



Dissecting earthworm biodiversity patterns in tropical rainforests through the use of DNA barcoding

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Rationale

Soil invertebrate functions

The values of soil invertebrates

- Ecological importance: regulators of soil processes
- Economic importance: maintenance of soil fertility
- Strategic importance: highly diverse communities
- Strategic importance: invasive species
- Practical importance: used as bioindicators of soil quality or pollution



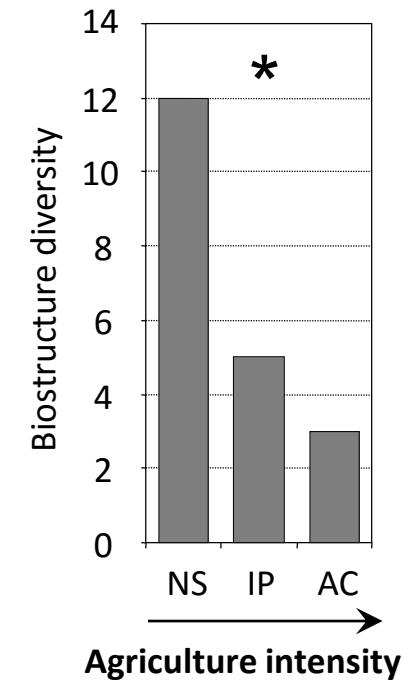
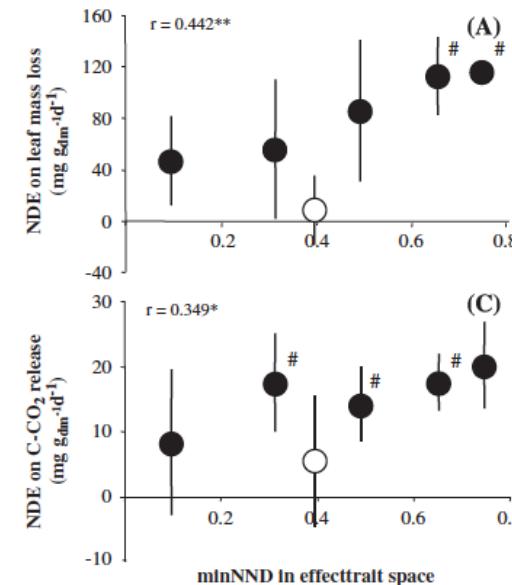
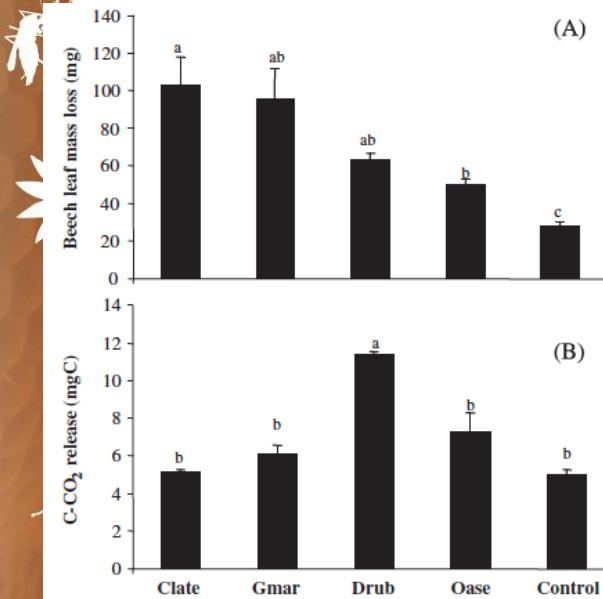


Rationale

Soil invertebrate functions

Diversity matters

- Different species perform differently in ecosystems
- Invertebrate functional diversity impacts ecological functions
- Global change can affect soil biodiversity and the production of related ecosystem services

