	0
Mionectes	Mionectes oleagineus	0	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0	0
Leporinus	<b>Leporinus affinis</b>	0	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0	0
Hoplias	<b>Hoplias malabaricus</b>	17,998	0	1,406
Prochilodus	<b>Prochilodus</b>	0	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0	0
Electrophorus	<b>Electrophorus electricus</b>	0	0	0
Gymnotus	<b>Gymnotus carapo</b>	543	0	0
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0	0	0
Potamotrygon	<i>Potamotrygon motoro</i>	0	0	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0	0
Tatia	<b>Tatia intermedia</b>	0	0	0
Doras	<b>Doras micropoeus</b>	0	0	0
Ancistrus	<b>Ancistrus</b>	0	0	0
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	0	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0	0
Farlowella	<b>Farlowella reticulata</b>	0	0	0
Harttia	<b>Harttia guianensis</b>	0	0	0
Hartiella	<b>Hartiella lucifer</b>	0	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	0	0
Lithoxus	<b>Lithoxus planquettei</b>	0	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0	0
Pseudancistrus	<b>Pseudancistrus</b>	0	0	0
	<b>Pseudancistrus barbatus</b>	0	0	0
Rineloricaria	<b>Rineloricaria</b>	0	0	0
Pimelabditus	<b>Pimelabditus moli</b>	0	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0	0

Genus	Taxon†	Site		
		Crique parare 5	Crique parare 8	Crique parare 2
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0	0
	Libellulidae	0	0	0
Caluromys	Caluromys philander	0	0	0
Cebus	Cebus albifrons	176	0	0
Homo	Homo	230	0	0
	Homo sapiens	0	0	0
	Homininae	0	0	0
Polychrus	Polychrus marmoratus	0	0	0
Rhinoclemmys	Rhinoclemmys	0	0	0
<b>Total</b>		<b>18,980</b>	<b>0</b>	<b>1,406</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site		
		Crique Marie	Crique SM	Crique Minette
Atelopus	Atelopus	0	0	0
	Atelopus flavescens	0	0	0
Rhinella	Rhinella	0	0	0
	Rhinella dapsilis	0	0	0
Anomaloglossus	Anomaloglossus baeobatrachus	0	0	0
Hyla	Hyla helenae	33	0	0
Hypsiboas	Hypsiboas boans	0	0	0
	Hypsiboas geographicus	0	0	0
	Hylinae	0	0	0
Osteocephalus	Osteocephalus cannatellai	0	0	0
	Osteocephalus oophagus	0	0	0
	Osteocephalus taurinus	0	0	0
Lithodytes	Lithodytes lineatus	0	0	0
Rana	Rana palmipes	0	0	0
Crax	Crax rubra	0	0	0
Mionectes	Mionectes oleagineus	0	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0	0
Leporinus	<b>Leporinus affinis</b>	0	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0	0
Hoplias	<b>Hoplias malabaricus</b>	45,004	11,685	0
Prochilodus	<b>Prochilodus</b>	0	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0	0
Electrophorus	<b>Electrophorus electricus</b>	40,116	0	0
Gymnotus	<b>Gymnotus carapo</b>	24	0	16
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0	0	0
Potamotrygon	<i>Potamotrygon motoro</i>	0	0	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0	0
Tatia	<b>Tatia intermedia</b>	0	0	0
Doras	<b>Doras micropoeus</b>	0	0	0
Ancistrus	<b>Ancistrus</b>	0	0	0
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	0	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0	0
Farlowella	<b>Farlowella reticulata</b>	0	0	0
Harttia	<b>Harttia guianensis</b>	0	0	0
Hartiella	<b>Hartiella lucifer</b>	0	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	0	0
Lithoxus	<b>Lithoxus planquettei</b>	0	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0	0
Pseudancistrus	<b>Pseudancistrus</b>	0	0	0
	<b>Pseudancistrus barbatus</b>	0	0	0
Rineloricaria	<b>Rineloricaria</b>	0	0	0
Pimelabditus	<b>Pimelabditus moli</b>	0	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0	0

Genus	Taxon†	Site		
		Crique Marie	Crique SM	Crique Minette
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0	0
	Libellulidae	0	0	0
Caluromys	Caluromys philander	0	0	0
Cebus	Cebus albifrons	0	0	0
Homo	Homo	0	0	0
	Homo sapiens	0	0	0
	Homininae	0	0	0
Polychrus	Polychrus marmoratus	0	0	0
Rhinoclemmys	Rhinoclemmys	0	0	0
<b>Total</b>		<b>85,177</b>	<b>11,685</b>	<b>16</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site		
		Crique Pied Saut	Crique Kapiri 2	Crique Kapiri 1
Atelopus	Atelopus	0	0	0
	Atelopus flavescens	0	0	0
Rhinella	Rhinella	0	9,608	0
	Rhinella dapsilis	0	71	759
Anomaloglossus	Anomaloglossus baebatrachus	0	0	0
Hyla	Hyla helenae	0	16	467
Hypsiboas	Hypsiboas boans	0	29	839
	Hypsiboas geographicus	0	0	0
	Hylinae	0	0	233
Osteocephalus	Osteocephalus cannatellai	0	0	0
	Osteocephalus oophagus	0	0	0
	Osteocephalus taurinus	0	0	0
Lithodytes	Lithodytes lineatus	0	0	0
Rana	Rana palmipes	0	0	0
Crax	Crax rubra	0	0	0
Mionectes	Mionectes oleagineus	0	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0	0
Leporinus	<b>Leporinus affinis</b>	0	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0	0
Hoplias	<b>Hoplias malabaricus</b>	951	1,534	8,561
Prochilodus	<b>Prochilodus</b>	0	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0	0
Electrophorus	<b>Electrophorus electricus</b>	0	1,446	0
Gymnotus	<b>Gymnotus carapo</b>	9,462	181	1,637
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0	0	0
Potamotrygon	<i>Potamotrygon motoro</i>	0	556	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0	0
Tatia	<b>Tatia intermedia</b>	0	183	0
Doras	<b>Doras micropoeus</b>	0	0	0
Ancistrus	<b>Ancistrus</b>	0	0	0
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	805	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0	0
Farlowella	<b>Farlowella reticulata</b>	0	0	0
Harttia	<b>Harttia guianensis</b>	0	0	0
Hartiella	<b>Hartiella lucifer</b>	0	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	0	0
Lithoxus	<b>Lithoxus planquettei</b>	0	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0	0
Pseudancistrus	<b>Pseudancistrus</b>	0	0	0
	<b>Pseudancistrus barbatus</b>	0	0	0
Rineloricaria	<b>Rineloricaria</b>	0	720	122
Pimelabditus	<b>Pimelabditus moli</b>	0	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0	0

Genus	Taxon†	Site		
		Crique Pied Saut	Crique Kapiri 2	Crique Kapiri 1
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0	0
	Libellulidae	0	0	0
Caluromys	Caluromys philander	0	0	0
Cebus	Cebus albifrons	0	0	0
Homo	Homo	0	0	505
	Homo sapiens	0	0	0
	Homininae	0	0	0
Polychrus	Polychrus marmoratus	0	0	0
Rhinoclemmys	Rhinoclemmys	0	0	0
<b>Total</b>		<b>10,413</b>	<b>15,149</b>	<b>13,123</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site	
		Crique Kapiri 6	Crique Petit Approuague
Atelopus	Atelopus	0	0
	Atelopus flavescens	0	0
Rhinella	Rhinella	0	2,336
	Rhinella dapsilis	119	520
Anomaloglossus	Anomaloglossus baeobatrachus	0	0
Hyla	Hyla helenae	29	69
Hypsiboas	Hypsiboas boans	0	255
	Hypsiboas geographicus	0	0
	Hylinae	0	0
Osteocephalus	Osteocephalus cannatai	0	0
	Osteocephalus oophagus	0	0
	Osteocephalus taurinus	0	0
Lithodytes	Lithodytes lineatus	0	0
Rana	Rana palmipes	0	0
Crax	Crax rubra	0	0
Mionectes	Mionectes oleagineus	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0
Leporinus	<b>Leporinus affinis</b>	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0
Hoplias	<b>Hoplias malabaricus</b>	8,297	14,623
Prochilodus	<b>Prochilodus</b>	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0
Electrophorus	<b>Electrophorus electricus</b>	823	168
Gymnotus	<b>Gymnotus carapo</b>	1,554	491
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0	0
Potamotrygon	<i>Potamotrygon motoro</i>	0	106
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0
Tatia	<b>Tatia intermedia</b>	197	512
Doras	<b>Doras micropoeus</b>	0	0
Ancistrus	<b>Ancistrus</b>	0	0
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	3,764
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0
Farlowella	<b>Farlowella reticulata</b>	0	0
Harttia	<b>Harttia guianensis</b>	0	0
Hartiella	<b>Hartiella lucifer</b>	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	0
Lithoxus	<b>Lithoxus planquettei</b>	0	281
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0
Pseudancistrus	<b>Pseudancistrus</b>	0	0
	<b>Pseudancistrus barbatus</b>	0	0
Rineloricaria	<b>Rineloricaria</b>	0	0
Pimelabditus	<b>Pimelabditus moli</b>	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0

Genus	Taxon†	Site	
		Crique Kapiri 6	Crique Petit Approuague
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0
	Libellulidae	0	0
Caluromys	Caluromys philander	0	0
Cebus	Cebus albifrons	108	0
Homo	Homo	191	0
	Homo sapiens	0	0
	Homininae	0	0
Polychrus	Polychrus marmoratus	0	0
Rhinoclemmys	Rhinoclemmys	0	0
<b>Total</b>		<b>11,318</b>	<b>23,125</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site		
		Crique Humus	Crique Eau Claire	Crique Paracou
Atelopus	Atelopus	0	0	0
	Atelopus flavescens	0	0	0
Rhinella	Rhinella	0	110	0
	Rhinella dapsilis	1,087	6,530	7,161
Anomaloglossus	Anomaloglossus baeobatrachus	0	0	0
Hyla	Hyla helenae	0	1,369	0
Hypsiboas	Hypsiboas boans	0	164	365
	Hypsiboas geographicus	0	0	0
	Hylinae	0	52	0
Osteocephalus	Osteocephalus cannatellai	0	87	0
	Osteocephalus oophagus	0	0	0
	Osteocephalus taurinus	0	0	0
Lithodytes	Lithodytes lineatus	0	0	0
Rana	Rana palmipes	0	0	0
Crax	Crax rubra	0	0	0
Mionectes	Mionectes oleagineus	0	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0	0
Leporinus	<b>Leporinus affinis</b>	0	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0	0
Hoplias	<b>Hoplias malabaricus</b>	8,960	3,911	993
Prochilodus	<b>Prochilodus</b>	0	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0	0
Electrophorus	<b>Electrophorus electricus</b>	7,778	3,516	0
Gymnotus	<b>Gymnotus carapo</b>	2,251	858	6,040
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0	0	3,312
Potamotrygon	<i>Potamotrygon motoro</i>	0	0	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0	0
Tatia	<b>Tatia intermedia</b>	24	81	0
Doras	<b>Doras micropoeus</b>	0	0	0
Ancistrus	<b>Ancistrus</b>	0	0	0
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	1,289	2,578
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0	0
Farlowella	<b>Farlowella reticulata</b>	0	0	0
Harttia	<b>Harttia guianensis</b>	0	0	0
Hartiella	<b>Hartiella lucifer</b>	0	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	0	0
Lithoxus	<b>Lithoxus planquettei</b>	0	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0	0
Pseudancistrus	<b>Pseudancistrus</b>	0	0	0
	<b>Pseudancistrus barbatus</b>	0	0	0
Rineloricaria	<b>Rineloricaria</b>	0	0	0
Pimelabditus	<b>Pimelabditus moli</b>	0	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0	0

Genus	Taxon†	Site		
		Crique Humus	Crique Eau Claire	Crique Paracou
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0	0
	Libellulidae	0	0	0
Caluromys	Caluromys philander	0	0	0
Cebus	Cebus albifrons	0	0	0
Homo	Homo	159	143	160
	Homo sapiens	0	0	0
	Homininae	0	0	0
Polychrus	Polychrus marmoratus	0	0	0
Rhinoclemmys	Rhinoclemmys	0	0	0
<b>Total</b>		<b>20,259</b>	<b>18,110</b>	<b>20,609</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site	
		Crique Toussaint	Crique Organabo
Atelopus	Atelopus	0	0
	Atelopus flavescens	0	12
Rhinella	Rhinella	37	0
	Rhinella dapsilis	884	0
Anomaloglossus	Anomaloglossus baeobatrachus	0	0
Hyla	Hyla helenae	0	0
Hypsiboas	Hypsiboas boans	0	0
	Hypsiboas geographicus	0	0
	Hylinae	0	0
Osteocephalus	Osteocephalus cannattellai	0	0
	Osteocephalus oophagus	0	0
	Osteocephalus taurinus	0	0
Lithodytes	Lithodytes lineatus	0	0
Rana	Rana palmipes	0	0
Crax	Crax rubra	0	0
Mionectes	Mionectes oleagineus	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0
Leporinus	<b>Leporinus affinis</b>	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0
Hoplias	<b>Hoplias malabaricus</b>	8,867	0
Prochilodus	<b>Prochilodus</b>	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0
Electrophorus	<b>Electrophorus electricus</b>	0	0
Gymnotus	<b>Gymnotus carapo</b>	3,737	108,621
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	481	0
Potamotrygon	<i>Potamotrygon motoro</i>	0	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0
Tatia	<b>Tatia intermedia</b>	0	0
Doras	<b>Doras micropoeus</b>	0	0
Ancistrus	<b>Ancistrus</b>	0	0
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0
Farlowella	<b>Farlowella reticulata</b>	0	0
Harttia	<b>Harttia guianensis</b>	0	0
Hartiella	<b>Hartiella lucifer</b>	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	0
Lithoxus	<b>Lithoxus planquettei</b>	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0
Pseudancistrus	<b>Pseudancistrus</b>	0	0
	<b>Pseudancistrus barbatus</b>	0	0
Rineloricaria	<b>Rineloricaria</b>	0	0
Pimelabditus	<b>Pimelabditus moli</b>	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0

Genus	Taxon†	Site	
		Crique Toussaint	Crique Organabo
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0
	Libellulidae	0	0
Caluromys	Caluromys philander	0	0
Cebus	Cebus albifrons	0	0
Homo	Homo	77	216
	Homo sapiens	0	0
	Homininae	0	29
Polychrus	Polychrus marmoratus	0	0
Rhinoclemmys	Rhinoclemmys	0	0
<b>Total</b>		<b>14,083</b>	<b>108,878</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site	
		Crique Petit laussat Aval	Crique Voltalia 2
Atelopus	Atelopus	13	0
	Atelopus flavescens	340	0
Rhinella	Rhinella	0	0
	Rhinella dapsilis	1,010	0
Anomaloglossus	Anomaloglossus baeobatrachus	0	0
Hyla	Hyla helenae	0	0
Hypsiboas	Hypsiboas boans	0	0
	Hypsiboas geographicus	0	0
	Hylinae	0	0
Osteocephalus	Osteocephalus cannatellai	0	0
	Osteocephalus oophagus	0	0
	Osteocephalus taurinus	0	0
Lithodytes	Lithodytes lineatus	0	0
Rana	Rana palmipes	0	0
Crax	Crax rubra	0	0
Mionectes	Mionectes oleagineus	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0
Leporinus	<b>Leporinus affinis</b>	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0
Hoplias	<b>Hoplias malabaricus</b>	4,563	5,911
Prochilodus	<b>Prochilodus</b>	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0
Electrophorus	<b>Electrophorus electricus</b>	0	0
Gymnotus	<b>Gymnotus carapo</b>	1,385	0
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0	0
Potamotrygon	<i>Potamotrygon motoro</i>	0	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0
Tatia	<b>Tatia intermedia</b>	0	0
Doras	<b>Doras micropoeus</b>	0	0
Ancistrus	<b>Ancistrus</b>	1,214	0
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0
Farlowella	<b>Farlowella reticulata</b>	0	0
Harttia	<b>Harttia guianensis</b>	0	0
Hartiella	<b>Hartiella lucifer</b>	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	0
Lithoxus	<b>Lithoxus planquettei</b>	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0
Pseudancistrus	<b>Pseudancistrus</b>	13,094	0
	<b>Pseudancistrus barbatus</b>	0	0
Rineloricaria	<b>Rineloricaria</b>	430	0
Pimelabditus	<b>Pimelabditus moli</b>	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0

Genus	Taxon†	Site	
		Crique Petit laussat Aval	Crique Voltalia 2
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0
	Libellulidae	0	0
Caluromys	Caluromys philander	0	0
Cebus	Cebus albifrons	0	0
Homo	Homo	297	1,723
	Homo sapiens	0	0
	Homininae	0	0
Polychrus	Polychrus marmoratus	239	0
Rhinoclemmys	Rhinoclemmys	180	0
<b>Total</b>		<b>22,765</b>	<b>7,634</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site	
		Crique Voltaia 3	Crique Voltaia 4
Atelopus	Atelopus	0	0
	Atelopus flavescens	0	0
Rhinella	Rhinella	0	0
	Rhinella dapsilis	0	55
Anomaloglossus	Anomaloglossus baeobatrachus	0	0
Hyla	Hyla helenae	0	0
Hypsiboas	Hypsiboas boans	0	719
	Hypsiboas geographicus	0	0
	Hylinae	0	0
Osteocephalus	Osteocephalus cannatellai	0	0
	Osteocephalus oophagus	0	0
	Osteocephalus taurinus	0	0
Lithodytes	Lithodytes lineatus	0	0
Rana	Rana palmipes	0	0
Crax	Crax rubra	0	0
Mionectes	Mionectes oleagineus	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0
Leporinus	<b>Leporinus affinis</b>	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0
Hoplias	<b>Hoplias malabaricus</b>	0	0
Prochilodus	<b>Prochilodus</b>	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0
Electrophorus	<b>Electrophorus electricus</b>	0	0
Gymnotus	<b>Gymnotus carapo</b>	0	0
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0	0
Potamotrygon	<i>Potamotrygon motoro</i>	0	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0
Tatia	<b>Tatia intermedia</b>	0	0
Doras	<b>Doras micropoeus</b>	0	0
Ancistrus	<b>Ancistrus</b>	0	36,590
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0
Farlowella	<b>Farlowella reticulata</b>	0	0
Harttia	<b>Harttia guianensis</b>	0	0
Hartiella	<b>Hartiella lucifer</b>	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	0
Lithoxus	<b>Lithoxus planquettei</b>	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0
Pseudancistrus	<b>Pseudancistrus</b>	0	0
	<b>Pseudancistrus barbatus</b>	0	0
Rineloricaria	<b>Rineloricaria</b>	0	0
Pimelabditus	<b>Pimelabditus moli</b>	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0

Genus	Taxon†	Site	
		Crique Voltalia 3	Crique Voltalia 4
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0
	Libellulidae	0	0
Caluromys	Caluromys philander	0	0
Cebus	Cebus albifrons	0	0
Homo	Homo	43,364	2,190
	Homo sapiens	0	78
	Homininae	0	0
Polychrus	Polychrus marmoratus	0	0
Rhinoclemmys	Rhinoclemmys	0	0
<b>Total</b>		<b>43,364</b>	<b>39,632</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site		
		Crique des Cascades	Apa	Crique Bastien
Atelopus	Atelopus	0	0	0
	Atelopus flavescens	0	0	0
Rhinella	Rhinella	0	0	20
	Rhinella dapsilis	0	5,668	457
Anomaloglossus	Anomaloglossus baeobatrachus	0	0	0
Hyla	Hyla helenae	0	571	0
Hypsiboas	Hypsiboas boans	42,440	115	0
	Hypsiboas geographicus	484	915	0
	Hylinae	0	0	0
Osteocephalus	Osteocephalus cannatellai	0	0	0
	Osteocephalus oophagus	305	0	0
	Osteocephalus taurinus	0	0	78
Lithodytes	Lithodytes lineatus	0	0	0
Rana	Rana palmipes	0	0	0
Crax	Crax rubra	0	0	0
Mionectes	Mionectes oleagineus	0	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0	0
Leporinus	<b>Leporinus affinis</b>	0	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0	0
Hoplias	<b>Hoplias malabaricus</b>	18,429	157	17,253
Prochilodus	<b>Prochilodus</b>	0	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0	0
Electrophorus	<b>Electrophorus electricus</b>	0	0	0
Gymnotus	<b>Gymnotus carapo</b>	24,355	0	0
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0	0	258
Potamotrygon	<i>Potamotrygon motoro</i>	0	0	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0	0
Tatia	<b>Tatia intermedia</b>	0	0	0
Doras	<b>Doras micropoeus</b>	0	0	0
Ancistrus	<b>Ancistrus</b>	0	0	637
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	4,748	97
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0	0
Farlowella	<b>Farlowella reticulata</b>	0	87	0
Harttia	<b>Harttia guianensis</b>	0	0	0
Hartiella	<b>Hartiella lucifer</b>	0	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	0	0
Lithoxus	<b>Lithoxus planquettei</b>	0	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0	0
Pseudancistrus	<b>Pseudancistrus</b>	0	0	0
	<b>Pseudancistrus barbatus</b>	0	0	0
Rineloricaria	<b>Rineloricaria</b>	0	781	0
Pimelabditus	<b>Pimelabditus moli</b>	0	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0	0