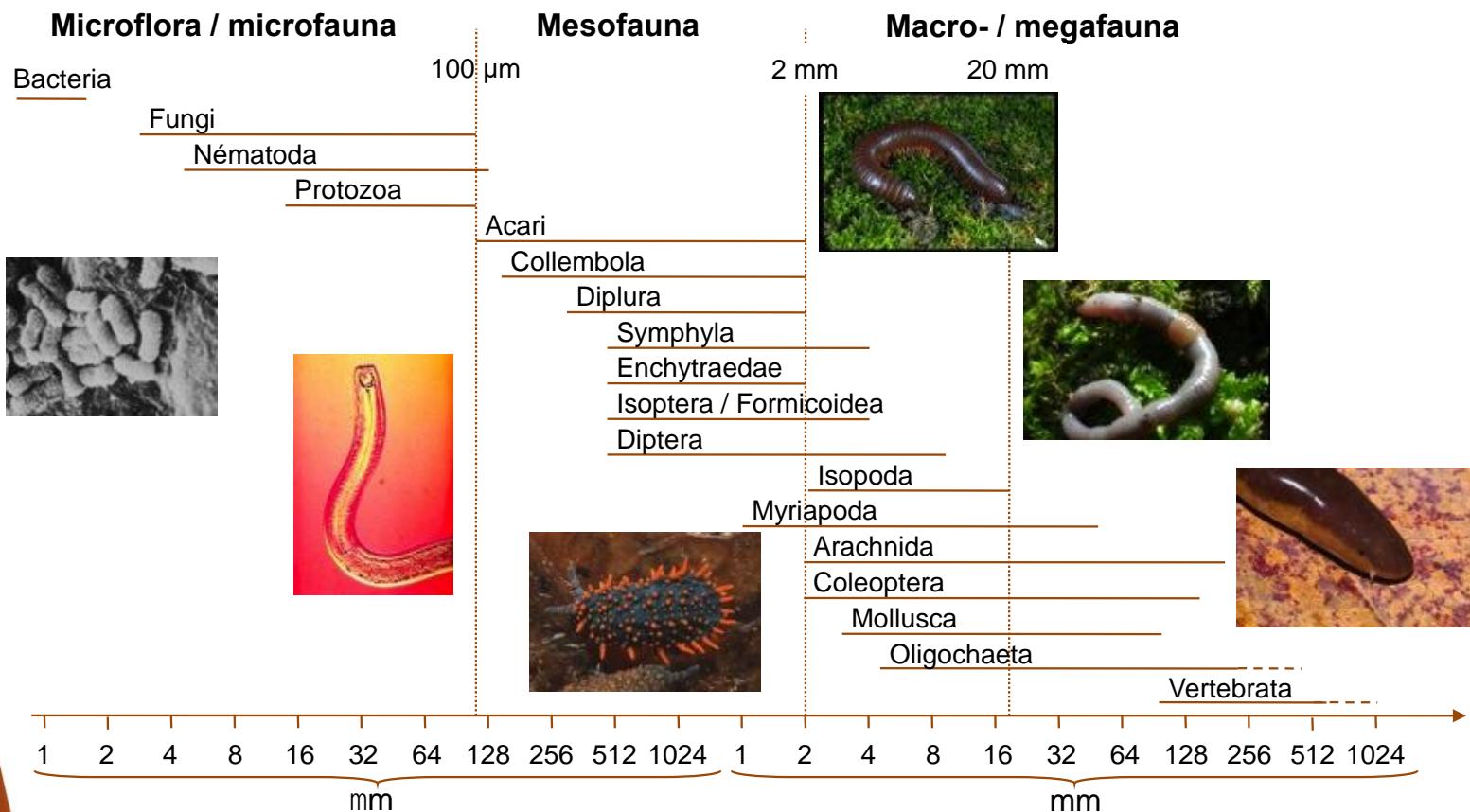




Introduction

Soil invertebrate diversity

Global soil biodiversity

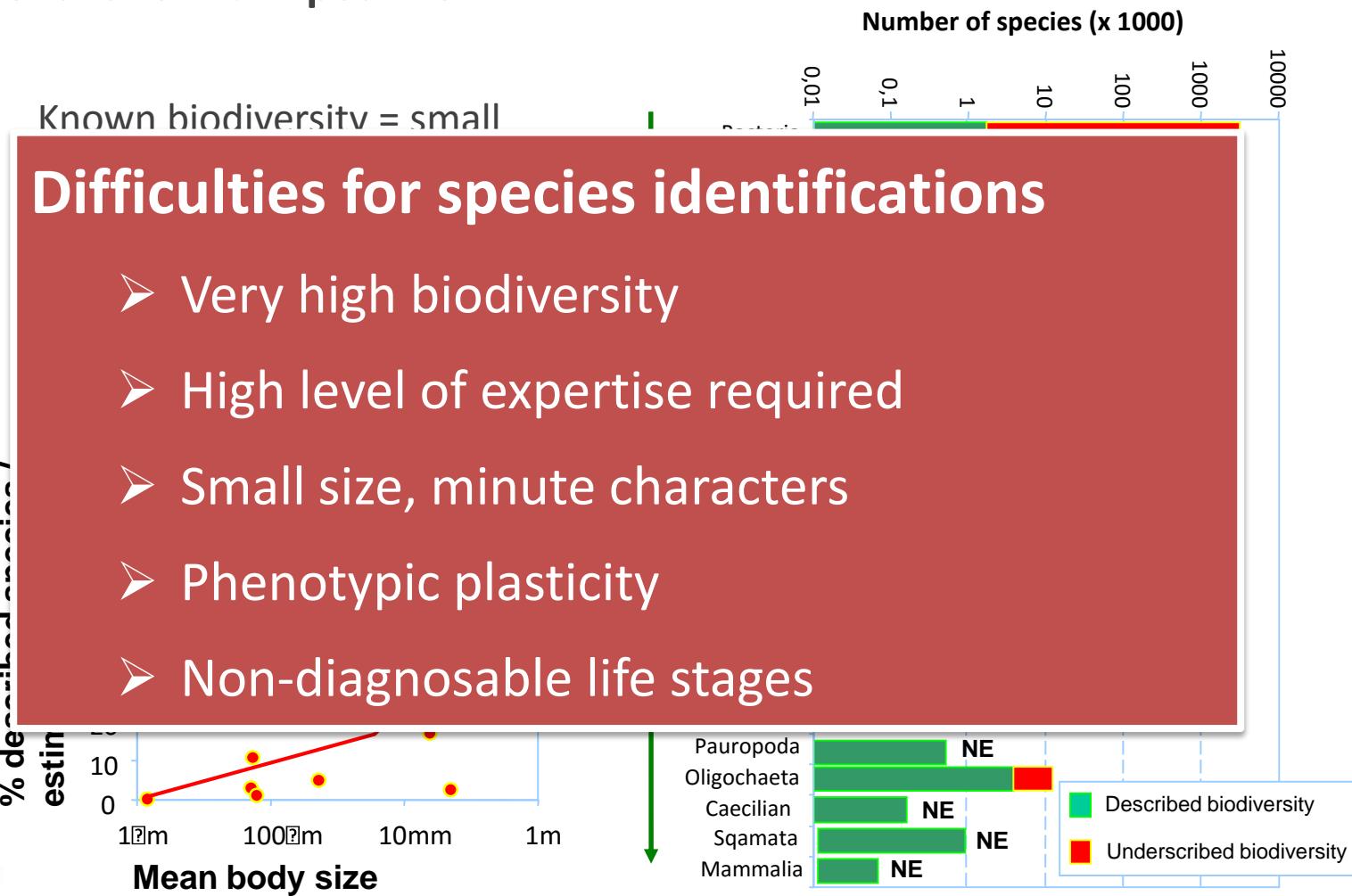




Introduction

Soil invertebrate diversity

The taxonomic impediment





Introduction

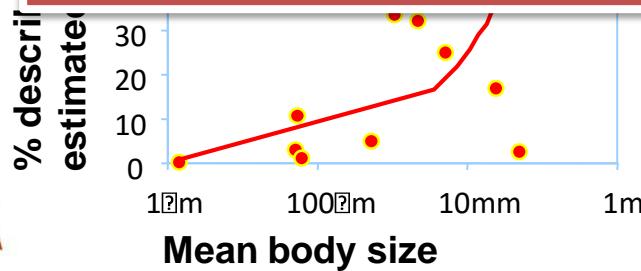
Soil invertebrate diversity

The taxonomic impediment

Known biodiversity = small fraction of real diversity

Important limitations for soil ecology:

- Deficit of taxonomists and legacy issues
- Problem for establishing reliable lists of species
- Problem for establishing the taxonomic status of important biological models





Rationale

Earthworms



Earthworm global patterns

- Are present in most soils of the world
- Represent the dominant component of soil faunal biomass in many ecosystems
- 5500 species described to date

DNA barcoding

- Rapid identifications, species discrimination
- Juveniles identification
- Cryptic diversity

→ **Molecular operational taxonomic units (MOTUs)**

→ Community scale barcoding to describe diversity patterns in regions where taxonomy is mostly unresolved

→ **Useful tool in soil ecology, macroecology, biogeography...**



Rationale

Earthworm diversity patterns
in tropical rainforests

Context

- Earthworm communities are poorly studied when compared to other groups
- We still know very few about earthworm diversity and distribution in tropical rainforests
- For example: only 22 sp listed from French Guiana (Pavlicek & Csuzdi, 2012)



The WormBank project:

- To build a library of DNA barcodes (COI) for earthworms of French Guiana
- To use the data to explore community patterns at different spatial scales

Funding : CEBA, CNRS (APEGE,
Nouragues), ANR, iBOL

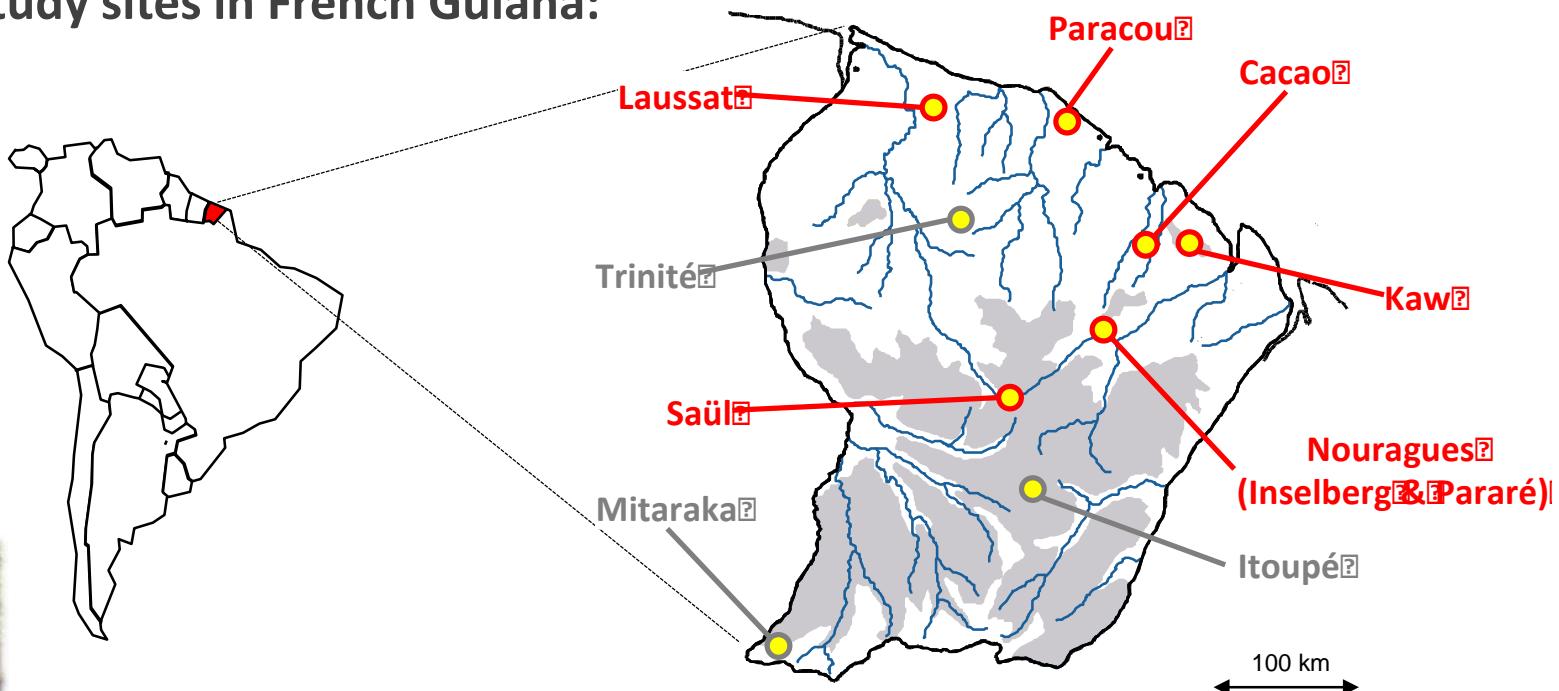




Material and methods

Sampling design

Study sites in French Guiana:





Material and methods

Sampling design



- A range of selected habitats in each site
- At least 3 replicates / habitats
- In each replicate:
 - Hand sorting of 3 soil blocks of 25x25cm
 - 2 hours (2 people) of qualitative sampling on a 1ha area
 - Soil, litter, decaying trunks, epiphytic soils...