Genus	Taxon†	Site		
		Crique des Cascades	Apa	Crique Bastien
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0	681	2,366
	Libellulidae	1,000	0	0
Caluromys	Caluromys philander	0	0	0
Cebus	Cebus albifrons	0	0	0
Homo	Homo	1,296	73	1,127
	Homo sapiens	362	0	0
	Homininae	0	0	0
Polychrus	Polychrus marmoratus	0	0	0
Rhinoclemmys	Rhinoclemmys	0	0	0
<b>Total</b>		<b>88,671</b>	<b>13,796</b>	<b>22,293</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site	
		Crique Penta	Crique Nouvelle France 1
Atelopus	Atelopus	128	413
	Atelopus flavescens	0	0
Rhinella	Rhinella	135	0
	Rhinella dapsilis	3,948	595
Anomaloglossus	Anomaloglossus baeobatrachus	0	0
Hyla	Hyla helenae	416	0
Hypsiboas	Hypsiboas boans	782	0
	Hypsiboas geographicus	0	0
	Hylinae	0	0
Osteocephalus	Osteocephalus cannatellai	0	0
	Osteocephalus oophagus	0	0
	Osteocephalus taurinus	0	0
Lithodytes	Lithodytes lineatus	0	0
Rana	Rana palmipes	0	0
Crax	Crax rubra	0	0
Mionectes	Mionectes oleagineus	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0
Leporinus	<b>Leporinus affinis</b>	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0
Hoplias	<b>Hoplias malabaricus</b>	1,747	4,671
Prochilodus	<b>Prochilodus</b>	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0
Electrophorus	<b>Electrophorus electricus</b>	0	0
Gymnotus	<b>Gymnotus carapo</b>	167	258
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	14	0
Potamotrygon	<i>Potamotrygon motoro</i>	0	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0
Tatia	<b>Tatia intermedia</b>	0	0
Doras	<b>Doras micropoeus</b>	0	0
Ancistrus	<b>Ancistrus</b>	971	8,583
	<i>Ancistrus sp. 2a Parente, 2015</i>	1,824	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0
Farlowella	<b>Farlowella reticulata</b>	0	433
Harttia	<b>Harttia guianensis</b>	0	0
Hartiella	<b>Hartiella lucifer</b>	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	0
Lithoxus	<b>Lithoxus planquettei</b>	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0
Pseudancistrus	<b>Pseudancistrus</b>	0	583
	<b>Pseudancistrus barbatus</b>	0	0
Rineloricaria	<b>Rineloricaria</b>	322	354
Pimelabditus	<b>Pimelabditus moli</b>	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0

Genus	Taxon†	Site	
		Crique Penta	Crique Nouvelle France 1
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0
	Libellulidae	0	0
Caluromys	Caluromys philander	0	0
Cebus	Cebus albifrons	0	0
Homo	Homo	162	0
	Homo sapiens	0	0
	Homininae	0	0
Polychrus	Polychrus marmoratus	0	0
Rhinoclemmys	Rhinoclemmys	0	0
<b>Total</b>		<b>10,616</b>	<b>15,890</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site
		Crique Nouvelle France 2
Atelopus	Atelopus	0
	Atelopus flavescens	0
Rhinella	Rhinella	0
	Rhinella dapsilis	777
Anomaloglossus	Anomaloglossus baeobatrachus	0
Hyla	Hyla helena	0
Hypsiboas	Hypsiboas boans	54
	Hypsiboas geographicus	0
	Hylinae	0
Osteocephalus	Osteocephalus cannatellai	0
	Osteocephalus oophagus	0
	Osteocephalus taurinus	0
Lithodytes	Lithodytes lineatus	0
Rana	Rana palmipes	0
Crax	Crax rubra	0
Mionectes	Mionectes oleagineus	0
Meiothermus	Meiothermus silvanus DSM 9946	0
Leporinus	<b>Leporinus affinis</b>	0
Chilodus	<b>Chilodus punctatus</b>	0
Hoplias	<b>Hoplias malabaricus</b>	1,476
Prochilodus	<b>Prochilodus</b>	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0
Electrophorus	<b>Electrophorus electricus</b>	0
Gymnotus	<b>Gymnotus carapo</b>	361
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0
Potamotrygon	<i>Potamotrygon motoro</i>	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0
Tatia	<b>Tatia intermedia</b>	0
Doras	<b>Doras micropoeus</b>	0
Ancistrus	<b>Ancistrus</b>	11,736
	<i>Ancistrus sp. 2a Parente, 2015</i>	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0
Farlowella	<b>Farlowella reticulata</b>	187
Harttia	<b>Harttia guianensis</b>	1,005
Hartiella	<b>Hartiella lucifer</b>	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	1,069
Lithoxus	<b>Lithoxus planquettei</b>	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0
Panaqolus	<b>Panaqolus koko</b>	0
Pseudancistrus	<b>Pseudancistrus</b>	283
	<b>Pseudancistrus barbatus</b>	0
Rineloricaria	<b>Rineloricaria</b>	585
Pimelabditus	<b>Pimelabditus moli</b>	0
Pimelodus	<b>Pimelodus ornatus</b>	50

Genus	Taxon†	Site
		Crique Nouvelle France 2
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0
	Libellulidae	0
Caluromys	Caluromys philander	0
Cebus	Cebus albifrons	0
Homo	Homo	0
	Homo sapiens	0
	Homininae	0
Polychrus	Polychrus marmoratus	0
Rhinoclemmys	Rhinoclemmys	0
<b>Total</b>		<b>17,583</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site
		Crique Nouvelle France 6
Atelopus	Atelopus	0
	Atelopus flavescens	0
Rhinella	Rhinella	0
	Rhinella dapsilis	0
Anomaloglossus	Anomaloglossus baeobatrachus	0
Hyla	Hyla helena	0
Hypsiboas	Hypsiboas boans	425
	Hypsiboas geographicus	0
	Hylinae	0
Osteocephalus	Osteocephalus cannatellai	0
	Osteocephalus oophagus	0
	Osteocephalus taurinus	0
Lithodytes	Lithodytes lineatus	0
Rana	Rana palmipes	0
Crax	Crax rubra	0
Mionectes	Mionectes oleagineus	13
Meiothermus	Meiothermus silvanus DSM 9946	0
Leporinus	<b>Leporinus affinis</b>	0
Chilodus	<b>Chilodus punctatus</b>	0
Hoplias	<b>Hoplias malabaricus</b>	56
Prochilodus	<b>Prochilodus</b>	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0
Electrophorus	<b>Electrophorus electricus</b>	0
Gymnotus	<b>Gymnotus carapo</b>	0
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0
Potamotrygon	<i>Potamotrygon motoro</i>	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0
Tatia	<b>Tatia intermedia</b>	0
Doras	<b>Doras micropoeus</b>	0
Ancistrus	<b>Ancistrus</b>	5,004
	<i>Ancistrus sp. 2a Parente, 2015</i>	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0
Farlowella	<b>Farlowella reticulata</b>	0
Harttia	<b>Harttia guianensis</b>	0
Hartiella	<b>Hartiella lucifer</b>	24,060
Hemiancistrus	<b>Hemiancistrus medians</b>	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0
Lithoxus	<b>Lithoxus planquettei</b>	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0
Panaqolus	<b>Panaqolus koko</b>	0
Pseudancistrus	<b>Pseudancistrus</b>	0
	<b>Pseudancistrus barbatus</b>	0
Rineloricaria	<b>Rineloricaria</b>	0
Pimelabditus	<b>Pimelabditus moli</b>	0
Pimelodus	<b>Pimelodus ornatus</b>	0

Genus	Taxon†	Site
		Crique Nouvelle France 6
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0
	Libellulidae	0
Caluromys	Caluromys philander	0
Cebus	Cebus albifrons	0
Homo	Homo	1,740
	Homo sapiens	363
	Homininae	0
Polychrus	Polychrus marmoratus	0
Rhinoclemmys	Rhinoclemmys	0
<b>Total</b>		<b>31,661</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site
		Crique Nouvelle France 5
Atelopus	Atelopus	2,211
	Atelopus flavescens	0
Rhinella	Rhinella	79
	Rhinella dapsilis	164
Anomaloglossus	Anomaloglossus baeobatrachus	12
Hyla	Hyla helena	0
Hypsiboas	Hypsiboas boans	0
	Hypsiboas geographicus	0
	Hylinae	0
Osteocephalus	Osteocephalus cannatellai	0
	Osteocephalus oophagus	33
	Osteocephalus taurinus	26
Lithodytes	Lithodytes lineatus	21
Rana	Rana palmipes	0
Crax	Crax rubra	0
Mionectes	Mionectes oleagineus	0
Meiothermus	Meiothermus silvanus DSM 9946	0
Leporinus	<b>Leporinus affinis</b>	0
Chilodus	<b>Chilodus punctatus</b>	0
Hoplias	<b>Hoplias malabaricus</b>	244
Prochilodus	<b>Prochilodus</b>	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0
Electrophorus	<b>Electrophorus electricus</b>	0
Gymnotus	<b>Gymnotus carapo</b>	1,208
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0
Potamotrygon	<i>Potamotrygon motoro</i>	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0
Tatia	<b>Tatia intermedia</b>	0
Doras	<b>Doras micropoeus</b>	0
Ancistrus	<b>Ancistrus</b>	12,535
	<i>Ancistrus sp. 2a Parente, 2015</i>	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0
Farlowella	<b>Farlowella reticulata</b>	276
Harttia	<b>Harttia guianensis</b>	72
Hartiella	<b>Hartiella lucifer</b>	1,959
Hemiancistrus	<b>Hemiancistrus medians</b>	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0
Lithoxus	<b>Lithoxus planquettei</b>	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0
Panaqolus	<b>Panaqolus koko</b>	0
Pseudancistrus	<b>Pseudancistrus</b>	2,207
	<b>Pseudancistrus barbatus</b>	0
Rineloricaria	<b>Rineloricaria</b>	861
Pimelabditus	<b>Pimelabditus moli</b>	0
Pimelodus	<b>Pimelodus ornatus</b>	0

Genus	Taxon†	Site
		Crique Nouvelle France 5
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0
	Libellulidae	0
Caluromys	Caluromys philander	130
Cebus	Cebus albifrons	0
Homo	Homo	383
	Homo sapiens	0
	Homininae	0
Polychrus	Polychrus marmoratus	0
Rhinoclemmys	Rhinoclemmys	0
<b>Total</b>		<b>22,421</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site
		Crique Nouvelle France 4
Atelopus	Atelopus	1,303
	Atelopus flavescens	0
Rhinella	Rhinella	80
	Rhinella dapsilis	673
Anomaloglossus	Anomaloglossus baeobatrachus	0
Hyla	Hyla helena	0
Hypsiboas	Hypsiboas boans	33
	Hypsiboas geographicus	0
	Hylinae	0
Osteocephalus	Osteocephalus cannatellai	0
	Osteocephalus oophagus	0
	Osteocephalus taurinus	0
Lithodytes	Lithodytes lineatus	0
Rana	Rana palmipes	0
Crax	Crax rubra	0
Mionectes	Mionectes oleagineus	0
Meiothermus	Meiothermus silvanus DSM 9946	0
Leporinus	<b>Leporinus affinis</b>	0
Chilodus	<b>Chilodus punctatus</b>	0
Hoplias	<b>Hoplias malabaricus</b>	1,419
Prochilodus	<b>Prochilodus</b>	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0
Electrophorus	<b>Electrophorus electricus</b>	0
Gymnotus	<b>Gymnotus carapo</b>	255
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0
Potamotrygon	<i>Potamotrygon motoro</i>	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0
Tatia	<b>Tatia intermedia</b>	0
Doras	<b>Doras micropoeus</b>	0
Ancistrus	<b>Ancistrus</b>	12,090
	<i>Ancistrus sp. 2a Parente, 2015</i>	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0
Farlowella	<b>Farlowella reticulata</b>	1,011
Harttia	<b>Harttia guianensis</b>	0
Hartiella	<b>Hartiella lucifer</b>	198
Hemiancistrus	<b>Hemiancistrus medians</b>	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0
Lithoxus	<b>Lithoxus planquettei</b>	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0
Panaqolus	<b>Panaqolus koko</b>	0
Pseudancistrus	<b>Pseudancistrus</b>	219
	<b>Pseudancistrus barbatus</b>	0
Rineloricaria	<b>Rineloricaria</b>	574
Pimelabditus	<b>Pimelabditus moli</b>	0
Pimelodus	<b>Pimelodus ornatus</b>	0

Genus	Taxon†	Site
		Crique Nouvelle France 4
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0
	Libellulidae	0
Caluromys	Caluromys philander	0
Cebus	Cebus albifrons	0
Homo	Homo	122
	Homo sapiens	14
	Homininae	0
Polychrus	Polychrus marmoratus	0
Rhinoclemmys	Rhinoclemmys	0
<b>Total</b>		<b>17,991</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site	
		Crique Nouvelle France 3	Crique à l'Est
Atelopus	Atelopus	1,529	143
	Atelopus flavescens	0	0
Rhinella	Rhinella	0	0
	Rhinella dapsilis	1,678	3,943
Anomaloglossus	Anomaloglossus baeobatrachus	0	0
Hyla	Hyla helenae	0	0
Hypsiboas	Hypsiboas boans	221	511
	Hypsiboas geographicus	0	183
	Hylinae	0	0
Osteocephalus	Osteocephalus cannatellai	0	0
	Osteocephalus oophagus	0	0
	Osteocephalus taurinus	0	0
Lithodytes	Lithodytes lineatus	0	0
Rana	Rana palmipes	0	0
Crax	Crax rubra	867	0
Mionectes	Mionectes oleagineus	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0
Leporinus	<b>Leporinus affinis</b>	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0
Hoplias	<b>Hoplias malabaricus</b>	4,583	4,304
Prochilodus	<b>Prochilodus</b>	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0
Electrophorus	<b>Electrophorus electricus</b>	0	0
Gymnotus	<b>Gymnotus carapo</b>	707	1,213
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0	0
Potamotrygon	<i>Potamotrygon motoro</i>	0	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0
Tatia	<b>Tatia intermedia</b>	0	0
Doras	<b>Doras micropoeus</b>	0	0
Ancistrus	<b>Ancistrus</b>	18,766	14,780
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	0
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	1,268
Farlowella	<b>Farlowella reticulata</b>	672	154
Harttia	<b>Harttia guianensis</b>	1,484	0
Hartiella	<b>Hartiella lucifer</b>	62	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	0
Lithoxus	<b>Lithoxus planquettei</b>	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0
Pseudancistrus	<b>Pseudancistrus</b>	3,610	5,045
	<b>Pseudancistrus barbatus</b>	0	0
Rineloricaria	<b>Rineloricaria</b>	2,262	1,073
Pimelabditus	<b>Pimelabditus moli</b>	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0

Genus	Taxon†	Site	
		Crique Nouvelle France 3	Crique à l'Est
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0
	Libellulidae	0	0
Caluromys	Caluromys philander	0	0
Cebus	Cebus albifrons	0	0
Homo	Homo	67	651
	Homo sapiens	0	0
	Homininae	0	0
Polychrus	Polychrus marmoratus	0	0
Rhinoclemmys	Rhinoclemmys	0	0
<b>Total</b>		<b>36,508</b>	<b>33,268</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

Genus	Taxon†	Site			
		Pikin Tabiki	Saut Sonelle	Papaichton	Saut Bief
Atelopus	Atelopus	0	0	0	0
	Atelopus flavescens	0	0	0	0
Rhinella	Rhinella	0	0	0	70
	Rhinella dapsilis	0	0	0	2,063
Anomaloglossus	Anomaloglossus baeobatrachus	0	0	0	0
Hyla	Hyla helenae	0	0	0	0
Hypsiboas	Hypsiboas boans	0	443	0	1,913
	Hypsiboas geographicus	0	0	0	0
	Hylinae	0	0	0	0
Osteocephalus	Osteocephalus cannatellai	0	0	0	0
	Osteocephalus oophagus	0	0	0	0
	Osteocephalus taurinus	0	0	0	0
Lithodytes	Lithodytes lineatus	0	0	0	0
Rana	Rana palmipes	0	0	0	147
Crax	Crax rubra	0	0	0	0
Mionectes	Mionectes oleagineus	0	0	0	0
Meiothermus	Meiothermus silvanus DSM 9946	8,040	80,231	29	0
Leporinus	<b>Leporinus affinis</b>	33,740	0	0	0
Chilodus	<b>Chilodus punctatus</b>	0	0	0	859
Hoplias	<b>Hoplias malabaricus</b>	0	0	0	1,329
Prochilodus	<b>Prochilodus</b>	0	0	0	0
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0	0	3,028
Electrophorus	<b>Electrophorus electricus</b>	0	0	0	13,825
Gymnotus	<b>Gymnotus carapo</b>	0	0	0	2,861
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0	0	0	0
Potamotrygon	<i>Potamotrygon motoro</i>	0	0	0	5,631
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0	0	0
Tatia	<b>Tatia intermedia</b>	0	0	0	2,083
Doras	<b>Doras micropoeus</b>	0	0	0	7,550
Ancistrus	<b>Ancistrus</b>	0	0	0	0
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	0	0	30,841
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0	0	0
Farlowella	<b>Farlowella reticulata</b>	0	0	0	0
Harttia	<b>Harttia guianensis</b>	0	0	0	0
Hartiella	<b>Hartiella lucifer</b>	0	0	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0	0	0
Hypostomus	<b>Hypostomus gymnorhynchus</b>	41,924	0	0	20,134
Lithoxus	<b>Lithoxus planquettei</b>	0	0	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0	0	0
Panaqolus	<b>Panaqolus koko</b>	0	0	0	0
Pseudancistrus	<b>Pseudancistrus</b>	0	0	0	0
	<b>Pseudancistrus barbatus</b>	111,178	0	0	0
Rineloricaria	<b>Rineloricaria</b>	0	0	0	1,104
Pimelabditus	<b>Pimelabditus moli</b>	0	0	0	0
Pimelodus	<b>Pimelodus ornatus</b>	0	0	0	0

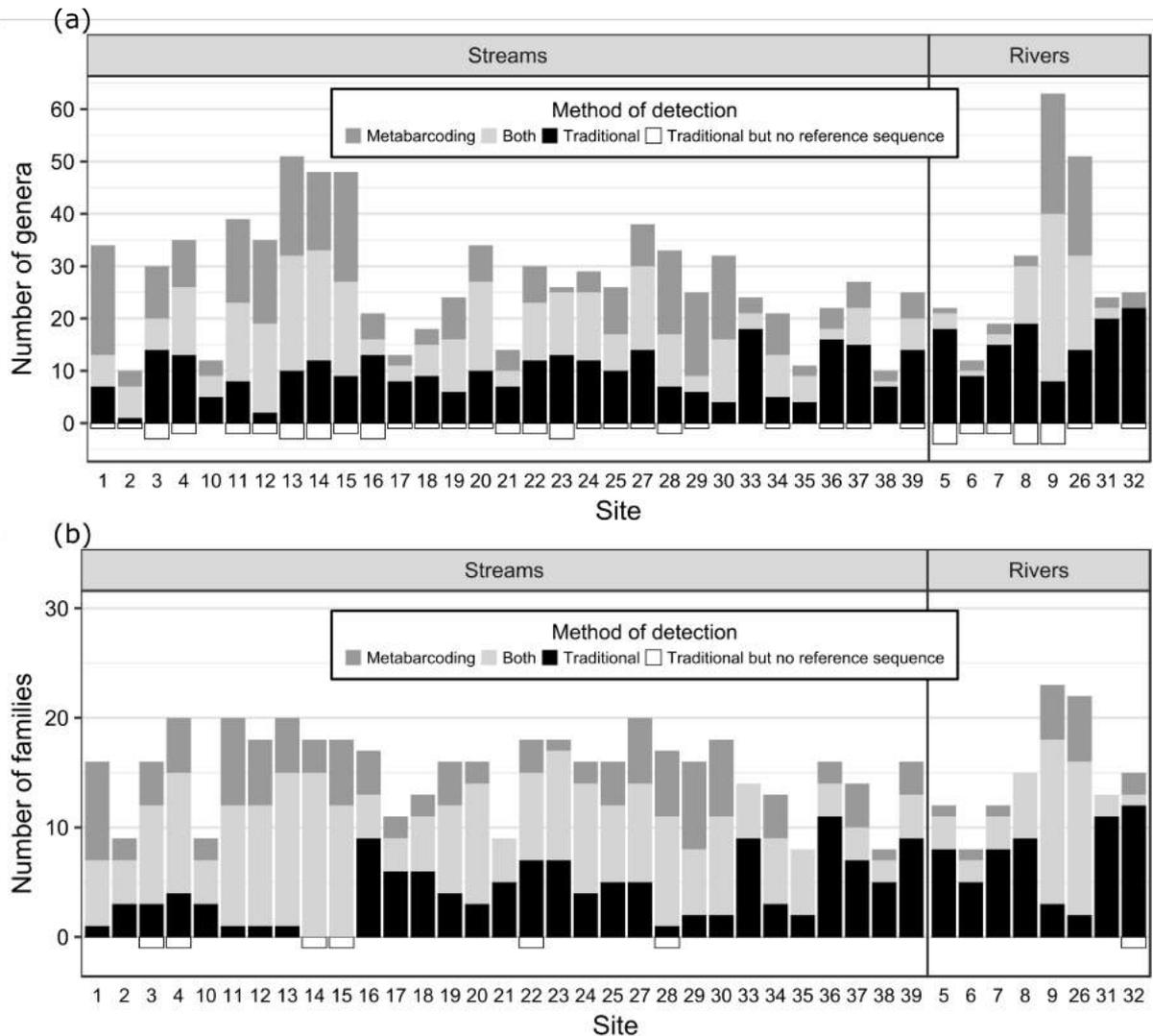
Genus	Taxon†	Site			
		Pikin Tabiki	Saut Sonelle	Papaichton	Saut Bief
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0	0	1,830
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0	0	0
	Libellulidae	0	0	0	0
Caluromys	Caluromys philander	0	0	0	0
Cebus	Cebus albifrons	0	0	0	0
Homo	Homo	40,951	54,627	220,895	413
	Homo sapiens	0	0	0	0
	Homininae	0	0	913	0
Polychrus	Polychrus marmoratus	0	0	0	0
Rhinoclemmys	Rhinoclemmys	0	0	0	0
<b>Total</b>		<b>235,833</b>	<b>135,301</b>	<b>221,837</b>	<b>95,681</b>

†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana

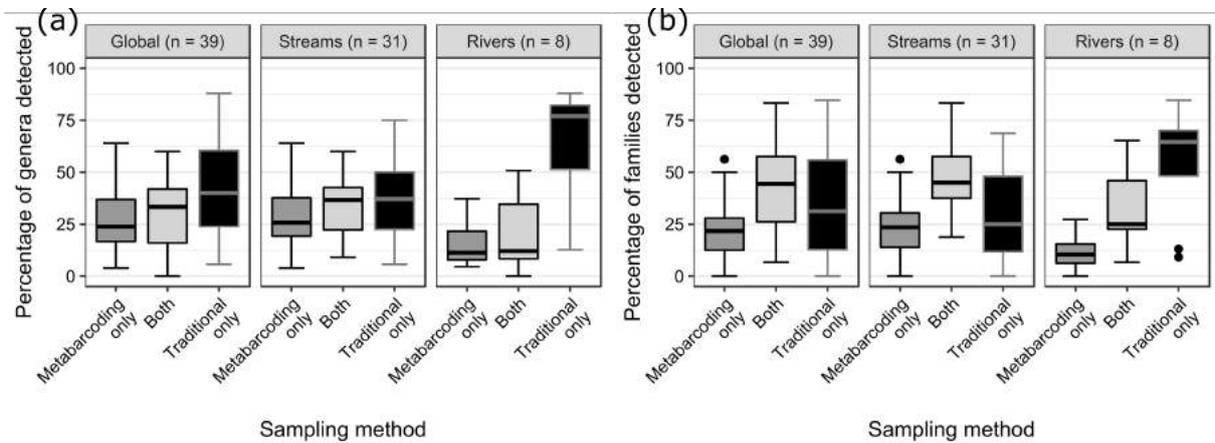
Genus	Taxon†	Site			
		Machicou	Athanase	Twenke	Apsik Icholi
Atelopus	Atelopus	0	0	0	0
	Atelopus flavescens	0	0	0	0
Rhinella	Rhinella	0	0	0	105
	Rhinella dapsilis	0	0	0	0
Anomaloglossus	Anomaloglossus baeobatrachus	0	0	0	0
Hyla	Hyla helenae	0	0	0	0
Hypsiboas	Hypsiboas boans	0	0	0	86
	Hypsiboas geographicus	0	0	0	0
	Hylinae	0	0	0	0
Osteocephalus	Osteocephalus cannatai	0	0	0	0
	Osteocephalus oophagus	0	0	0	0
	Osteocephalus taurinus	0	0	0	0
Lithodytes	Lithodytes lineatus	0	0	0	0
Rana	Rana palmipes	0	0	0	0
Crax	Crax rubra	0	0	0	0
Mionectes	Mionectes oleagineus	0	0	0	0
Meiothermus	Meiothermus silvanus DSM 9946	0	0	0	0
Leporinus	<b>Leporinus affinis</b>	0	0	0	7,578
Chilodus	<b>Chilodus punctatus</b>	0	0	0	0
Hoplias	<b>Hoplias malabaricus</b>	0	0	0	1,049
Prochilodus	<b>Prochilodus</b>	0	0	0	2,776
Pterengraulis	<b>Pterengraulis atherinoides</b>	0	0	0	0
Electrophorus	<b>Electrophorus electricus</b>	0	0	0	0
Gymnotus	<b>Gymnotus carapo</b>	0	0	0	0
Eigenmannia	<i>Eigenmannia sp. CBM-ZF-10620</i>	0	0	0	0
Potamotrygon	<i>Potamotrygon motoro</i>	0	0	0	0
Auchenipterus	<i>Auchenipterus demerarae</i>	0	0	0	37
Tatia	<b>Tatia intermedia</b>	0	0	0	0
Doras	<b>Doras micropoeus</b>	0	0	0	8,723
Ancistrus	<b>Ancistrus</b>	0	0	0	22
	<i>Ancistrus sp. 2a Parente, 2015</i>	0	0	0	1,296
Cteniloricaria	<b>Cteniloricaria platystoma</b>	0	0	0	84
Farlowella	<b>Farlowella reticulata</b>	0	0	0	0
Harttia	<b>Harttia guianensis</b>	0	0	0	2,940
Hartiella	<b>Hartiella lucifer</b>	0	0	0	0
Hemiancistrus	<b>Hemiancistrus medians</b>	0	0	0	3,826
Hypostomus	<b>Hypostomus gymnorhynchus</b>	0	20	0	8,828
Lithoxus	<b>Lithoxus planquettei</b>	0	0	0	0
Metaloricaria	<b>Metaloricaria paucidens</b>	0	0	0	1,363
Panaqolus	<b>Panaqolus koko</b>	0	0	0	792
Pseudancistrus	<b>Pseudancistrus</b>	0	0	0	16,115
	<b>Pseudancistrus barbatus</b>	0	0	13,822	2,766
Rineloricaria	<b>Rineloricaria</b>	0	0	0	0
Pimelabditus	<b>Pimelabditus moli</b>	0	0	0	621
Pimelodus	<b>Pimelodus ornatus</b>	0	0	0	1,966

Genus	Taxon†	Site			
		Machicou	Athanase	Twenke	Apsik Icholi
Pseudoplatystoma	<b>Pseudoplatystoma fasciatum</b>	0	0	63,666	3,299
Polycentrus	<b>Polycentrus schomburgkii</b>	0	0	0	0
	Libellulidae	0	0	0	0
Caluromys	Caluromys philander	0	0	0	0
Cebus	Cebus albifrons	0	0	0	0
Homo	Homo	32,689	296,897	58,549	505
	Homo sapiens	0	0	0	100
	Homininae	0	0	0	0
Polychrus	Polychrus marmoratus	0	0	0	0
Rhinoclemmys	Rhinoclemmys	0	0	0	0
<b>Total</b>		<b>32,689</b>	<b>296,917</b>	<b>136,037</b>	<b>64,877</b>

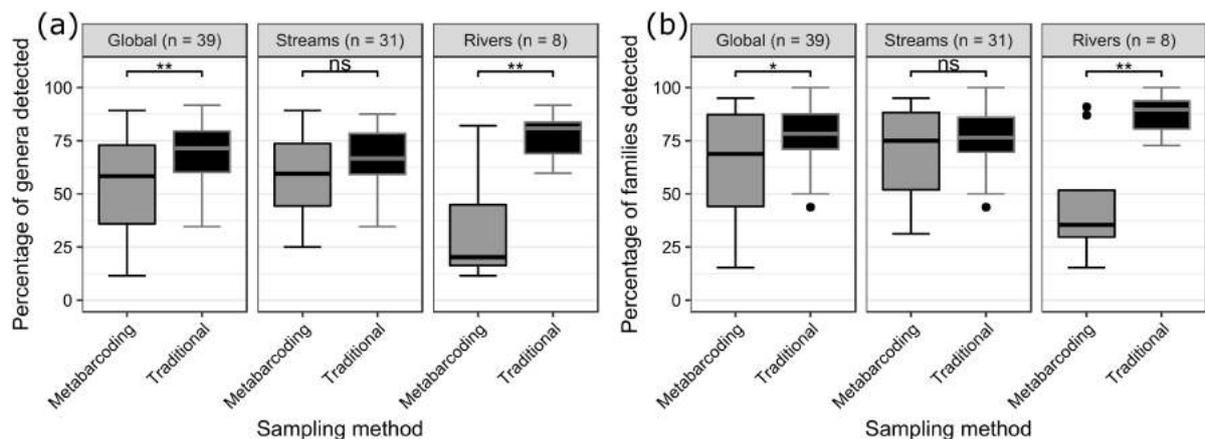
†: Bold taxa are fish present in French Guiana; italicized taxa are fish not known from French Guiana



**Figure IV.S1:** Number of genera (a) and families (b) detected at each site with traditional and eDNA metabarcoding methods. Taxa caught only with traditional methods are in black (taxa informed in the reference database) and white (absent from the reference database), detected only with eDNA metabarcoding in dark grey and by both eDNA and traditional methods in light grey. Sites are indicated by numbers (see Table 1 in the main text for number signification)



**Figure IV.S2:** Percentage of genera (a) and families (b) detected with traditional and eDNA metabarcoding methods. Only taxa informed in the reference database were considered. Taxa caught only with traditional methods are in black, those detected only with eDNA metabarcoding in dark grey and those detected by both eDNA and traditional methods (shared) in light grey. Global refers to all the 39 sites considered together (stream and river sites are pooled). Streams refers only to the 31 stream sites and Rivers refer to the 8 rivers sites



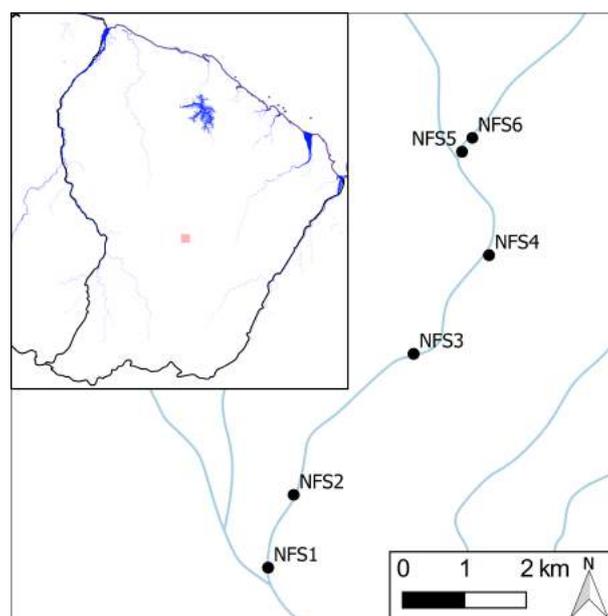
**Figure IV.S3:** Percentage of genera (a) and families (b) detected, with eDNA metabarcoding (dark grey) or traditional methods (black). Global refers to all the 39 sites considered together (stream and river sites are pooled). Streams refers only to the 31 stream sites and Rivers refer to the 8 rivers sites. Difference between eDNA and traditional methods were tested using Wilcoxon rank-sum test, ns:  $p > .05$ ; \*:  $p < .05$ ; \*\*:  $p < .01$

### IV.3 Additional analyses

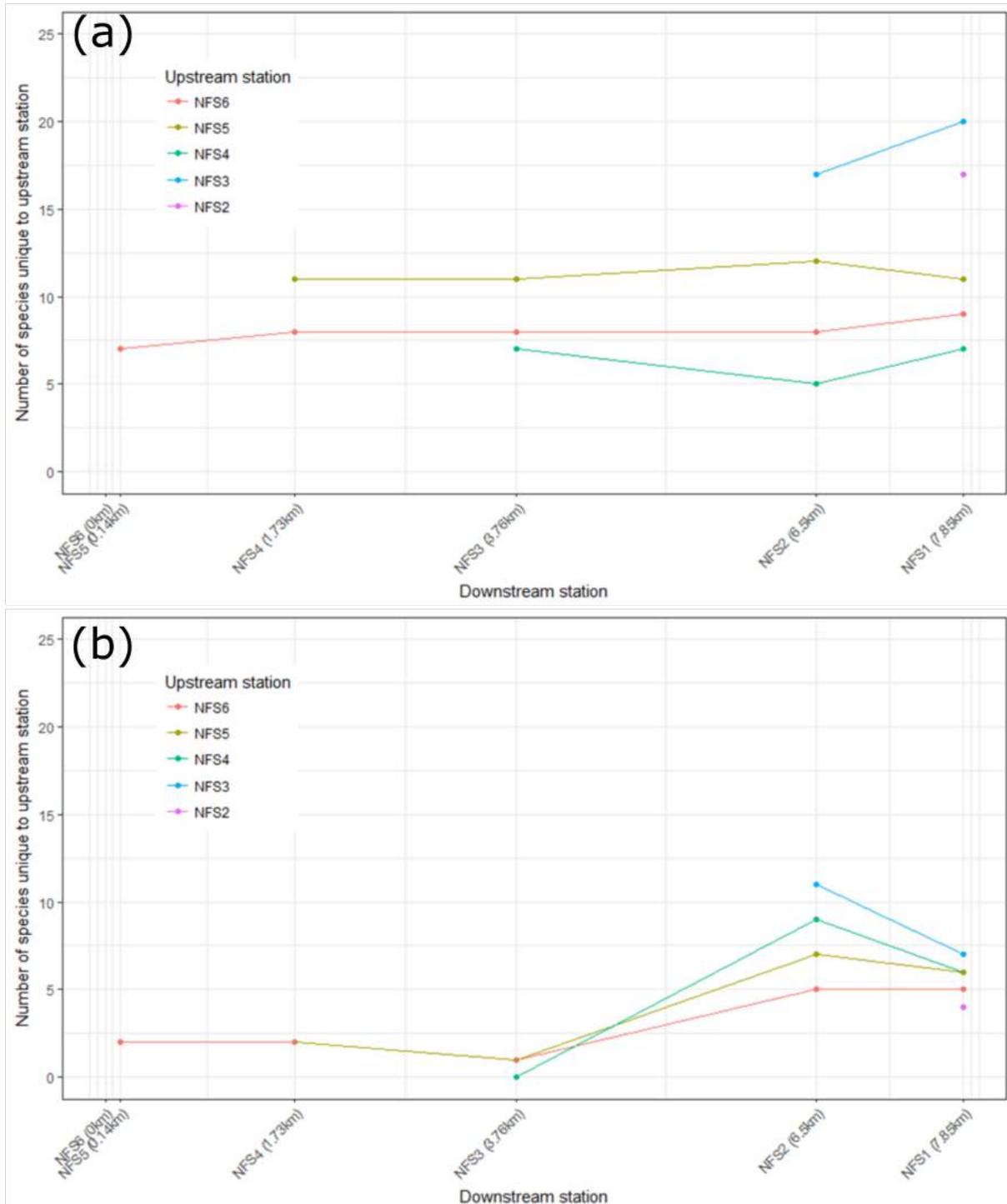
#### Analyses of the distance of detection on one stream

In addition to this analysis but not included in the submitted/accepted version, I used a subset of the stream sites that were sampled along the same stream (Nouvelle France stream, Figure IV.9) to roughly estimate the distance of detection for eDNA.

This study remains a preliminary approach of the distance of detection (no replicate), but it can nevertheless provide some hints for future researches. We compared each site to all the downstream sites and counted the number of unique species to this site, *i.e.* species that are found only in the upstream site, for both traditional sampling and eDNA metabarcoding. We have seen in the Chapter II that species assemblages change along the upstream-downstream gradient. We thus expect that the number of unique species should increase with the distance between sites. If eDNA is transported along the stream, the species occurring only in upstream sites should still be detected downstream, and the number of unique species in the upstream site should be lower when using eDNA metabarcoding than for traditional sampling. We observed that, with traditional sampling, the number of species unique to the upstream site was always higher than five and did not increase with distance (Figure IV.10a). In comparison, the number of unique species was lower with eDNA metabarcoding (Figure IV.10b) and increase with the distance when comparing to NFS2. For the most upstream sites (NFS4 to NFS6), the species were almost all detected from one site to the others. The detection distance of eDNA thus correspond to the distance between the upstream and the downstream sites (about 8 km). In temperate streams, this distance was evaluated between hundreds of meters (Jane *et al.*, 2015) to kilometers (about 2km in Civade *et al.*, 2016, 10km in Deiner &



**Figure IV.9:** Location of the 6 sampling sites (from upstream NFS6 site to downstream NFS1 site) on the Nouvelle France stream. The location of the stream is indicated in red on the map of French Guiana



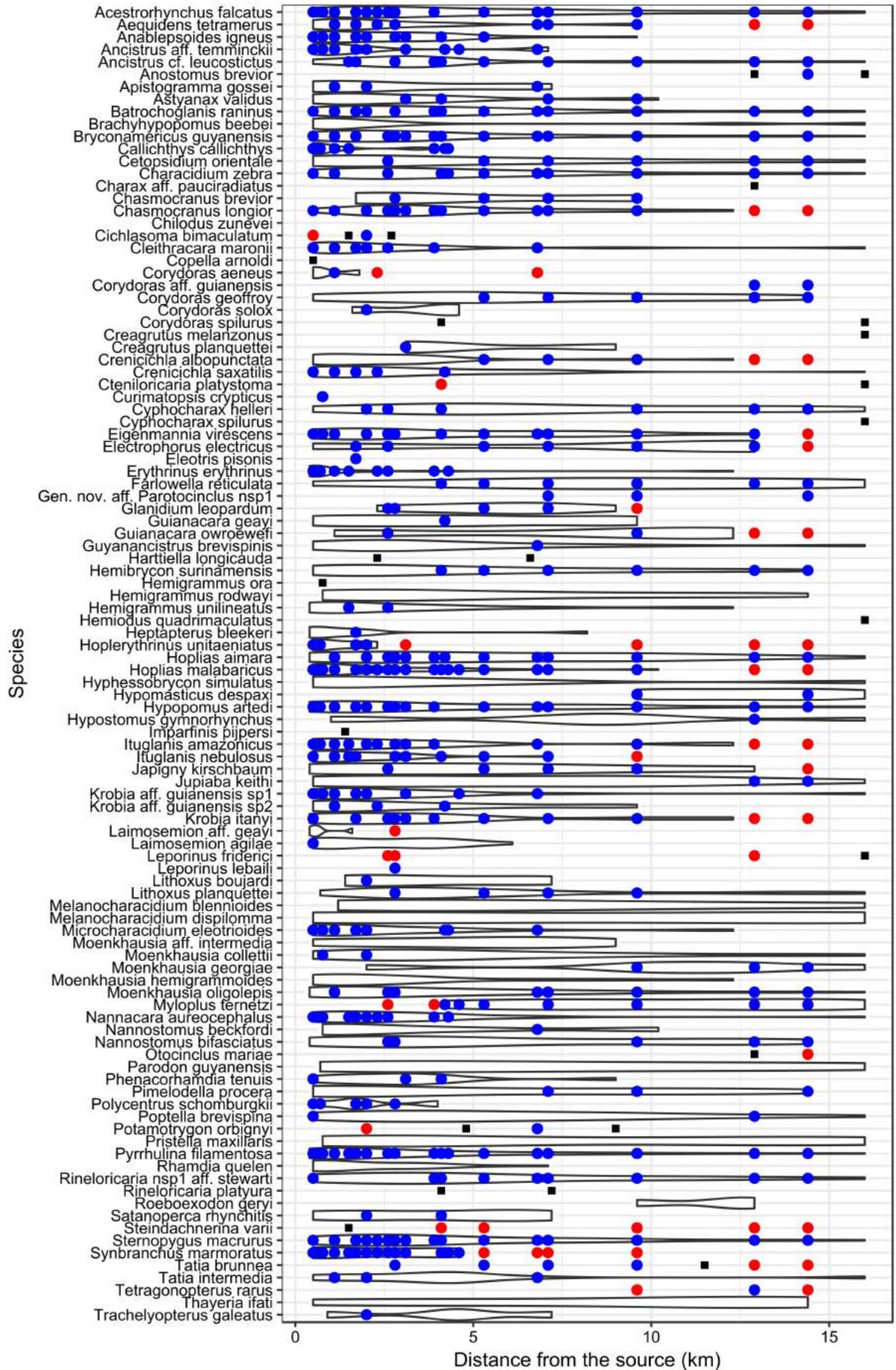
**Figure IV.10:** For each site (colored line), the number of species only found in this site compared to the downstream sites is indicated when using rotenone (a) or eDNA metabarcoding (b). Distance from the first sites (NSF6) is indicated.

Altermatt, 2014). The environmental characteristics of Guianese streams that could have affected DNA degradation (acidity, turbidity or UV), and reduce eDNA metabarcoding efficiency, seem to have little effect on this detection distance.

### **Detection and environmental range**

In another exploratory analysis, I checked if species detection fell into the known environmental range of species distribution based on traditional sampling. Their distributions are highly influenced by the upstream-downstream gradient and I thus used the distance of the site from the source to compare the detection and the known range. A large number of species have distributed across a wide range of sites IV.11, but for species with smaller range, little detection fell outside the known range (10% of wrong detections). These wrong detections were often at higher distance from the source, that can partly be explained by the detection distance of the eDNA. However, wrong assignments or the lack of reference for some species, as suggested in the submitted work, might also caused this wrong detection. We should also be aware that traditional sampling methods do not provide an exhaustive image of the fish fauna and some inconsistencies of species distributions can either be due to traditional or eDNA biases. Nevertheless, the majority of species are detected within their known distribution range and focusing our attention on species for which detections are not congruent would be beneficial to understand the specificity of eDNA in tropical freshwaters. Once this achieved, eDNA metabarcoding will help to refine species distribution range.

**Figure IV.11:** Known distribution of species along the distance from the source gradient based on traditional sampling (violin, when more than 2 occurrences with height proportional to the number of occurrences, and ■ when less than 3 occurrences). Dots represent sites where the species was detected using eDNA metabarcoding, with ● representing detection within the known range and ● for detection outside the known range



### **Comparison of the cost and specificity of traditional and eDNA metabarcoding methods**

If we compare both methods from technical and financial point of view (Table IV.2), eDNA metabarcoding is much easier to achieve compared to traditional sampling, since a single operator with a peristaltic pump is needed, compared to a 3 to 5 persons team with heavy equipment for traditional samples (gill-nets, block-nets, hand nets). For the sampling cost, the complete eDNA analysis (sampling tools, sampling and subsequent analyses) are estimated to 530€per sample, once the reference database is implemented. The cost for metabarcoding is lower compared to traditional sampling (around 700€per sample) and it will probably decreases in the future, with the decrease of molecular analysis cost.

In conclusion, eDNA metabarcoding is applicable to assess freshwater fish assemblage in French Guiana and given these first results, it is for the moment more a complementary sampling methods than an alternative to the traditional samples. It nevertheless represents the only efficient sampling method that can still be used in streams since rotenone samples are no longer allowed. Environmental DNA developments are still pending and determining an optimal sampling effort (filtration volume, number of replicate) and distance of detection, but also to complement the genetic reference database appear as the most urgent developments needed to enhances eDNA metabarcoding relevance in Guyanese freshwaters.

**Table IV.2:** Sampling characteristics and costs of eDNA metabarcoding and traditional sampling methods

		<b>Metabarcoding</b>	<b>Traditional</b>	
			Rotenone	Nets
Duration of	Sampling	30 min/site	$\frac{1}{2}$ day/site	$\frac{1}{2}$ day/site
	Species assignment or determination	< 3 months for all sites	2h/site	$\frac{1}{2}$ day/site
Determination based on		Genetic reference database	Specialist/identification key	
Cost for 1 sampling		Kit + biomolecular and bioinformatic analyses (414€)	Rotenone (30€) + Nets (20€)	Nets (246€)
	Human resources for sampling (3-5 years experienced technician; 16.47€/h)	1 minimum (8.28€)	3 minimum (271.76€)	3 minimum (345.87€+ 90€)
Total cost/site		422.28€	321.76€	681.87€
Applicable to		All type of water	Small streams (but banned)	Rivers
Sampling detection distance		Variable (all upstream waters, from hundred meters to kilometers)	local (dozens of meters)	Local (hundreds of meters)
Specificity		Can detect a wide range of organisms	Habitat selective	Species selective (habitat and size)



# Chapter V: Conclusion

## V.1 Assembly processes of Guianese freshwater assemblages

These works is in continuation of previous works that have allowed increasing the knowledge about Guianese fish ecology. This thesis allowed to extend the previous database about fish species assemblages with the integration of new sampling sites (especially the rivers), the development of a genetic database that will be used for two different goals (phylogeny and metabarcoding) and the inclusion of morphological measurements to measure functional traits. The database updating is still an ongoing project, with new sampling planned to increase the tissue and photographic collections, and local assemblage information. This will allow extending the approach I used in this thesis to other part of the hydrological network (other river sites or estuary areas).

All these data allowed me to characterize the taxonomic, phylogenetic and functional diversities of Guianese freshwater fish assemblages, and these three facets of diversity revealed the processes that shaped fish assemblages. The results of these works highlight that high diversity of fish observed in the tropical waters in French Guiana is structured along an upstream to downstream gradient, with higher local diversity downstream (Chapter II). This pattern is coupled with the effect of the spatial structure of the hydrological network, that limits dispersion of fish species between assemblages (Chapters II and III). Rivers act as a barrier to dispersal for stream associated species, thus creating a strong turnover of species between stream assemblages. We also observed dispersal limitation between river assemblages, but the biogeographical history of Guianese region had differently impacted streams and rivers assemblages. Streams assemblages are isolated for a longer time compared to rivers assemblages, which had exchange species during low sea level period.

## V.2 Phylogenetic characteristics of assemblages

Working with different types of data requires using the intersection of these data and thus some species are excluded from the analysis. Here, in my thesis, 239 species were identified in the assemblage data (both rivers and streams) but only 195 species were informed both in the assemblage dataset, in the morphological traits database and in the phylogenetic tree. We thus excluded 44 species from the analyses, either due to their low occurrence or their absence in the phylogenetic tree. For the species that are missing from this phylogenetic tree, some are missing due to the fact that no tissue sample was available for a minority of them (and will thus need supplementary fieldworks), or that sequence amplification did not work (especially species from the Loricariidae family). For those last species, supplementary collections of tissue and changing the body part collected (muscle instead of fin tissue, Raphael Covain, pers. comm.) can solve amplification problem. An alternative could be to incorporate them

as polytomies within their respective genera (Qian *et al.*, 2013; Webb & Donoghue, 2005). However for species that are the unique representative of their genera (e.g. *Metynnus lippincottianus*), their placement in the phylogenetic tree will create polytomies at higher taxonomic level (subfamily or family) and thus will impact phylogenetic diversity estimates (Davies *et al.*, 2012). The best estimates of phylogenetic diversity would be given by a phylogeny including all the species found in Guianese assemblages, and even more if those species are replaced in a larger context like the evolutive history of Neotropical ichthyofauna.

### **V.3 Functional characteristics of assemblages**

Recently, the intraspecific variability in functional traits has been highlighted as an important component to estimate functional diversity and can help to assess the processes structuring assemblages that can be missed when using mean values (Albert *et al.*, 2012; Schleuter *et al.*, 2010; Spasojevic *et al.*, 2016). Measuring this intraspecific variability at the individual level would however be impossible for Guianese freshwater assemblages. Indeed, nets sampling select individuals based on their size and their habitat and this will lead to underestimate intraspecific variability in assemblages. Carmona *et al.* (2016) proposed a framework based trait probability density to overcome this problem. The distribution of trait values can be estimated from the measures and an estimated variability. This probabilistic framework however needs to be tested, but is a first step to incorporate individual variability for tropical fish assemblages. This inclusion of intraspecific variability should also be accompanied with a reflexion on the spatial scale at which it will be taken into account. We could expect that for species inhabiting both streams and rivers, individuals of these species will differ between these two habitats. Differentiating intraspecific variability estimations for these species between the different habitats will thus be needed to correctly estimate functional diversity of assemblages. This needs to be tested, and maybe thought at other spatial scales (difference between upstream and downstream, between drainage basins) to draw robust conclusions on assembly rules.

### **V.4 Assembly processes and anthropogenic disturbances**

Although some processes needs to be confirmed by further analyses (Chapter III.2), this work can be used as a reference to understand how anthropogenic disturbances affect fish assemblage. Data for fish assemblages in disturbed streams were already available (Allard, 2014 PhD thesis) and we now have the information for river site under anthropogenic disturbances. With these data at hand, doing the same complete analysis of assembly rules (Chapter III) in these disturbed sites will allow to identify the dominant processes under disturbed conditions. We can then verify if local neutral processes and dispersal limitation are still the dominant processes explaining the diversity and the structure of fish assemblages or if disturbances modified the relative strength of the different processes that shifts assemblage composition and structure.

The observed changes of assemblage compositions between non-impacted and disturbed sites (Allard *et al.*, 2016; Dedieu *et al.*, 2015) might be explained by a shift in local processes from neutral to deterministic processes, with a strong environmental filtering for taller and ubiquitous species.

However, the future studies of assembly rules rely on the development of a sampling method that give exhaustive picture of assemblages. We see that eDNA metabarcoding is at the moment in a development phase and several tests are need before it can be routinely used to asses fish assemblages. The first essential step will be to evaluate the sampling effort of the eDNA metabarcoding (the number of replicates or the volume of filtered water). Metabarcoding samples from the same location can greatly differ in their assemblage composition (Ficetola *et al.*, 2015). The number of replicates to obtain an almost full picture of the local assemblages will create a financial constrain and a choice between the number of sampling sites (and thus the generalization of the results) and the representativeness of local assemblages (and thus the confidence in the results). In addition, if assemblages detected with metabarcoding are biased toward particular species irrespective of the environmental conditions (the rarest species are never detected for example), looking at assembly rules might give the same results as for the river assemblages studied in the Chapter III.2, *i.e.* the difference between the scale at which processes act and the scale at which we observe assemblages might blur the effect of deterministic processes. Once this calibration phase of the metabarcoding method for tropical stream completed, its use will be a major step for inventories or ecological and conservation studies in these areas.

## V.5 Leaving the “process from pattern” way of thinking?

Finally, I think that the greatest improvement that can be done in the assessment of assembly rules will be to shift the way we assess them. For the moment, a large part of studies assembly rules rely on the “process from pattern” template, which have been highly criticized (see Chapter I) and led to the development of strong frameworks and hypotheses about the patterns expected under each process or their combinations (as developed in these thesis). This warning about the inductive way of thinking has been also reported as biogeography (Crisp *et al.*, 2011) or in spatial ecology (McIntire & Fajardo, 2009). A process from experimental test template maybe will strengthen our view on assembly rules. This type of experimental approach is mainly used in plants communities (Baer *et al.*, 2016; Loranger *et al.*, 2016; HilleRisLambers *et al.*, 2012), because abiotic and biotic conditions are easily controlled and the strength of each processes can be modified. An alternative is to associate observational and experimental studies, as Fayle *et al.* (2015) who confirmed their field observations by experimentally induced interactions between ant species. These two ways would be more difficult (if not impossible) to implement for larger species from speciose area, and will require a lot of work, but tending to them (for example looking at the ecological and life history traits of species in addition to their morphological traits) would be the greatest help in understanding the processes

that structure assemblages.

Community ecology is a field that successfully conciliated and integrated a variety of theoretical work (e.g. metacommunity and neutral theories, eco-phylogenetic, functional trait). However, this has made community ecology complex (numerous available indices, spatial and taxonomic scales importance, phylogenetic and functional relationships) and it raised warnings by several authors about its new fuzziness (Kraft *et al.*, 2015; Cadotte & Tucker, 2017; Gerhold *et al.*, 2015). This might be the call for a rethinking of community ecology concepts to reach a new synthesis of community ecology.





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# Glossary

**$\alpha$ -diversity** the diversity found within a spatial unit.

**$\beta$ -diversity** the diversity between two spatial units.

**$\gamma$ -diversity** the diversity over a group of spatial units. It combines the information of  $\alpha$  and  $\beta$ -diversity.

**assemblage** the set of taxonomically related species co-occurring in space and time, *i.e.* a subset of the community (Stroud *et al.*, 2015). Here, an assemblage is the co-occurring fish species in a locality. All the considered fish belong to the Teleosts, to the exception of the freshwater stingray which belongs to the Chondrichthyans.

**assembly rule** deterministic and neutral processes (competition, environmental filtering, historical contingency), that specify which subset of species in the total pool would form a community (Keddy, 1992).

**biodiversity** the variety of life, at all levels of organization, classified both by evolutionary (phylogenetic) and ecological (functional) criteria (Colwell, 2012).

**community** a group of interacting species (directly or indirectly), co-occurring in space and time (Stroud *et al.*, 2015).

**dispersal limitation** rare dispersal events prevents species establishment in suitable habitat (Shurin *et al.*, 2000). Dispersion can be limited due to species intrinsic capacities or due to the presence of barriers to the dispersion (mountain, river, sea).

**environmental filtering** environmental filtering occurs when a species arrives at a site but failed to establish or persist due to the local abiotic environment (Kraft *et al.*, 2014). The common hypothesis related is that assemblages are composed of species with similar trait values. Extended to multiple sites, environmental filtering occurs if sites have different environments that promote species with different ecological strategies.

**functional diversity** the diversity in the traits of species that influence their performance and thus ecosystem functioning (Díaz & Cabido, 2001).

**functional trait** any trait which impact fitness indirectly via its effects on growth, reproduction and survival (Violle *et al.*, 2007).

**limiting similarity** there is a limit to the similarity of competing species and to their number (MacArthur & Levins, 1967). The co-occurrence between species with

similar functions must thus be limited. The common hypothesis related is that assemblages are composed of species with divergent trait values.

**nestedness** one of the component of  $\beta$ -diversity. It represents the difference in species richness between two assemblages, where one assemblage is a subset of the other. Nestedness reflects selective extinction or colonization or nestedness of habitat (Baselga, 2007).

**phylogenetic diversity** the diversity in evolutionary history of species (Faith, 1992).

**phylogenetic signal** the tendency of related species to have similar ecology and phenotype (Losos, 2008).

**phylogenetic trait or niche conservatism** the extent to which species retain ancestral ecological traits and environmental distributions (Crisp *et al.*, 2009). Generally, closely related species have more similar ecology or phenotype than expected based on their phylogenetic relationships (Losos, 2008).

**species pool** a set of candidate species that could occur at smaller scale (Srivastava, 1999).

**taxonomic diversity** the diversity in species, with each species treated equally. One of the traditional measures of taxonomic diversity is the species richness (the number of species found in one spatial unit).

**trait convergence** species occurring in a particular set of environmental conditions tend to share similar functional and phenotypic traits (Pillar & Duarte, 2010).

**turnover** one of the component of  $\beta$ -diversity. It represents the change in species composition due to environmental sorting, dispersal limitation or speciation (Baselga, 2007).

## Bibliography

- Abascal, F., Zardoya, R. & Telford, M.J. (2010). TranslatorX: Multiple alignment of nucleotide sequences guided by amino acid translations. *Nucleic acids research*, **38**, W7–W13.
- Abell, R., Thieme, M.L., Revenga, C., Bryer, M., Kottelat, M., Bogutskaya, N., Coad, B., Mandrak, N., Balderas, S.C., Bussing, W. & others (2008). Freshwater ecoregions of the world: A new map of biogeographic units for freshwater biodiversity conservation. *BioScience*, **58**, 403–414.
- Adams, R.I., Amend, A.S., Taylor, J.W. & Bruns, T.D. (2013). A unique signal distorts the perception of species richness and composition in high-throughput sequencing surveys of microbial communities: A case study of fungi in indoor dust. *Microbial Ecology*, **66**, 735–741.
- Albanese, B., Angermeier, P.L. & Dorai-Raj, S. (2004). Ecological correlates of fish movement in a network of Virginia streams. *Canadian Journal of Fisheries and Aquatic Sciences*, **61**, 857–869.
- Albert, C.H., de Bello, F., Boulangeat, I., Pellet, G., Lavorel, S. & Thuiller, W. (2012). On the importance of intraspecific variability for the quantification of functional diversity. *Oikos*, **121**, 116–126.
- Aljanabi, S.M. & Martinez, I. (1997). Universal and rapid salt-extraction of high quality genomic DNA for PCR-based techniques. *Nucleic Acids Research*, **25**, 4692–4693.
- Allan, J.D., Abell, R., Hogan, Z., Revenga, C., Taylor, B.W., Welcomme, R.L. & Winemiller, K. (2005). Overfishing of inland waters. *BioScience*, **55**, 1041–1051.
- Allan, J.D. & Castillo, M.M. (2007). *Stream Ecology: Structure and Function of Running Waters*. Springer, New York, USA.
- Allard, L. (2014). *Elaboration d'un Indice de Qualité Des Eaux Basé Sur La Structure Taxonomique et Fonctionnelle Des Assemblages de Poissons Des Petits Cours d'eau de Guyane*. Ph.D. thesis, Université Toulouse III Paul Sabatier, Toulouse, France.
- Allard, L., Grenouillet, G., Khazraie, K., Tudesque, L., Vigouroux, R. & Brosse, S. (2014). Electrofishing efficiency in low conductivity Neotropical streams: Towards a non-destructive fish sampling method. *Fisheries Management and Ecology*, **21**, 234–243.
- Allard, L., Popée, M., Vigouroux, R. & Brosse, S. (2016). Effect of reduced impact logging and small-scale mining disturbances on Neotropical stream fish assemblages. *Aquatic Sciences*, **78**, 315–325.

- Altschul, S.F., Gish, W., Miller, W., Myers, E.W. & Lipman, D.J. (1990). Basic local alignment search tool. *Journal of Molecular Biology*, **215**, 403–410.
- Anderson, E.P. & Maldonado-Ocampo, J.A. (2011). A regional perspective on the diversity and conservation of tropical Andean fishes. *Conservation Biology*, **25**, 30–39.
- Anderson, M.J. (2001). A new method for non-parametric multivariate analysis of variance. *Austral Ecology*, **26**, 32–46.
- Anderson, M.J. (2006). Distance-based tests for homogeneity of multivariate dispersions. *Biometrics*, **62**, 245–253.
- Anderson, M.J., Crist, T.O., Chase, J.M., Vellend, M., Inouye, B.D., Freestone, A.L., Sanders, N.J., Cornell, H.V., Comita, L.S., Davies, K.F., Harrison, S.P., Kraft, N.J.B., Stegen, J.C. & Swenson, N.G. (2011). Navigating the multiple meanings of  $\beta$  diversity: A roadmap for the practicing ecologist. *Ecology Letters*, **14**, 19–28.
- Araújo, F.G., Pinto, B.C.T. & Teixeira, T.P. (2009). Longitudinal patterns of fish assemblages in a large tropical river in southeastern Brazil: Evaluating environmental influences and some concepts in river ecology. *Hydrobiologia*, **618**, 89–107.
- Ardura, A., Planes, S. & Garcia-Vazquez, E. (2013). Applications of DNA barcoding to fish landings: Authentication and diversity assessment. *ZooKeys*, **365**, 49–65.
- Aristotle (1910). *Historia Animalium*. Translated by D’Arcy Wentworth Thompson, Clarendon Press, Oxford, UK.
- Baer, S.G., Blair, J.M. & Collins, S.L. (2016). Environmental heterogeneity has a weak effect on diversity during community assembly in tallgrass prairie. *Ecological Monographs*, **86**, 94–106.
- Bahram, M., Kõljalg, U., Courty, P.E., Diédhiou, A.G., Kjøller, R., Põlme, S., Ryberg, M., Veldre, V. & Tedersoo, L. (2013). The distance decay of similarity in communities of ectomycorrhizal fungi in different ecosystems and scales. *Journal of Ecology*, **101**, 1335–1344.
- Baraloto, C., Hardy, O.J., Paine, C.E.T., Dexter, K.G., Cruaud, C., Dunning, L.T., Gonzalez, M.A., Molino, J.F., Sabatier, D., Savolainen, V. & Chave, J. (2012). Using functional traits and phylogenetic trees to examine the assembly of tropical tree communities. *Journal of Ecology*, **100**, 690–701.
- Barnes, M.A., Turner, C.R., Jerde, C.L., Renshaw, M.A., Chadderton, W.L. & Lodge, D.M. (2014). Environmental conditions influence eDNA persistence in aquatic systems. *Environmental Science & Technology*, **48**, 1819–1827.

- Barthélemy-Saint-Hilaire, J. (1883). Preface. In: *Histoire des animaux*. Hachette et cie, Paris, Œuvres d'Aristote.
- Barton, K. (2016). *MuMIn: Multi-Model Inference*. R package version 1.15.6. Available at <https://CRAN.R-project.org/package=MuMIn>.
- Baselga, A. (2007). Disentangling distance decay of similarity from richness gradients: Response to Soininen et al. 2007. *Ecography*, **30**, 838–841.
- Baselga, A. (2012). The relationship between species replacement, dissimilarity derived from nestedness, and nestedness. *Global Ecology and Biogeography*, **21**, 1223–1232.
- Baselga, A., Orme, D., Villegger, S., Bortoli, J.D. & Leprieur, F. (2013). *Betapart: Partitioning Beta Diversity into Turnover and Nestedness Components*. R package version 1.3. Available at <https://CRAN.R-project.org/package=betapart>.
- Bates, D., Mächler, M., Bolker, B. & Walker, S. (2015). Fitting linear mixed-effects models using lme4. *Journal of Statistical Software*, **67**, 1–48.
- Baur, H. & Leuenberger, C. (2011). Analysis of ratios in multivariate morphometry. *Systematic Biology*, **60**, 813–825.
- Bellemain, E., Patricio, H., Gray, T., Guegan, F., Valentini, A., Miaud, C. & Dejean, T. (2016). Trails of river monsters: Detecting critically endangered Mekong giant catfish *Pangasianodon gigas* using environmental DNA. *Global Ecology and Conservation*, **7**, 148–156.
- Bellwood, D.R., Goatley, C.H.R., Brandl, S.J. & Bellwood, O. (2014). Fifty million years of herbivory on coral reefs: Fossils, fish and functional innovations. *Proceedings of the Royal Society of London B: Biological Sciences*, **281**, 20133046.
- Belon, P. (1555). *La nature et diversité des poissons, avec leurs pourtraicts, représentez au plus près du naturel*. C. Estienne, Paris, France.
- Benson, D.A., Cavanaugh, M., Clark, K., Karsch-Mizrachi, I., Lipman, D.J., Ostell, J. & Sayers, E.W. (2017). GenBank. *Nucleic Acids Research*, **45**, D37–D42.
- Betancur-R, R., Broughton, R.E., Wiley, E.O., Carpenter, K., López, J.A., Li, C., Holcroft, N.I., Arcila, D., Sanciangco, M., Cureton li, J.C., Zhang, F., Buser, T., Campbell, M.A., Ballesteros, J.A., Roa-Varon, A., Willis, S., Borden, W.C., Rowley, T., Reneau, P.C., Hough, D.J., Lu, G., Grande, T., Arratia, G. & Ortí, G. (2013). The tree of life and a new classification of bony fishes. *PLoS currents*, **5**.
- Blanchet, F.G., Legendre, P. & Borcard, D. (2008). Forward selection of explanatory variables. *Ecology*, **89**, 2623–2632.

- Blanchet, S., Helmus, M.R., Brosse, S. & Grenouillet, G. (2014). Regional vs local drivers of phylogenetic and species diversity in stream fish communities. *Freshwater Biology*, **59**, 450–462.
- Blomberg, S.P., Garland, T. & Ives, A.R. (2003). Testing for phylogenetic signal in comparative data: Behavioral traits are more labile. *Evolution*, **57**, 717–745.
- Borcard, D. & Legendre, P. (2002). All-scale spatial analysis of ecological data by means of principal coordinates of neighbour matrices. *Ecological Modelling*, **153**, 51–68.
- Borcard, D., Legendre, P. & Drapeau, P. (1992). Partialling out the spatial component of ecological variation. *Ecology*, **73**, 1045–1055.
- Boujard, T., Pascal, M. & Meunier, F. (1990). Micro-répartition spatio-temporelle du peuplement ichthyologique d'un haut bassin fluvial de Guyane, l'Arataye. *Revue D'Ecologie-La Terre Et La Vie*, **45**, 357–373.
- Boujard, T. & Rojas-Beltran, R. (1988). Zonation longitudinale du peuplement ichthyologique du fleuve Sinnamary (Guyane Française). *Revue d'Hydrobiologie Tropicale*, **21**, 47–61.
- Boujard, T. & Tito de Morais, L. (1992). Biogéographie des poissons d'eau douce de Guyane française. In: *Evolution des littoraux de Guyane et de la zone caraïbe méridionale pendant le quaternaire* (eds. Prost, M.T. & Symposium PICG 274/ORSTOM, Cayenne (GUF), 1990/11/09-14). ORSTOM, Paris, Colloques et Séminaires, pp. 17–24.
- Boyer, F., Mercier, C., Bonin, A., Le Bras, Y., Taberlet, P. & Coissac, E. (2016). Obitools: A unix-inspired software package for DNA metabarcoding. *Molecular Ecology Resources*, **16**, 176–182.
- Boyle, K.S. & Horn, M.H. (2006). Comparison of feeding guild structure and ecomorphology of intertidal fish assemblages from central California and central Chile. *Marine Ecology Progress Series*, **319**, 65–84.
- Brandl, S.J., Robbins, W.D. & Bellwood, D.R. (2015). Exploring the nature of ecological specialization in a coral reef fish community: Morphology, diet and foraging microhabitat use. *Proceedings of the Royal Society of London B: Biological Sciences*, **282**, 20151147.
- Bray, J.R. & Curtis, J.T. (1957). An Ordination of the Upland Forest Communities of Southern Wisconsin. *Ecological Monographs*, **27**, 325–349.
- Brosse, S., Grenouillet, G., Gevrey, M., Khazraie, K. & Tudesque, L. (2011). Small-scale gold mining erodes fish assemblage structure in small neotropical streams. *Biodiversity and Conservation*, **20**, 1013–1026.

- Brosse, S. & Lek, S. (2000). Modelling roach (*Rutilus rutilus*) microhabitat using linear and nonlinear techniques. *Freshwater Biology*, **44**, 441–452.
- Brosse, S., Montoya-Burgos, J.I., Grenouillet, G. & Surugue, N. (2013). Determinants of fish assemblage structure in Mount Itoupé mountain streams (French Guiana). *Annales de Limnologie - International Journal of Limnology*, **49**, 43–49.
- Brown, E.A., Chain, F.J.J., Crease, T.J., Maclsaac, H.J. & Cristescu, M.E. (2015). Divergence thresholds and divergent biodiversity estimates: Can metabarcoding reliably describe zooplankton communities? *Ecology and Evolution*, **5**, 2234–2251.
- Buffon Comte de, G.L.L. (1761). *Histoire Naturelle Générale et Particulière*. Imprimerie Royale, Paris, France.
- Burnham, K.P. & Anderson, D.R. (2002). *Model Selection and Multimodel Inference: A Practical Information-Theoretic Approach*. Springer-Verlag, New York, USA.
- Bush, A., Sollmann, R., Wilting, A., Bohmann, K., Cole, B., Balzter, H., Martius, C., Zlinszky, A., Calvignac-Spencer, S., Cobbold, C.A., Dawson, T.P., Emerson, B.C., Ferrier, S., Gilbert, M.T.P., Herold, M., Jones, L., Leendertz, F.H., Matthews, L., Millington, J.D.A., Olson, J.R., Ovaskainen, O., Raffaelli, D., Reeve, R., Rödel, M.O., Rodgers, T.W., Snape, S., Visseren-Hamakers, I., Vogler, A.P., White, P.C.L., Wooster, M.J. & Yu, D.W. (2017). Connecting Earth observation to high-throughput biodiversity data. *Nature Ecology & Evolution*, **1**, 176.
- Cadotte, M.W., Carscadden, K. & Mirotchnick, N. (2011). Beyond species: Functional diversity and the maintenance of ecological processes and services. *Journal of Applied Ecology*, **48**, 1079–1087.
- Cadotte, M.W. & Tucker, C.M. (2017). Should Environmental Filtering be Abandoned? *Trends in Ecology & Evolution*, **32**, 429–437.
- Cailleux, A. (1954). Limites dimensionnelles et noms des fractions granulométriques. *Bulletin de la Société Géologique de France*, **S6-IV**, 643–646.
- Cardoso, Y.P. & Montoya-Burgos, J.I. (2009). Unexpected diversity in the catfish *Pseudancistrus brevispinis* reveals dispersal routes in a Neotropical center of endemism: The Guyanas Region. *Molecular Ecology*, **18**, 947–964.
- Carmona, C.P., de Bello, F., Mason, N.W.H. & Lepš, J. (2016). Traits without borders: Integrating functional diversity across scales. *Trends in Ecology & Evolution*, **31**, 382–394.
- Castresana, J. (2000). Selection of conserved blocks from multiple alignments for their use in phylogenetic analysis. *Molecular biology and evolution*, **17**, 540–552.
- Cavender-Bares, J., Ackerly, D.D., Baum, D.A. & Bazzaz, F.A. (2004). Phylogenetic overdispersion in Floridian oak communities. *The American Naturalist*, **163**, 823–843.

- Cilleros, K., Allard, L., Grenouillet, G. & Brosse, S. (2016). Taxonomic and functional diversity patterns reveal different processes shaping European and Amazonian stream fish assemblages. *Journal of Biogeography*, **43**, 1832–1843.
- Civade, R., Dejean, T., Valentini, A., Roset, N., Raymond, J.C., Bonin, A., Taberlet, P. & Pont, D. (2016). Spatial representativeness of environmental DNA metabarcoding signal for fish biodiversity assessment in a natural freshwater system. *PLOS ONE*, **11**, e0157366.
- Colwell, R.K. (2012). Biodiversity: Concepts, patterns and measurement. In: *The Princeton Guide to Ecology* (ed. Levin, S.). Princeton University Press, Princeton, NJ, USA, pp. 257–263.
- Colwell, R.K., Gotelli, N.J., Ashton, L.A., Beck, J., Brehm, G., Fayle, T.M., Fiedler, K., Forister, M.L., Kessler, M., Kitching, R.L., Klimes, P., Kluge, J., Longino, J.T., Maunsell, S.C., McCain, C.M., Moses, J., Noben, S., Sam, K., Sam, L., Shapiro, A.M., Wang, X. & Novotny, V. (2016). Midpoint attractors and species richness: Modelling the interaction between environmental drivers and geometric constraints. *Ecology Letters*, **19**, 1009–1022.
- Colwell, R.K. & Hurtt, G.C. (1994). Nonbiological Gradients in Species Richness and a Spurious Rapoport Effect. *The American Naturalist*, **144**, 570–595.
- Connell, J.H. (1971). On the role of natural enemies in preventing competitive exclusion in some marine animals and in rain forest trees. In: *Dynamics of Populations* (eds. den Boer, P.J. & Gradwell, G.R.). Centre for Agricultural Publishing and Documentation, Wageningen, The Netherlands, pp. 298–312.
- Cornwell, W.K., Schwilk, D.W. & Ackerly, D.D. (2006). A trait-based test for habitat filtering: Convex hull volume. *Ecology*, **87**, 1465–1471.
- Crisp, M.D., Arroyo, M.T.K., Cook, L.G., Gandolfo, M.A., Jordan, G.J., McGlone, M.S., Weston, P.H., Westoby, M., Wilf, P. & Linder, H.P. (2009). Phylogenetic biome conservatism on a global scale. *Nature*, **458**, 754–756.
- Crisp, M.D., Trewick, S.A. & Cook, L.G. (2011). Hypothesis testing in biogeography. *Trends in ecology & evolution*, **26**, 66–72.
- Currie, D.J., Mittelbach, G.G., Cornell, H.V., Field, R., Guégan, J.F., Hawkins, B.A., Kaufman, D.M., Kerr, J.T., Oberdorff, T., O'Brien, E. & others (2004). Predictions and tests of climate-based hypotheses of broad-scale variation in taxonomic richness. *Ecology letters*, **7**, 1121–1134.
- Dahl, C., Novotny, V., Moravec, J. & Richards, S.J. (2009). Beta diversity of frogs in the forests of New Guinea, Amazonia and Europe: Contrasting tropical and temperate communities. *Journal of Biogeography*, **36**, 896–904.

- Dalu, T., Wasserman, R.J., Jordaan, M., Froneman, W.P. & Weyl, O.L.F. (2015). An assessment of the effect of rotenone on selected non-target aquatic fauna. *PLOS ONE*, **10**, e0142140.
- Datry, T., Melo, A.S., Moya, N., Zubieta, J., De la Barra, E. & Oberdorff, T. (2016). Metacommunity patterns across three Neotropical catchments with varying environmental harshness. *Freshwater Biology*, **61**, 277–292.
- Davidar, P., Rajagopal, B., Mohandass, D., Puyravaud, J.P., Condit, R., Wright, S.J. & Leigh, E.G. (2007). The effect of climatic gradients, topographic variation and species traits on the beta diversity of rain forest trees. *Global Ecology and Biogeography*, **16**, 510–518.
- Davies, T.J., Kraft, N.J.B., Salamin, N. & Wolkovich, E.M. (2012). Incompletely resolved phylogenetic trees inflate estimates of phylogenetic conservatism. *Ecology*, **93**, 242–247.
- De Barba, M., Miquel, C., Boyer, F., Mercier, C., Rioux, D., Coissac, E. & Taberlet, P. (2014). DNA metabarcoding multiplexing and validation of data accuracy for diet assessment: Application to omnivorous diet. *Molecular Ecology Resources*, **14**, 306–323.
- de Mérona, B., Tejerina-Garro, F.L. & Vigouroux, R. (2012). Fish-habitat relationships in French Guiana rivers : A review. *Cybium*, **36**, 7–15.
- de Mérona, B., Vigouroux, R. & Tejerina-Garro, F.L. (2005). Alteration of fish diversity downstream from Petit-Saut Dam in French Guiana. Implication of ecological strategies of fish species. *Hydrobiologia*, **551**, 33–47.
- Declerck, S.A.J., Coronel, J.S., Legendre, P. & Brendonck, L. (2011). Scale dependency of processes structuring metacommunities of cladocerans in temporary pools of High-Andes wetlands. *Ecography*, **34**, 296–305.
- Dedieu, N. (2014). *Mise au point d'un outil d'évaluation de la qualité biologique des petites rivières de Guyane sur la base des invertébrés benthiques*. Ph.D. thesis, Université Toulouse III Paul Sabatier, Toulouse, France.
- Dedieu, N., Allard, L., Vigouroux, R., Brosse, S. & Céréghino, R. (2014). Physical habitat and water chemistry changes induced by logging and gold mining in French Guiana streams. *Knowledge and Management of Aquatic Ecosystems*, **415**, 02.
- Dedieu, N., Rhone, M., Vigouroux, R. & Céréghino, R. (2015). Assessing the impact of gold mining in headwater streams of Eastern Amazonia using Ephemeroptera assemblages and biological traits. *Ecological Indicators*, **52**, 332–340.
- Deiner, K. & Altermatt, F. (2014). Transport distance of invertebrate environmental DNA in a natural river. *PLOS ONE*, **9**, e88786.

- Deiner, K., Fronhofer, E.A., Mächler, E., Walser, J.C. & Altermatt, F. (2016). Environmental DNA reveals that rivers are conveyor belts of biodiversity information. *Nature Communications*, **7**, 12544.
- Delacoste, M., Baran, P., Lek, S. & Lascaux, J.M. (1995). Classification et clé de détermination des faciès d'écoulements en rivières de montagne. *Bulletin Français de la Pêche et de la Pisciculture*, pp. 149–156.
- Devictor, V., Mouillot, D., Meynard, C., Jiguet, F., Thuiller, W. & Mouquet, N. (2010). Spatial mismatch and congruence between taxonomic, phylogenetic and functional diversity: The need for integrative conservation strategies in a changing world. *Ecology Letters*, **13**, 1030–1040.
- Dias, M.S., Magnusson, W.E. & Zuanon, J. (2010). Effects of reduced-impact logging on fish assemblages in Central Amazonia. *Conservation Biology*, **24**, 278–286.
- Dias, M.S., Oberdorff, T., Hugueny, B., Leprieur, F., Jézéquel, C., Cornu, J.F., Brosse, S., Grenouillet, G. & Tedesco, P.A. (2014). Global imprint of historical connectivity on freshwater fish biodiversity. *Ecology Letters*, **17**, 1130–1140.
- Diáz, S. & Cabido, M. (2001). Vive la différence: Plant functional diversity matters to ecosystem processes. *Trends in Ecology & Evolution*, **16**, 646–655.
- Dinno, A. (2017). *Dunn.Test: Dunn's Test of Multiple Comparisons Using Rank Sums*. R package version 1.3.4. Available at <https://CRAN.R-project.org/package=dunn.test>.
- Dodds, W.K., Gido, K., Whiles, M.R., Daniels, M.D. & Grudzinski, B.P. (2015). The Stream Biome Gradient Concept: Factors controlling lotic systems across broad biogeographic scales. *Freshwater Science*, **34**, 1–19.
- Dolnicar, S., Grabler, K. & Mazanec, J. (2000). A tale of three cities: Perceptual charting for analysing destination images. In: *Consumer Psychology of Tourism, Hospitality and Leisure* (eds. Woodside, A.G., Crouch, G.I., Mazanec, J.A., Oppermann, M. & Sakai, M.Y.). CAB International, New York, USA, pp. 39–62.
- Downes, B.J., Lake, P.S., Schreiber, E.S.G. & Glaister, A. (1998). Habitat structure and regulation of local species diversity in a stony, upland stream. *Ecological Monographs*, **68**, 237–257.
- Dray, S., Blanchet, G., Borcard, D., Guenard, G., Jombart, T., Larocque, G., Legendre, P., Madi, N. & Wagner, H.H. (2017). *Adespatial: Multivariate Multiscale Spatial Analysis*. R package version 0.0-8. Available at.
- Dray, S., Legendre, P. & Peres-Neto, P.R. (2006). Spatial modelling: A comprehensive framework for principal coordinate analysis of neighbour matrices (PCNM). *Ecological Modelling*, **196**, 483–493.

- Dumay, O., Tari, P.S., Tomasini, J.A. & Mouillot, D. (2004). Functional groups of lagoon fish species in Languedoc Roussillon, southern France. *Journal of Fish Biology*, **64**, 970–983.
- Edgar, R.C. (2010). Search and clustering orders of magnitude faster than BLAST. *Bioinformatics*, **26**, 2460–2461.
- Elith, J. & Leathwick, J.R. (2009). Species distribution models: Ecological explanation and prediction across space and time. *Annual Review of Ecology, Evolution, and Systematics*, **40**, 677–697.
- Ellender, B.R., Becker, A., Weyl, O.L.F. & Swartz, E.R. (2012). Underwater video analysis as a non-destructive alternative to electrofishing for sampling imperilled headwater stream fishes. *Aquatic Conservation: Marine and Freshwater Ecosystems*, **22**, 58–65.
- Elton, C. (1946). Competition and the Structure of Ecological Communities. *Journal of Animal Ecology*, **15**, 54–68.
- Engler, A. (1879). *Versuch einer Entwicklungsgeschichte der Pflanzenwelt, insbesondere der Florengebiete seit der Tertiärperiode: Die extratropischen Gebiete der nördlichen Hemisphäre*. Wilhelm Engelmann, Leipzig, Germany.
- Faith, D.P. (1992). Conservation evaluation and phylogenetic diversity. *Biological Conservation*, **61**, 1–10.
- Farris, J.S. (1969). On the cophenetic correlation coefficient. *Systematic Biology*, **18**, 279–285.
- Fausch, K.D. (1993). Experimental analysis of microhabitat selection by juvenile steelhead (*Oncorhynchus mykiss*) and coho salmon (*O. kisutch*) in a British Columbia stream. *Canadian Journal of Fisheries and Aquatic Sciences*, **50**, 1198–1207.
- Fayle, T.M., Eggleton, P., Manica, A., Yusah, K.M. & Foster, W.A. (2015). Experimentally testing and assessing the predictive power of species assembly rules for tropical canopy ants. *Ecology Letters*, **18**, 254–262.
- Fernandes, C.C., Podos, J. & Lundberg, J.G. (2004). Amazonian ecology: Tributaries enhance the diversity of electric fishes. *Science*, **305**, 1960–1962.
- Ficetola, G.F., Pansu, J., Bonin, A., Coissac, E., Giguet-Covex, C., De Barba, M., Gielly, L., Lopes, C.M., Boyer, F., Pompanon, F., Rayé, G. & Taberlet, P. (2015). Replication levels, false presences and the estimation of the presence/absence from eDNA metabarcoding data. *Molecular Ecology Resources*, **15**, 543–556.

## BIBLIOGRAPHY

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- Finlayson, B.J., Schnick, R.A., Cailteux, R.L., DeMong, L., Horton, W.D., McClay, W., Thompson, C.W. & Tichacek, G. (2000). *Rotenone Use in Fisheries Management: Administrative and Technical Guidelines Manual*. American Fisheries Society, Bethesda, MD, USA.
- Fischer, A.G. (1960). Latitudinal variations in organic diversity. *Evolution*, **14**, 64–81.
- Flynn, J.M., Brown, E.A., Chain, F.J.J., Maclsaac, H.J. & Cristescu, M.E. (2015). Toward accurate molecular identification of species in complex environmental samples: Testing the performance of sequence filtering and clustering methods. *Ecology and Evolution*, **5**, 2252–2266.
- Fortunel, C., Paine, C.E.T., Fine, P.V.A., Kraft, N.J.B. & Baraloto, C. (2014). Environmental factors predict community functional composition in Amazonian forests. *Journal of Ecology*, **102**, 145–155.
- Freilich, M.A. & Connolly, S.R. (2015). Phylogenetic community structure when competition and environmental filtering determine abundances. *Global Ecology and Biogeography*, **24**, 1390–1400.
- Froese, R. & Pauly, D. (2015). FishBase. World Wide Web electronic publication. Version 10/2015. Available at <http://www.fishbase.org>.
- Fukami, T. (2015). Historical contingency in community assembly: Integrating niches, species pools, and priority effects. *Annual Review of Ecology, Evolution, and Systematics*, **46**, 1–23.
- Fukami, T., Martijn Bezemer, T., Mortimer, S.R. & van der Putten, W.H. (2005). Species divergence and trait convergence in experimental plant community assembly. *Ecology Letters*, **8**, 1283–1290.
- Fukumoto, S., Ushimaru, A. & Minamoto, T. (2015). A basin-scale application of environmental DNA assessment for rare endemic species and closely related exotic species in rivers: A case study of giant salamanders in Japan. *Journal of Applied Ecology*, **52**, 358–365.
- Fulton, C., Bellwood, D. & Wainwright, P. (2001). The relationship between swimming ability and habitat use in wrasses (Labridae). *Marine Biology*, **139**, 25–33.
- Garzon-Lopez, C.X., Jansen, P.A., Bohlman, S.A., Ordonez, A. & Olf, H. (2014). Effects of sampling scale on patterns of habitat association in tropical trees. *Journal of Vegetation Science*, **25**, 349–362.
- Gaston, K. & Blackburn, T. (2008). *Pattern and Process in Macroecology*. Blackwell Science, Oxford, UK.
- Gaston, K.J. & Blackburn, T.M. (1996). The tropics as a museum of biological diversity: An analysis of the New World avifauna. *Proceedings of the Royal Society of London B: Biological Sciences*, **263**, 63–68.

- Gatz Jr., A. (1968). Ecological morphology of freshwater stream fishes. *Tulane Studies in Zoology and Botany*, **21**, 91–124.
- Gerhold, P., Cahill, J.F., Winter, M., Bartish, I.V. & Prinzing, A. (2015). Phylogenetic patterns are not proxies of community assembly mechanisms (they are far better). *Functional Ecology*, **29**, 600–614.
- Gorman, O.T. & Karr, J.R. (1978). Habitat structure and stream fish communities. *Ecology*, **59**, 507–515.
- Gotelli, N.J. & Colwell, R.K. (2001). Quantifying biodiversity: Procedures and pitfalls in the measurement and comparison of species richness. *Ecology Letters*, **4**, 379–391.
- Gotelli, N.J. & Graves, G.R. (1996). *Null Models in Ecology*. Smithsonian Institution Press, Washington, DC, USA.
- Graham, C.H. & Fine, P.V.A. (2008). Phylogenetic beta diversity: Linking ecological and evolutionary processes across space in time. *Ecology Letters*, **11**, 1265–1277.
- Grassle, J.F. (2000). The Ocean Biogeographic Information System (OBIS): An on-line, worldwide atlas for accessing, modeling and mapping marine biological data in a multidimensional geographic context. *Oceanography*, **13**, 5–7.
- Griffiths, D. (2006). Pattern and process in the ecological biogeography of European freshwater fish. *Journal of Animal Ecology*, **75**, 734–751.
- Grossman, G.D., Moyle, P.B. & Whitaker, J.O. (1982). Stochasticity in structural and functional characteristics of an indiana stream fish assemblage: A test of community theory. *The American Naturalist*, **120**, 423–454.
- Grossman, G.D., Ratajczak, R.E., Crawford, M. & Freeman, M.C. (1998). Assemblage organization in stream fishes: Effects of environmental variation and interspecific interactions. *Ecological Monographs*, **68**, 395–420.
- Grossman, G.D., Ratajczak, R.E., Petty, J.T., Hunter, M.D., Peterson, J.T. & Grenouillet, G. (2006). Population dynamics of mottled sculpin (pisces) in a variable environment: Information theoretic approaches. *Ecological Monographs*, **76**, 217–234.
- Grossman, G.D., Ratajczak Jr, R.E., Farr, M.D., Wagner, C.M. & Petty, J.T. (2010). Why there are fewer fish upstream. In: *Community Ecology of Stream Fishes: Concepts, Approaches, and Techniques*. American Fisheries Society, Symposium. vol. 73, pp. 63–81.
- Gunzburger, M.S. (2007). Evaluation of seven aquatic sampling methods for amphibians and other aquatic fauna. *Applied Herpetology*, **4**, 47–63.
- Hammond, D.S., Gond, V., de Thoisy, B., Forget, P.M. & DeDijn, B.P.E. (2007). Causes and Consequences of a Tropical Forest Gold Rush in the Guiana Shield, South America. *AMBIO: A Journal of the Human Environment*, **36**, 661–670.

## BIBLIOGRAPHY

---

- Hänfling, B., Lawson Handley, L., Read, D.S., Hahn, C., Li, J., Nichols, P., Blackman, R.C., Oliver, A. & Winfield, I.J. (2016). Environmental DNA metabarcoding of lake fish communities reflects long-term data from established survey methods. *Molecular Ecology*, **25**, 3101–3119.
- Hawkins, B.A. & Porter, E.E. (2003). Relative influences of current and historical factors on mammal and bird diversity patterns in deglaciated North America. *Global Ecology and Biogeography*, **12**, 475–481.
- Hercos, A.P., Sobansky, M., Queiroz, H.L. & Magurran, A.E. (2013). Local and regional rarity in a diverse tropical fish assemblage. *Proceedings of the Royal Society of London B: Biological Sciences*, **280**, 20122076.
- Hickey, M.A. & Closs, G.P. (2006). Evaluating the potential of night spotlighting as a method for assessing species composition and brown trout abundance: A comparison with electrofishing in small streams. *Journal of Fish Biology*, **69**, 1513–1523.
- Hillebrand, H. (2004). On the Generality of the Latitudinal Diversity Gradient. *The American Naturalist*, **163**, 192–211.
- HilleRisLambers, J., Adler, P., Harpole, W., Levine, J. & Mayfield, M. (2012). Rethinking community assembly through the lens of coexistence theory. *Annual Review of Ecology, Evolution, and Systematics*, **43**, 227–248.
- Hoeinghaus, D.J., Winemiller, K.O. & Birnbaum, J.S. (2007). Local and regional determinants of stream fish assemblage structure: Inferences based on taxonomic vs. functional groups. *Journal of Biogeography*, **34**, 324–338.
- Hof, C., Brändle, M. & Brandl, R. (2008). Latitudinal variation of diversity in European freshwater animals is not concordant across habitat types. *Global Ecology and Biogeography*, **17**, 539–546.
- Hubbell, S.P. (2011). *The Unified Neutral Theory of Biodiversity and Biogeography*. Princeton University Press, Princeton, NJ, USA.
- Hubert, N. & Renno, J.F. (2006). Historical biogeography of South American freshwater fishes. *Journal of Biogeography*, **33**, 1414–1436.
- Huet, M. (1959). Profiles and biology of western European streams as related to fish management. *Transactions of the American Fisheries Society*, **88**, 155–163.
- Hugueny, B. (1989). West African rivers as biogeographic islands: Species richness of fish communities. *Oecologia*, **79**, 236–243.
- Hurlbert, A.H. & Stegen, J.C. (2014). When should species richness be energy limited, and how would we know? *Ecology Letters*, **17**, 401–413.

- Hutchinson, G.E. (1957). Concluding remarks. *Cold Spring Harbor Symposia on Quantitative Biology*, **22**, 415–427.
- Hutchinson, G.E. (1959). Homage to Santa Rosalia or Why Are There So Many Kinds of Animals? *The American Naturalist*, **93**, 145–159.
- Ibarra, A.A., Park, Y.S., Brosse, S., Reyjol, Y., Lim, P. & Lek, S. (2005). Nested patterns of spatial diversity revealed for fish assemblages in a west European river. *Ecology of Freshwater Fish*, **14**, 233–242.
- Jaccard, P. (1912). The distribution of the flora in the Alpine zone. *New Phytologist*, **11**, 37–50.
- Jackson, D.A., Peres-Neto, P.R. & Olden, J.D. (2001). What controls who is where in freshwater fish communities: The roles of biotic, abiotic, and spatial factors. *Canadian Journal of Fisheries and Aquatic Sciences*, **58**, 157–170.
- Jane, S.F., Wilcox, T.M., McKelvey, K.S., Young, M.K., Schwartz, M.K., Lowe, W.H., Letcher, B.H. & Whiteley, A.R. (2015). Distance, flow and PCR inhibition: eDNA dynamics in two headwater streams. *Molecular Ecology Resources*, **15**, 216–227.
- Janzen, D.H. (1970). Herbivores and the number of tree species in tropical forests. *The American Naturalist*, **104**, 501–528.
- Jaramillo-Villa, U., Maldonado-Ocampo, J.A. & Escobar, F. (2010). Altitudinal variation in fish assemblage diversity in streams of the central Andes of Colombia. *Journal of Fish Biology*, **76**, 2401–2417.
- Jarvinen, O. (1982). Species-To-Genus Ratios in Biogeography: A Historical Note. *Journal of Biogeography*, **9**, 363–370.

- Jarvis, E.D., Mirarab, S., Aberer, A.J., Li, B., Houde, P., Li, C., Ho, S.Y.W., Faircloth, B.C., Nabholz, B., Howard, J.T., Suh, A., Weber, C.C., da Fonseca, R.R., Li, J., Zhang, F., Li, H., Zhou, L., Narula, N., Liu, L., Ganapathy, G., Boussau, B., Bayzid, M.S., Zavidovych, V., Subramanian, S., Gabaldón, T., Capella-Gutiérrez, S., Huerta-Cepas, J., Rekepalli, B., Munch, K., Schierup, M., Lindow, B., Warren, W.C., Ray, D., Green, R.E., Bruford, M.W., Zhan, X., Dixon, A., Li, S., Li, N., Huang, Y., Derryberry, E.P., Bertelsen, M.F., Sheldon, F.H., Brumfield, R.T., Mello, C.V., Lovell, P.V., Wirthlin, M., Schneider, M.P.C., Prosdociimi, F., Samaniego, J.A., Velazquez, A.M.V., Alfaro-Núñez, A., Campos, P.F., Petersen, B., Sicheritz-Ponten, T., Pas, A., Bailey, T., Scofield, P., Bunce, M., Lambert, D.M., Zhou, Q., Perelman, P., Driskell, A.C., Shapiro, B., Xiong, Z., Zeng, Y., Liu, S., Li, Z., Liu, B., Wu, K., Xiao, J., Yinqi, X., Zheng, Q., Zhang, Y., Yang, H., Wang, J., Smeds, L., Rheindt, F.E., Braun, M., Fjeldsa, J., Orlando, L., Barker, F.K., Jönsson, K.A., Johnson, W., Koepfli, K.P., O'Brien, S., Haussler, D., Ryder, O.A., Rahbek, C., Willerslev, E., Graves, G.R., Glenn, T.C., McCormack, J., Burt, D., Ellegren, H., Alström, P., Edwards, S.V., Stamatakis, A., Mindell, D.P., Cracraft, J., Braun, E.L., Warnow, T., Jun, W., Gilbert, M.T.P. & Zhang, G. (2014). Whole-genome analyses resolve early branches in the tree of life of modern birds. *Science*, **346**, 1320–1331.
- Jerde, C.L., Mahon, A.R., Chadderton, W.L. & Lodge, D.M. (2011). “Sight-unseen” detection of rare aquatic species using environmental DNA. *Conservation Letters*, **4**, 150–157.
- Johnson, R.K., Furse, M.T., Hering, D. & Sandin, L. (2007). Ecological relationships between stream communities and spatial scale: Implications for designing catchment-level monitoring programmes. *Freshwater Biology*, **52**, 939–958.
- Junk, W.J., Soares, M.G.M. & Bayley, P.B. (2007). Freshwater fishes of the Amazon River basin: Their biodiversity, fisheries, and habitats. *Aquatic Ecosystem Health & Management*, **10**, 153–173.
- Jurasinski, G. & Retzer, V. (2012). *Simba: A Collection of Functions for Similarity Analysis of Vegetation*. R package version 0.3-5. Available at <http://CRAN.R-project.org/package=simba>.
- Kappas, I., Vittas, S., Pantzartzi, C.N., Drosopoulou, E. & Scouras, Z.G. (2016). A Time-Calibrated Mitogenome Phylogeny of Catfish (Teleostei: Siluriformes). *PLOS ONE*, **11**, e0166988.
- Katoh, K. & Standley, D.M. (2013). MAFFT multiple sequence alignment software version 7: Improvements in performance and usability. *Molecular biology and evolution*, **30**, 772–780.

- Kearse, M., Moir, R., Wilson, A., Stones-Havas, S., Cheung, M., Sturrock, S., Buxton, S., Cooper, A., Markowitz, S., Duran, C., Thierer, T., Ashton, B., Meintjes, P. & Drummond, A. (2012). Geneious basic: An integrated and extendable desktop software platform for the organization and analysis of sequence data. *Bioinformatics*, **28**, 1647–1649.
- Keddy, P.A. (1992). Assembly and response rules: Two goals for predictive community ecology. *Journal of Vegetation Science*, **3**, 157–164.
- Keith, P., Le Bail, P.Y. & Planquette, P. (2000). *Atlas des Poissons d'eau douce de Guyane. Tome 2, fascicule 1. Batrachoidiformes, Mugiliformes, Beloniformes, Cyprinodontiformes, Synbranchiformes, Perciformes, Pleuronectiformes, Tetraodontiformes*. vol. 43(I) of *Collection du patrimoine naturel*. M.N.H.N./S.P.N., Paris, France.
- Kelly, R.P., Port, J.A., Yamahara, K.M. & Crowder, L.B. (2014). Using environmental DNA to census marine fishes in a large mesocosm. *PLOS ONE*, **9**, e86175.
- Koeck, B., Tessier, A., Brind'Amour, A., Pastor, J., Bijaoui, B., Dalias, N., Astruch, P., Saragoni, G. & Lenfant, P. (2014). Functional differences between fish communities on artificial and natural reefs: A case study along the French Catalan coast. *Aquatic Biology*, **20**, 219–234.
- Koleff, P., Gaston, K.J. & Lennon, J.J. (2003). Measuring beta diversity for presence–absence data. *Journal of Animal Ecology*, **72**, 367–382.
- Kotlík, P. & Berrebi, P. (2001). Phylogeography of the barbel (*Barbus barbus*) assessed by mitochondrial DNA variation. *Molecular Ecology*, **10**, 2177–2185.
- Kovalenko, K.E., Thomaz, S.M. & Warfe, D.M. (2011). Habitat complexity: Approaches and future directions. *Hydrobiologia*, **685**, 1–17.
- Kraft, N.J.B. & Ackerly, D.D. (2010). Functional trait and phylogenetic tests of community assembly across spatial scales in an Amazonian forest. *Ecological Monographs*, **80**, 401–422.
- Kraft, N.J.B., Adler, P.B., Godoy, O., James, E.C., Fuller, S. & Levine, J.M. (2015). Community assembly, coexistence and the environmental filtering metaphor. *Functional Ecology*, **29**, 592–599.
- Kraft, N.J.B., Comita, L.S., Chase, J.M., Sanders, N.J., Swenson, N.G., Crist, T.O., Stegen, J.C., Vellend, M., Boyle, B., Anderson, M.J., Cornell, H.V., Davies, K.F., Freestone, A.L., Inouye, B.D., Harrison, S.P. & Myers, J.A. (2011). Disentangling the drivers of  $\beta$ -diversity along latitudinal and elevational gradients. *Science*, **333**, 1755–1758.

- Kraft, N.J.B., Crutsinger, G.M., Forrestel, E.J. & Emery, N.C. (2014). Functional trait differences and the outcome of community assembly: An experimental test with vernal pool annual plants. *Oikos*, **123**, 1391–1399.
- Kraft, N.J.B., Valencia, R. & Ackerly, D.D. (2008). Functional traits and niche-based tree community assembly in an Amazonian forest. *Science*, **322**, 580–582.
- Lamanna, C., Blonder, B., Violle, C., Kraft, N.J.B., Sandel, B., Šímová, I., Donoghue, J.C., Svenning, J.C., McGill, B.J., Boyle, B., Buzzard, V., Dolins, S., Jørgensen, P.M., Marcuse-Kubitza, A., Morueta-Holme, N., Peet, R.K., Piel, W.H., Regetz, J., Schildhauer, M., Spencer, N., Thiers, B., Wisser, S.K. & Enquist, B.J. (2014). Functional trait space and the latitudinal diversity gradient. *Proceedings of the National Academy of Sciences*, **111**, 13745–13750.
- Landeiro, V.L., Magnusson, W.E., Melo, A.S., Espírito-Santo, H.M.V. & Bini, L.M. (2011). Spatial eigenfunction analyses in stream networks: Do watercourse and overland distances produce different results? *Freshwater Biology*, **56**, 1184–1192.
- Lanfear, R., Frandsen, P.B., Wright, A.M., Senfeld, T. & Calcott, B. (2016). PartitionFinder 2: New methods for selecting partitioned models of evolution for molecular and morphological phylogenetic analyses. *Molecular biology and evolution*, **34**, 772–773.
- Lawes, M.J., Eeley, H.A.C., Findlay, N.J. & Forbes, D. (2007). Resilient forest faunal communities in South Africa: A legacy of palaeoclimatic change and extinction filtering? *Journal of Biogeography*, **34**, 1246–1264.
- Le Bail, P.Y., Covain, R., Jegu, M., Fisch-Muller, S., Vigouroux, R. & Keith, P. (2012). Updated checklist of the freshwater and estuarine fishes of French Guiana. *Cybium*, **36**, 293–319.
- Le Bail, P.Y., Keith, P. & Planquette, P. (2000). *Atlas Des Poissons d'eau Douce de Guyane. Tome 2, Fascicule II : Siluriformes*. vol. 43(II) of *Collection du patrimoine naturel*. M.N.H.N./S.P.N., Paris, France.
- Lefcheck, J.S., Buchheister, A., Laumann, K.M., Stratton, M.A., Sobocinski, K.L., Chak, S.T.C., Clardy, T.R., Reynolds, P.L., Latour, R.J. & Duffy, J.E. (2014). Dimensions of biodiversity in Chesapeake Bay demersal fishes: Patterns and drivers through space and time. *Ecosphere*, **5**, 1–48.
- Legendre, P. & Legendre, L. (2012). *Numerical Ecology*. Developments in Environmental Modelling. Elsevier Science, Amsterdam, The Netherlands.
- Leitão, R.P., Zuanon, J., Mouillot, D., Leal, C.G., Hughes, R.M., Kaufmann, P.R., Villéger, S., Pompeu, P.S., Kasper, D., de Paula, F.R., Ferraz, S.F.B. & Gardner, T.A. (2017). Disentangling the pathways of land use impacts on the functional structure of fish assemblages in Amazon streams. *Ecography*, p. doi:10.1111/ecog.02845.

- Leitão, R.P., Zuanon, J., Villéger, S., Williams, S.E., Baraloto, C., Fortunel, C., Mendonça, F.P. & Mouillot, D. (2016). Rare species contribute disproportionately to the functional structure of species assemblages. *Proceedings of the Royal Society of London B: Biological Sciences*, **283**, 20160084.
- Leprieur, F., Albouy, C., De Bortoli, J., Cowman, P.F., Bellwood, D.R. & Mouillot, D. (2012). Quantifying phylogenetic beta diversity: Distinguishing between 'true' turnover of lineages and phylogenetic diversity gradients. *PLoS ONE*, **7**, e42760.
- Lepš, J. & Šmilauer, P. (2003). *Multivariate Analysis of Ecological Data Using CANOCO*. Cambridge University Press, Cambridge, UK.
- Lévêque, C., Oberdorff, T., Paugy, D., Stiassny, M.L.J. & Tedesco, P.A. (2007). Global diversity of fish (Pisces) in freshwater. *Hydrobiologia*, **595**, 545–567.
- Lindahl, P.E. & Öberg, K.E. (1961). The effect of rotenone on respiration and its point of attack. *Experimental Cell Research*, **23**, 228–237.
- Lopes, C.M., Sasso, T., Valentini, A., Dejean, T., Martins, M., Zamudio, K.R. & Haddad, C.F.B. (2017). eDNA metabarcoding: A promising method for anuran surveys in highly diverse tropical forests. *Molecular Ecology Resources*, pp. doi:10.1111/1755–0998.12643.
- Loranger, J., Violle, C., Shipley, B., Lavorel, S., Bonis, A., Cruz, P., Louault, F., Loucougaray, G., Mesléard, F., Yavercovski, N. & Garnier, É. (2016). Recasting the dynamic equilibrium model through a functional lens: The interplay of trait-based community assembly and climate. *Journal of Ecology*, **104**, 781–791.
- Losos, J.B. (2008). Phylogenetic niche conservatism, phylogenetic signal and the relationship between phylogenetic relatedness and ecological similarity among species. *Ecology Letters*, **11**, 995–1003.
- Lovejoy, T.E. (1980). Environment Projections. In: *The Global 2000 Report to the President of the U.S., Vol. 2 (The Technical Report)* (ed. Barney, G.O.). Penguin, Harmondsworth, UK, pp. 327–332.
- Lozupone, C., Hamady, M. & Knight, R. (2006). UniFrac – An online tool for comparing microbial community diversity in a phylogenetic context. *BMC Bioinformatics*, **7**, 371.
- Lozupone, C., Lladser, M.E., Knights, D., Stombaugh, J. & Knight, R. (2011). UniFrac: An effective distance metric for microbial community comparison. *The ISME Journal*, **5**, 169–172.
- Lujan, N.K. & Armbruster, J.W. (2011). The Guiana Shield. In: *Historical Biogeography of Neotropical Freshwater Fishes*. (eds. Albert, J. & Reis, R.E.). University of California Press, Berkeley, CA, USA, pp. 211–224.

- MacArthur, R. & Levins, R. (1967). The limiting similarity, convergence, and divergence of coexisting species. *The American Naturalist*, **101**, 377–385.
- MacArthur, R.H. & Wilson, E.O. (1967). *The Theory of Island Biogeography*. Princeton University Press, Princeton, NJ, USA.
- Macnaughton, C.J., Harvey-Lavoie, S., Senay, C., Lanthier, G., Bourque, G., Legendre, P. & Boisclair, D. (2015). A comparison of electrofishing and visual surveying methods for estimating fish community structure in temperate rivers. *River Research and Applications*, **31**, 1040–1051.
- Maire, E., Grenouillet, G., Brosse, S. & Villéger, S. (2015). How many dimensions are needed to accurately assess functional diversity? A pragmatic approach for assessing the quality of functional spaces. *Global Ecology and Biogeography*, **24**, 728–740.
- Malavoi, J.R. & Souchon, Y. (2002). Description standardisée des principaux faciès d'écoulement observables en rivière: Clé de détermination qualitative et mesures physiques. *Bulletin Français de la Pêche et de la Pisciculture*, pp. 357–372.
- Matheson, C.D., Gurney, C., Esau, N. & Lehto, R. (2014). Assessing PCR inhibition from humic substances. *The Open Enzyme Inhibition Journal*, **3**, 38–45.
- Mayfield, M.M. & Levine, J.M. (2010). Opposing effects of competitive exclusion on the phylogenetic structure of communities. *Ecology letters*, **13**, 1085–1093.
- McIntire, E.J.B. & Fajardo, A. (2009). Beyond description: The active and effective way to infer processes from spatial patterns. *Ecology*, **90**, 46–56.
- Melki, F. (2016). *Poissons d'eau douce de Guyane: plongée dans les eaux de l'Amazonie française*. Biotope éditions, Mèze, France.
- Melo, A.S., Rangel, T.F.L.V.B. & Diniz-Filho, J.A.F. (2009). Environmental drivers of beta-diversity patterns in New-World birds and mammals. *Ecography*, **32**, 226–236.
- Mérigoux, S. & Ponton, D. (1999). Spatio-temporal distribution of young fish in tributaries of natural and flow-regulated sections of a neotropical river in French Guiana. *Freshwater Biology*, **42**, 177–198.
- Mesquita, N., Coelho, M.M. & Filomena, M.M. (2006). Spatial variation in fish assemblages across small Mediterranean drainages: Effects of habitat and landscape context. *Environmental Biology of Fishes*, **77**, 105–120.
- Michener, C.D. & Sokal, R.R. (1957). A Quantitative Approach to a Problem in Classification. *Evolution*, **11**, 130–162.
- Miklós, I. & Podani, J. (2004). Randomization of presence–absence matrices: Comments and new algorithms. *Ecology*, **85**, 86–92.

- Mittelbach, G.G., Schemske, D.W., Cornell, H.V., Allen, A.P., Brown, J.M., Bush, M.B., Harrison, S.P., Hurlbert, A.H., Knowlton, N., Lessios, H.A., McCain, C.M., McCune, A.R., McDade, L.A., McPeck, M.A., Near, T.J., Price, T.D., Ricklefs, R.E., Roy, K., Sax, D.F., Schluter, D., Sobel, J.M. & Turelli, M. (2007). Evolution and the latitudinal diversity gradient: Speciation, extinction and biogeography. *Ecology Letters*, **10**, 315–331.
- Miya, M., Sato, Y., Fukunaga, T., Sado, T., Poulsen, J.Y., Sato, K., Minamoto, T., Yamamoto, S., Yamanaka, H., Araki, H., Kondoh, M. & Iwasaki, W. (2015). MiFish, a set of universal PCR primers for metabarcoding environmental DNA from fishes: Detection of more than 230 subtropical marine species. *Royal Society Open Science*, **2**, 150088.
- Mojica, J.I., Lobón-Cerviá, J. & Castellanos, C. (2014). Quantifying fish species richness and abundance in Amazonian streams: Assessment of a multiple gear method suitable for Terra firme stream fish assemblages. *Fisheries Management and Ecology*, **21**, 220–233.
- Mol, J.H. & Ouboter, P.E. (2004). Downstream effects of erosion from small-scale gold mining on the instream habitat and fish community of a small neotropical rainforest stream. *Conservation Biology*, **18**, 201–214.
- Mol, J.H., Wan Tong You, K., Vrede, I., Flynn, A., Ouboter, P. & Van der Lugt, F. (2007). Fishes of Lely and Nassau Mountains, Suriname. In: *RAP Bulletin of Biological Assessment: A Rapid Biological Assessment of the Lely and Nassau Plateaus, Suriname (with Additional Information on the Brownsberg Plateau)* (eds. Alonso, L.A. & Mol, J.H.). Conservation International, Arlington, VA, USA, RAP Bulletin of Biological Assessment, pp. 107–118.
- Morinière, J., Van Dam, M.H., Hawlitschek, O., Bergsten, J., Michat, M.C., Hendrich, L., Ribera, I., Toussaint, E.F. & Balke, M. (2016). Phylogenetic niche conservatism explains an inverse latitudinal diversity gradient in freshwater arthropods. *Scientific Reports*, **6**.
- Mouillot, D. (2007). Niche-assembly vs. dispersal-assembly rules in coastal fish metacommunities: Implications for management of biodiversity in brackish lagoons. *Journal of Applied Ecology*, **44**, 760–767.
- Mouillot, D., Bellwood, D.R., Baraloto, C., Chave, J., Galzin, R., Harmelin-Vivien, M., Kulbicki, M., Lavergne, S., Lavorel, S., Mouquet, N., Paine, C.E.T., Renaud, J. & Thuiller, W. (2013). Rare Species Support Vulnerable Functions in High-Diversity Ecosystems. *PLOS Biology*, **11**, e1001569.
- Mouillot, D., Dumay, O. & Tomasini, J.A. (2007). Limiting similarity, niche filtering and functional diversity in coastal lagoon fish communities. *Estuarine, Coastal and Shelf Science*, **71**, 443–456.

## BIBLIOGRAPHY

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- Mouillot, D., Villéger, S., Scherer-Lorenzen, M. & Mason, N.W.H. (2011). Functional structure of biological communities predicts ecosystem multifunctionality. *PLoS ONE*, **6**, e17476.
- Munch, K., Boomsma, W., Huelsenbeck, J.P., Willerslev, E. & Nielsen, R. (2008). Statistical Assignment of DNA Sequences Using Bayesian Phylogenetics. *Systematic Biology*, **57**, 750–757.
- Murphy, B.R. & Willis, D.W. (1996). *Fisheries Techniques, 2nd Edition*. 2nd edn. American Fisheries Society, Bethesda, MD, USA.
- Myers, J.A., Chase, J.M., Jiménez, I., Jørgensen, P.M., Araujo-Murakami, A., Paniagua-Zambrana, N. & Seidel, R. (2013). Beta-diversity in temperate and tropical forests reflects dissimilar mechanisms of community assembly. *Ecology Letters*, **16**, 151–157.
- Nakagawa, H. (2014). Contribution of environmental and spatial factors to the structure of stream fish assemblages at different spatial scales. *Ecology of Freshwater Fish*, **23**, 208–223.
- Nakagawa, S. & Schielzeth, H. (2013). A general and simple method for obtaining R<sup>2</sup> from generalized linear mixed-effects models. *Methods in Ecology and Evolution*, **4**, 133–142.
- Nekola, J.C. & White, P.S. (1999). The distance decay of similarity in biogeography and ecology. *Journal of Biogeography*, **26**, 867–878.
- Novotny, V., Drozd, P., Miller, S.E., Kulfan, M., Janda, M., Basset, Y. & Weiblen, G.D. (2006). Why are there so many species of herbivorous insects in tropical rainforests? *Science*, **313**, 1115–1118.
- Oberdorff, T., Pont, D., Hugueny, B. & Chessel, D. (2001). A probabilistic model characterizing fish assemblages of French rivers: A framework for environmental assessment. *Freshwater Biology*, **46**, 399–415.
- Oberdorff, T., Tedesco, P.A., Hugueny, B., Leprieur, F., Beauchard, O., Brosse, S. & Dürr, H.H. (2011). Global and regional patterns in riverine fish species richness: A review. *International Journal of Ecology*, **2011**, e967631.
- Oksanen, J., Blanchet, F.G., Friendly, M., Kindt, R., Legendre, P., McGlenn, D., Minchin, P.R., O'Hara, R.B., Simpson, G.L., Solymos, P., Stevens, M.H.H., Szoecs, E. & Wagner, H. (2016). *Vegan: Community Ecology Package*. R package version 2.4-1c. Available at <https://CRAN.R-project.org/package=adespatial>.
- Olds, B.P., Jerde, C.L., Renshaw, M.A., Li, Y., Evans, N.T., Turner, C.R., Deiner, K., Mahon, A.R., Brueseke, M.A., Shirey, P.D., Pfrender, M.E., Lodge, D.M. & Lamberti, G.A. (2016). Estimating species richness using environmental DNA. *Ecology and Evolution*, **6**, 4214–4226.

- Olson, D.M., Dinerstein, E., Wikramanayake, E.D., Burgess, N.D., Powell, G.V., Underwood, E.C., D'Amico, J.A., Itoua, I., Strand, H.E., Morrison, J.C. & others (2001). Terrestrial Ecoregions of the World: A New Map of Life on Earth: A new global map of terrestrial ecoregions provides an innovative tool for conserving biodiversity. *BioScience*, **51**, 933–938.
- Pavoine, S. & Bonsall, M.B. (2011). Measuring biodiversity to explain community assembly: A unified approach. *Biological Reviews*, **86**, 792–812.
- Peres-Neto, P.R., Legendre, P., Dray, S. & Borcard, D. (2006). Variation partitioning of species data matrices: Estimation and comparison of fractions. *Ecology*, **87**, 2614–2625.
- Petry, A.C. & Schulz, U.H. (2006). Longitudinal changes and indicator species of the fish fauna in the subtropical Sinos River, Brazil. *Journal of Fish Biology*, **69**, 272–290.
- Pianka, E.R. (1966). Latitudinal Gradients in Species Diversity: A Review of Concepts. *The American Naturalist*, **100**, 33–46.
- Pillar, V.D. & Duarte, L.d.S. (2010). A framework for metacommunity analysis of phylogenetic structure. *Ecology Letters*, **13**, 587–596.
- Pilliod, D.S., Goldberg, C.S., Arkle, R.S. & Waits, L.P. (2014). Factors influencing detection of eDNA from a stream-dwelling amphibian. *Molecular Ecology Resources*, **14**, 109–116.
- Planquette, P., Keith, P. & Le Bail, P.Y. (1996). *Atlas des poissons d'eau douce de Guyane (tome 1)*. vol. 22 of *Collection du patrimoine naturel*. IEBG - M.N.H.N., INRA, CSP, Min. Env., Paris, France.
- Pochon, X., Zaiko, A., Hopkins, G.A., Banks, J.C. & Wood, S.A. (2015). Early detection of eukaryotic communities from marine biofilm using high-throughput sequencing: An assessment of different sampling devices. *Biofouling*, **31**, 241–251.
- Poff, N.L. (1997). Landscape filters and species traits: Towards mechanistic understanding and prediction in stream ecology. *Journal of the North American Benthological Society*, **16**, 391–409.
- Ponton, D. & Copp, G.H. (1997). Early dry-season community structure and habitat use of young fish in tributaries of the River Sinnamary (French Guiana, South America) before and after hydrodam operation. *Environmental Biology of Fishes*, **50**, 235–256.
- Portt, C.B., Coker, G.A., Ming, D.L. & Randall, R.G. (2006). *A Review of Fish Sampling Methods Commonly Used in Canadian Freshwater Habitats*. vol. 2604. Fisheries and Oceans Canada, Burlington, ON, Canada.
- Poulet, N., Beaulaton, L. & Dembski, S. (2011). Time trends in fish populations in metropolitan France: Insights from national monitoring data. *Journal of Fish Biology*, **79**, 1436–1452.

- Preston, F.W. (1962). The canonical distribution of commonness and rarity: Part I. *Ecology*, **43**, 185–215.
- Pyron, R.A., Costa, G.C., Patten, M.A. & Burbrink, F.T. (2015). Phylogenetic niche conservatism and the evolutionary basis of ecological speciation. *Biological Reviews of the Cambridge Philosophical Society*, **90**, 1248–1262.
- Pyron, R.A. & Wiens, J.J. (2013). Large-scale phylogenetic analyses reveal the causes of high tropical amphibian diversity. *Proceedings of the Royal Society of London B: Biological Sciences*, **280**, 20131622.
- QGIS Development Team (2016). QGIS Geographic Information System. Available at <http://qgis.osgeo.org>.
- Qian, H., Swenson, N.G. & Zhang, J. (2013). Phylogenetic beta diversity of angiosperms in North America. *Global Ecology and Biogeography*, **22**, 1152–1161.
- R Core Team (2015). R: A language and environment for statistical computing.
- Rahel, F.J. & Hubert, W.A. (1991). Fish assemblages and habitat gradients in a rocky mountain–great plains stream: Biotic zonation and additive patterns of community change. *Transactions of the American Fisheries Society*, **120**, 319–332.
- Rees, H.C., Gough, K.C., Middleditch, D.J., Patmore, J.R.M. & Maddison, B.C. (2015). Applications and limitations of measuring environmental DNA as indicators of the presence of aquatic animals. *Journal of Applied Ecology*, **52**, 827–831.
- Reyjol, Y., Hugueny, B., Pont, D., Bianco, P.G., Beier, U., Caiola, N., Casals, F., Cowx, I., Economou, A., Ferreira, T., Haidvogel, G., Noble, R., De Sostoa, A., Vigneron, T. & Virbickas, T. (2007). Patterns in species richness and endemism of European freshwater fish. *Global Ecology and Biogeography*, **16**, 65–75.
- Rhodes, J.R., McAlpine, C.A., Zuur, A.F., Smith, G.M. & Ieno, E.N. (2009). GLMM applied on the spatial distribution of koalas in a fragmented landscape. In: *Mixed Effects Models and Extensions in Ecology with R* (eds. Zuur, A.F., Ieno, E.N., Walker, N., Saveliev, A.A. & Smith, G.M.). Springer, New York, USA, Statistics for Biology and Health, pp. 469–492.
- Ricklefs, R.E. & O'Rourke, K. (1975). Aspect diversity in moths: A temperate-tropical comparison. *Evolution*, **29**, 313–324.
- Robson, B.J. & Chester, E.T. (1999). Spatial patterns of invertebrate species richness in a river: The relationship between riffles and microhabitats. *Australian Journal of Ecology*, **24**, 599–607.
- Robson, H.L.A., Noble, T.H., Saunders, R.J., Robson, S.K.A., Burrows, D.W. & Jerry, D.R. (2016). Fine-tuning for the tropics: Application of eDNA technology for invasive fish detection in tropical freshwater ecosystems. *Molecular Ecology Resources*, **16**, 922–932.

- Rolland, J., Condamine, F.L., Jiguet, F. & Morlon, H. (2014). Faster speciation and reduced extinction in the tropics contribute to the mammalian latitudinal diversity gradient. *PLoS Biol*, **12**, e1001775.
- Rondelet, G. (1558). *L'Histoire entière des poissons, composée premièrement en latin par maistre Guillaume Rondelet..., maintenant traduite en françois..., avec leurs pourtraits au naïf*. Translated by Laurent Joubert, M. Bonhome, Lyon, France.
- Rossetto, M., Kooyman, R., Sherwin, W. & Jones, R. (2008). Dispersal limitations, rather than bottlenecks or habitat specificity, can restrict the distribution of rare and endemic rainforest trees. *American Journal of Botany*, **95**, 321–329.
- Roussel, J.M., Paillisson, J.M., Tréguier, A. & Petit, E. (2015). The downside of eDNA as a survey tool in water bodies. *Journal of Applied Ecology*, **52**, 823–826.
- Safi, K., Cianciaruso, M.V., Loyola, R.D., Brito, D., Armour-Marshall, K. & Diniz-Filho, J.A.F. (2011). Understanding global patterns of mammalian functional and phylogenetic diversity. *Philosophical Transactions of the Royal Society of London B: Biological Sciences*, **366**, 2536–2544.
- Sanciango, M.D., Carpenter, K.E. & Betancur-R., R. (2016). Phylogenetic placement of enigmatic percomorph families (Teleostei: Percomorphaceae). *Molecular Phylogenetics and Evolution*, **94, Part B**, 565–576.
- Sanderson, M.J. (2002). Estimating absolute rates of molecular evolution and divergence times: A penalized likelihood approach. *Molecular biology and evolution*, **19**, 101–109.
- Schemske, D.W., Mittelbach, G.G., Cornell, H.V., Sobel, J.M. & Roy, K. (2009). Is there a latitudinal gradient in the importance of biotic interactions? *Annual Review of Ecology, Evolution, and Systematics*, **40**, 245–269.
- Schleuter, D., Daufresne, M., Massol, F. & Argillier, C. (2010). A user's guide to functional diversity indices. *Ecological Monographs*, **80**, 469–484.
- Schnell, I.B., Bohmann, K. & Gilbert, M.T.P. (2015). Tag jumps illuminated – reducing sequence-to-sample misidentifications in metabarcoding studies. *Molecular Ecology Resources*, **15**, 1289–1303.
- Sclater, P.L. (1858). On the general Geographical Distribution of the Members of the Class Aves. *Journal of the Proceedings of the Linnean Society of London. Zoology*, **2**, 130–136.
- Shannon, C.E. (1948). A mathematical theory of communication. *Bell System Technical Journal*, **27**, 379–423.
- Shurin, J.B., Havel, J.E., Leibold, M.A. & Pinel-Alloul, B. (2000). Local and regional zooplankton species richness: A scale-independent test for saturation. *Ecology*, **81**, 3062–3073.

- Simpfendorfer, C.A., Kyne, P.M., Noble, T.H., Goldsbury, J., Basiita, R.K., Lindsay, R., Shields, A., Perry, C. & Jerry, D.R. (2016). Environmental DNA detects Critically Endangered largemouth sawfish in the wild. *Endangered Species Research*, **30**, 109–116.
- Smith, S.A., Beaulieu, J.M. & Donoghue, M.J. (2009). Mega-phylogeny approach for comparative biology: An alternative to supertree and supermatrix approaches. *BMC Evolutionary Biology*, **9**, 37.
- Snyder, D.E. (2003). Invited overview: Conclusions from a review of electrofishing and its harmful effects on fish. *Reviews in Fish Biology and Fisheries*, **13**, 445–453.
- Soininen, J., Lennon, J.J. & Hillebrand, H. (2007). A multivariate analysis of beta diversity across organisms and environments. *Ecology*, **88**, 2830–2838.
- Sørensen, T.J. (1948). *A Method of Establishing Groups of Equal Amplitude in Plant Sociology Based on Similarity of Species Content and Its Application to Analyses of the Vegetation on Danish Commons*. I kommission hos E. Munksgaard, Copenhagen, Denmark.
- Spasojevic, M.J., Turner, B.L. & Myers, J.A. (2016). When does intraspecific trait variation contribute to functional beta-diversity? *Journal of Ecology*, **104**, 487–496.
- Srivastava, D.S. (1999). Using local–regional richness plots to test for species saturation: Pitfalls and potentials. *Journal of Animal Ecology*, **68**, 1–16.
- Stamatakis, A. (2014). RAxML version 8: A tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics*, **30**, 1312–1313.
- Stegen, J.C., Lin, X., Konopka, A.E. & Fredrickson, J.K. (2012). Stochastic and deterministic assembly processes in subsurface microbial communities. *The ISME Journal*, **6**, 1653–1664.
- Stein, A., Gerstner, K. & Kreft, H. (2014). Environmental heterogeneity as a universal driver of species richness across taxa, biomes and spatial scales. *Ecology Letters*, **17**, 866–880.
- Steinbauer, M.J., Dolos, K., Reineking, B. & Beierkuhnlein, C. (2012). Current measures for distance decay in similarity of species composition are influenced by study extent and grain size. *Global Ecology and Biogeography*, **21**, 1203–1212.
- Stevens, G.C. (1989). The Latitudinal Gradient in Geographical Range: How so Many Species Coexist in the Tropics. *The American Naturalist*, **133**, 240–256.
- Stroud, J.T., Bush, M.R., Ladd, M.C., Nowicki, R.J., Shantz, A.A. & Sweatman, J. (2015). Is a community still a community? Reviewing definitions of key terms in community ecology. *Ecology and Evolution*, **5**, 4757–4765.

- Svenning, J.C. & Skov, F. (2007). Ice age legacies in the geographical distribution of tree species richness in Europe. *Global Ecology and Biogeography*, **16**, 234–245.
- Swenson, N.G. & Enquist, B.J. (2009). Opposing assembly mechanisms in a neotropical dry forest: Implications for phylogenetic and functional community ecology. *Ecology*, **90**, 2161–2170.
- Swenson, N.G., Enquist, B.J., Pither, J., Kerkhoff, A.J., Boyle, B., Weiser, M.D., Elser, J.J., Fagan, W.F., Forero-Montaña, J., Fyllas, N., Kraft, N.J.B., Lake, J.K., Moles, A.T., Patiño, S., Phillips, O.L., Price, C.A., Reich, P.B., Quesada, C.A., Stegen, J.C., Valencia, R., Wright, I.J., Wright, S.J., Andelman, S., Jørgensen, P.M., Lacher Jr, T.E., Monteagudo, A., Núñez-Vargas, M.P., Vasquez-Martínez, R. & Nolting, K.M. (2012a). The biogeography and filtering of woody plant functional diversity in North and South America. *Global Ecology and Biogeography*, **21**, 798–808.
- Swenson, N.G., Erickson, D.L., Mi, X., Bourg, N.A., Forero-Montaña, J., Ge, X., Howe, R., Lake, J.K., Liu, X., Ma, K., Pei, N., Thompson, J., Uriarte, M., Wolf, A., Wright, S.J., Ye, W., Zhang, J., Zimmerman, J.K. & Kress, W.J. (2012b). Phylogenetic and functional alpha and beta diversity in temperate and tropical tree communities. *Ecology*, **93**, S112–S125.
- Taberlet, P., Coissac, E., Hajibabaei, M. & Rieseberg, L.H. (2012). Environmental DNA. *Molecular Ecology*, **21**, 1789–1793.
- Taberlet, P., Fumagalli, L., Wust-Saucy, A.G. & Cosson, J.F. (1998). Comparative phylogeography and postglacial colonization routes in Europe. *Molecular Ecology*, **7**, 453–464.
- ter Braak, C.J.F. & Šmilauer, P. (2002). *CANOCO Reference Manual and CanoDraw for Windows User's Guide: Software for Canonical Community Ordination (Version 4.5)*. Microcomputer Power, Ithaca, USA.

- ter Steege, H., Pitman, N.C.A., Sabatier, D., Baraloto, C., Salomão, R.P., Guevara, J.E., Phillips, O.L., Castilho, C.V., Magnusson, W.E., Molino, J.F., Monteagudo, A., Vargas, P.N., Montero, J.C., Feldpausch, T.R., Coronado, E.N.H., Killeen, T.J., Mostacedo, B., Vasquez, R., Assis, R.L., Terborgh, J., Wittmann, F., Andrade, A., Laurance, W.F., Laurance, S.G.W., Marimon, B.S., Marimon, B.H., Vieira, I.C.G., Amaral, I.L., Brienen, R., Castellanos, H., López, D.C., Duivenvoorden, J.F., Mogollón, H.F., Matos, F.D.d.A., Dávila, N., García-Villacorta, R., Diaz, P.R.S., Costa, F., Emilio, T., Levis, C., Schiatti, J., Souza, P., Alonso, A., Dallmeier, F., Montoya, A.J.D., Piedade, M.T.F., Araujo-Murakami, A., Arroyo, L., Gribel, R., Fine, P.V.A., Peres, C.A., Toledo, M., C, G.A.A., Baker, T.R., Cerón, C., Engel, J., Henkel, T.W., Maas, P., Petronelli, P., Stropp, J., Zartman, C.E., Daly, D., Neill, D., Silveira, M., Paredes, M.R., Chave, J., Filho, D.d.A.L., Jørgensen, P.M., Fuentes, A., Schöngart, J., Valverde, F.C., Fiore, A.D., Jimenez, E.M., Mora, M.C.P., Phillips, J.F., Rivas, G., van Andel, T.R., von Hildebrand, P., Hoffman, B., Zent, E.L., Malhi, Y., Prieto, A., Rudas, A., Ruschell, A.R., Silva, N., Vos, V., Zent, S., Oliveira, A.A., Schutz, A.C., Gonzales, T., Nascimento, M.T., Ramirez-Angulo, H., Sierra, R., Tirado, M., Medina, M.N.U., van der Heijden, G., Vela, C.I.A., Torre, E.V., Vriesendorp, C., Wang, O., Young, K.R., Baider, C., Balslev, H., Ferreira, C., Mesones, I., Torres-Lezama, A., Giraldo, L.E.U., Zagt, R., Alexiades, M.N., Hernandez, L., Huamantupa-Chuquimaco, I., Milliken, W., Cuenca, W.P., Pauletto, D., Sandoval, E.V., Gamarra, L.V., Dexter, K.G., Feeley, K., Lopez-Gonzalez, G. & Silman, M.R. (2013). Hyperdominance in the Amazonian tree flora. *Science*, **342**, 1243092.
- Terra, B.d.F., Hughes, R.M. & Araújo, F.G. (2016). Fish assemblages in Atlantic Forest streams: The relative influence of local and catchment environments on taxonomic and functional species. *Ecology of Freshwater Fish*, **25**, 527–544.
- Thomsen, P.F., Kielgast, J., Iversen, L.L., Møller, P.R., Rasmussen, M. & Willerslev, E. (2012). Detection of a diverse marine fish fauna using environmental DNA from seawater samples. *PLoS ONE*, **7**, e41732.
- Thomsen, P.F. & Willerslev, E. (2015). Environmental DNA – An emerging tool in conservation for monitoring past and present biodiversity. *Biological Conservation*, **183**, 4–18.
- Thorp, J.H., Thoms, M.C. & DeLong, M.D. (2006). The riverine ecosystem synthesis: Biocomplexity in river networks across space and time. *River Research and Applications*, **22**, 123–147.
- Tito de Morais, L. & Lauzanne, L. (1994). Zonation longitudinale des peuplements ichtyques avant mise en eau de la retenue de Petit-Saut (Guyane française). *Revue d'Hydrobiologie Tropicale*, **27**, 467–483.
- Tonn, W.M. (1990). Climate change and fish communities: A conceptual framework. *Transactions of the American Fisheries Society*, **119**, 337–352.

- Toussaint, A., Beauchard, O., Oberdorff, T., Brosse, S. & Villéger, S. (2014). Historical assemblage distinctiveness and the introduction of widespread non-native species explain worldwide changes in freshwater fish taxonomic dissimilarity. *Global Ecology and Biogeography*, **23**, 574–584.
- Toussaint, A., Charpin, N., Brosse, S. & Villéger, S. (2016). Global functional diversity of freshwater fish is concentrated in the Neotropics while functional vulnerability is widespread. *Scientific Reports*, **6**, 22125.
- Tréguier, A., Paillisson, J.M., Dejean, T., Valentini, A., Schlaepfer, M.A. & Roussel, J.M. (2014). Environmental DNA surveillance for invertebrate species: Advantages and technical limitations to detect invasive crayfish *Procambarus clarkii* in freshwater ponds. *Journal of Applied Ecology*, **51**, 871–879.
- Valentini, A., Pompanon, F. & Taberlet, P. (2009). DNA barcoding for ecologists. *Trends in Ecology & Evolution*, **24**, 110–117.
- Valentini, A., Taberlet, P., Miaud, C., Civade, R., Herder, J., Thomsen, P.F., Bellemain, E., Besnard, A., Coissac, E., Boyer, F., Gaboriaud, C., Jean, P., Poulet, N., Roset, N., Copp, G.H., Geniez, P., Pont, D., Argillier, C., Baudoin, J.M., Peroux, T., Crivelli, A.J., Olivier, A., Acqueberge, M., Le Brun, M., Møller, P.R., Willerslev, E. & Dejean, T. (2016). Next-generation monitoring of aquatic biodiversity using environmental DNA metabarcoding. *Molecular Ecology*, **25**, 929–942.
- Vane-Wright, R.I., Humphries, C.J. & Williams, P.H. (1991). What to protect? Systematics and the agony of choice. *Biological Conservation*, **55**, 235–254.
- Vannote, R.L., Minshall, G.W., Cummins, K.W., Sedell, J.R. & Cushing, C.E. (1980). The River Continuum Concept. *Canadian Journal of Fisheries and Aquatic Sciences*, **37**, 130–137.
- Villéger, S., Blanchet, S., Beauchard, O., Oberdorff, T. & Brosse, S. (2011a). Homogenization patterns of the world's freshwater fish faunas. *Proceedings of the National Academy of Sciences*, **108**, 18003–18008.
- Villéger, S., Brosse, S., Mouchet, M., Mouillot, D. & Vanni, M.J. (2017). Functional ecology of fish: Current approaches and future challenges. *Aquatic Sciences*, **79**, 1–19.
- Villéger, S., Grenouillet, G. & Brosse, S. (2013). Decomposing functional  $\beta$ -diversity reveals that low functional  $\beta$ -diversity is driven by low functional turnover in European fish assemblages. *Global Ecology and Biogeography*, **22**, 671–681.
- Villéger, S., Mason, N.W.H. & Mouillot, D. (2008). New multidimensional functional diversity indices for a multifaceted framework in functional ecology. *Ecology*, **89**, 2290–2301.

- Villéger, S., Novack-Gottshall, P.M. & Mouillot, D. (2011b). The multidimensionality of the niche reveals functional diversity changes in benthic marine biotas across geological time. *Ecology Letters*, **14**, 561–568.
- Violle, C., Navas, M.L., Vile, D., Kazakou, E., Fortunel, C., Hummel, I. & Garnier, E. (2007). Let the concept of trait be functional! *Oikos*, **116**, 882–892.
- Vitorino Júnior, O.B., Fernandes, R., Agostinho, C.S. & Pelicice, F.M. (2016). Riverine networks constrain  $\beta$ -diversity patterns among fish assemblages in a large Neotropical river. *Freshwater Biology*, **61**, 1733–1745.
- Wallace, A.R. (1876). *The Geographical Distribution of Animals: With a Study of the Relations of Living and Extinct Faunas as Elucidating the Past Changes of Earth's Surface*. MacMillan and Company, London, UK.
- Ward, R.D., Zemlak, T.S., Innes, B.H., Last, P.R. & Hebert, P.D. (2005). DNA barcoding Australia's fish species. *Philosophical Transactions of the Royal Society B: Biological Sciences*, **360**, 1847–1857.
- Webb, C.O. (2000). Exploring the phylogenetic structure of ecological communities: An example for rain forest trees. *The American Naturalist*, **156**, 145–155.
- Webb, C.O., Ackerly, D.D., McPeck, M.A. & Donoghue, M.J. (2002). Phylogenies and community ecology. *Annual Review of Ecology and Systematics*, **33**, 475–505.
- Webb, C.O. & Donoghue, M.J. (2005). Phylomatic: Tree assembly for applied phylogenetics. *Molecular Ecology Notes*, **5**, 181–183.
- Webb, P.W. (1984). Body Form, Locomotion and Foraging in Aquatic Vertebrates. *American Zoologist*, **24**, 107–120.
- Weinstein, B.G., Tinoco, B., Parra, J.L., Brown, L.M., McGuire, J.A., Stiles, F.G. & Graham, C.H. (2014). Taxonomic, phylogenetic, and trait beta diversity in South American hummingbirds. *The American Naturalist*, **184**, 211–224.
- Whittaker, R.H. (1960). Vegetation of the Siskiyou Mountains, Oregon and California. *Ecological Monographs*, **30**, 279–338.
- Whittaker, R.H. (1972). Evolution and Measurement of Species Diversity. *Taxon*, **21**, 213–251.
- Wiens, J.J. & Donoghue, M.J. (2004). Historical biogeography, ecology and species richness. *Trends in Ecology & Evolution*, **19**, 639–644.
- Wiens, J.J., Pyron, R.A. & Moen, D.S. (2011). Phylogenetic origins of local-scale diversity patterns and the causes of Amazonian megadiversity. *Ecology Letters*, **14**, 643–652.

- Willis, S.C., Winemiller, K.O. & Lopez-Fernandez, H. (2004). Habitat structural complexity and morphological diversity of fish assemblages in a Neotropical floodplain river. *Oecologia*, **142**, 284–295.
- Willoughby, J.R., Wijayawardena, B.K., Sundaram, M., Swihart, R.K. & DeWoody, J.A. (2016). The importance of including imperfect detection models in eDNA experimental design. *Molecular Ecology Resources*, **16**, 837–844.
- Wilson, E.O. & Peter, F.M. (1988). Biodiversity. In: *National Forum on BioDiversity (1986: Washington, DC)*. National Academy of Sciences (US). Smithsonian Institution., Washington, DC, USA.
- Winemiller, K.O. (1991). Ecomorphological diversification in lowland freshwater fish assemblages from five biotic regions. *Ecological Monographs*, **61**, 343–365.
- Winemiller, K.O., Agostinho, A.A. & Caramaschi, É.P. (2008). Fish ecology in tropical streams. In: *Tropical Stream Ecology* (ed. Dudgeon, D.). Academic Press, London, UK, pp. 107–146.
- Winemiller, K.O., McIntyre, P.B., Castello, L., Fluet-Chouinard, E., Giarrizzo, T., Nam, S., Baird, I.G., Darwall, W., Lujan, N.K., Harrison, I., Stiassny, M.L.J., Silvano, R.a.M., Fitzgerald, D.B., Pelicice, F.M., Agostinho, A.A., Gomes, L.C., Albert, J.S., Baran, E., Petreere, M., Zarfl, C., Mulligan, M., Sullivan, J.P., Arantes, C.C., Sousa, L.M., Koning, A.A., Hoinghaus, D.J., Sabaj, M., Lundberg, J.G., Armbruster, J., Thieme, M.L., Petry, P., Zuanon, J., Vilara, G.T., Snoeks, J., Ou, C., Rainboth, W., Pavanelli, C.S., Akama, A., van Soesbergen, A. & Sáenz, L. (2016). Balancing hydropower and biodiversity in the Amazon, Congo, and Mekong. *Science*, **351**, 128–129.
- Winemiller, K.O. & Rose, K.A. (1992). Patterns of life-history diversification in north american fishes: Implications for population regulation. *Canadian Journal of Fisheries and Aquatic Sciences*, **49**, 2196–2218.
- Wisz, M.S., Pottier, J., Kissling, W.D., Pellissier, L., Lenoir, J., Damgaard, C.F., Dormann, C.F., Forchhammer, M.C., Grytnes, J.A., Guisan, A., Heikkinen, R.K., Høye, T.T., Kühn, I., Luoto, M., Maiorano, L., Nilsson, M.C., Normand, S., Öckinger, E., Schmidt, N.M., Termansen, M., Timmermann, A., Wardle, D.A., Aastrup, P. & Svenning, J.C. (2013). The role of biotic interactions in shaping distributions and realised assemblages of species: Implications for species distribution modelling. *Biological Reviews*, **88**, 15–30.
- Wright, D.H. (1983). Species-energy theory: An extension of species-area theory. *Oikos*, **41**, 496–506.
- Xiang, Q.Y.J., Zhang, W.H., Ricklefs, R.E., Qian, H., Chen, Z.D., Wen, J., Li, J.H. & Williams, C.F. (2004). Regional differences in rates of plant speciation and molecular evolution: A comparison between eastern asia and eastern north america. *Evolution*, **58**, 2175–2184.

## BIBLIOGRAPHY

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- Yang, J., Swenson, N.G., Zhang, G., Ci, X., Cao, M., Sha, L., Li, J., Ferry Slik, J.W. & Lin, L. (2015). Local-scale partitioning of functional and phylogenetic beta diversity in a tropical tree assemblage. *Scientific Reports*, **5**, 12731.
- Yunoki, T. & Velasco, L.T. (2016). Fish metacommunity dynamics in the patchy heterogeneous habitats of varzea lakes, turbid river channels and transparent clear and black water bodies in the Amazonian Lowlands of Bolivia. *Environmental Biology of Fishes*, **99**, 391–408.





# **Diversity and assembly processes of Guianese freshwater fish assemblages**

Tropical ecosystems, especially Amazonian ecosystems, host a great diversity of terrestrial and aquatic organisms. However, the causes and the processes behind this high diversity for freshwater fish assemblages are little known, but their identification will be an asset in the assessment of anthropogenic impacts that are increasing in these regions. We studied the processes that shape the diversity and the structure of freshwater fish assemblages of non-impacted streams and rivers located in French Guiana. Within-assemblage diversity increased along an upstream/downstream gradient and was higher in sites where the habitat was diversified. Species identity changed along this gradient, which created zones along the stream. Spatial relationships between assemblages and their isolation also greatly impacted species assemblages. Using information about species traits (functional diversity) and their phylogenetic relationships (phylogenetic diversity), we showed that within-assemblage diversity was not influenced by the environment or by species interactions. We also confirmed that dispersal limitation, linked with the past history of drainage basins, had a strong effect on assemblage structure in both streams and in rivers. Future investigations on the processes structuring fish assemblages will need to acquire more exhaustive biological data, and therefore to develop an efficient, and non-destructive sampling method. To this aim, we evaluated the efficiency of environmental metabarcoding applied to aquatic assemblages (the molecular identification of species present from a water sample) and compared it to traditional sampling methods. Currently, metabarcoding gives complementary information to traditional sampling. It thus needs developments and further tests to increase its efficiency and allow its use for assembly processes studies. Pursuing the formalization of a conceptual framework to investigate assembly rules together with the development of an efficient fish sampling protocol are now needed to better understand the structure of tropical fish assemblages. Those theoretical and practical developments will contribute to better evaluate anthropogenic disturbances on aquatic ecosystems.

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TITRE: Diversité et règles d'assemblage des communautés de poissons d'eau douce de Guyane

DIRECTEUR DE THÈSE : Sébastien BROSSE

LIEU ET DATE DE SOUTENANCE : Université Paul Sabatier, le 4 décembre 2017

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Les écosystèmes tropicaux, en particulier les écosystèmes amazoniens, sont connus pour abriter une importante diversité d'organismes, terrestres ou aquatiques. Cependant, les causes et les processus responsables de cette grande diversité dans les assemblages de poissons d'eau douce restent encore peu connus, et leur identification est un fort enjeu dans l'évaluation future des impacts des perturbations humaines qui sont grandissantes dans ces milieux. Nous avons étudié les processus qui façonnent la diversité et la structure des assemblages de poissons d'eau douce de Guyane dans les petits cours d'eau et les fleuves non impactés par les activités humaines. La diversité au sein des assemblages de petits cours d'eau augmente le long du gradient amont/aval et dans les milieux où l'habitat est plus diversifié. L'identité des espèces change le long de ce gradient, définissant des zones le long du cours d'eau. Les relations spatiales entre les assemblages et leur isolement ont aussi un fort effet sur les assemblages. En incorporant les informations sur les traits des espèces (diversité fonctionnelle) et leur relation de parenté (diversité phylogénétique), nous avons montré que la diversité au sein des assemblages n'était pas influencée par l'environnement ou les interactions entre les espèces. Nous avons aussi confirmé le fort effet de la limite à la dispersion entre les assemblages, en lien avec l'histoire passée des bassins versants, dans les petits cours d'eau et dans les fleuves. De telles études sur les processus structurant les assemblages nécessitent l'acquisition de données biologiques plus complètes, et donc le développement d'une nouvelle méthode d'échantillonnage qui soit exhaustive et non invasive. Pour cela, nous avons testé le metabarcoding environnemental (l'identification moléculaire des espèces présentes à partir d'un échantillon d'eau). Cette méthode donne des résultats complémentaires aux pêches traditionnelles et nécessite encore un travail de développement et des tests supplémentaires pour améliorer son efficacité et permettre son utilisation pour identifier les processus structurant les assemblages. Ces travaux, aussi bien pratiques que théoriques, sont nécessaires au développement d'un meilleur cadre conceptuel sur la structure des assemblages de poissons tropicaux, et dans la construction d'indicateurs d'impacts d'activités humaines sur les écosystèmes.

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MOTS-CLÉS : Ecologie des communautés, règles d'assemblages, écomorphologie, éco-phylogénétique, poisson, metabarcoding environnemental, Guyane

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