Material and methods

Sampling design



Laussat white sands



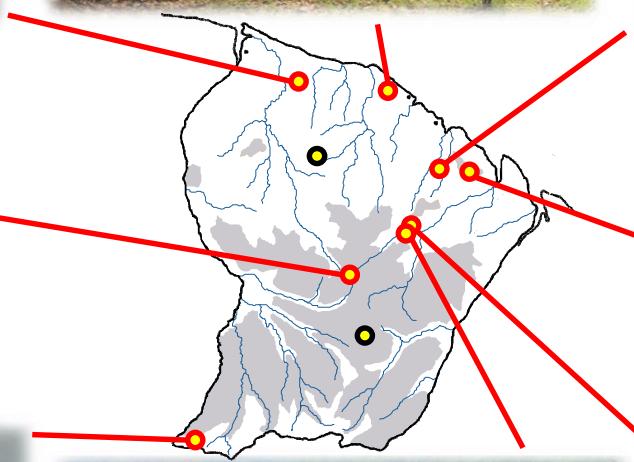
Paracou station



Saül



Mitaraka inselbergs



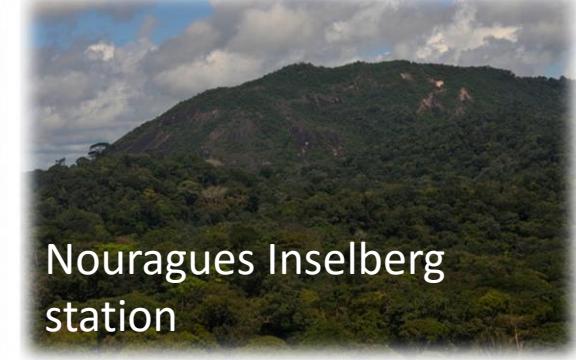
Nouragues Pararé station



Cacao



Kaw mountain



Nouragues Inselberg station

ceba
centre d'étude de la
biodiversité
amazonenne

6th
International
Barcode of Life
Conference
August 18-21, 2015



Material and methods

Sample processing

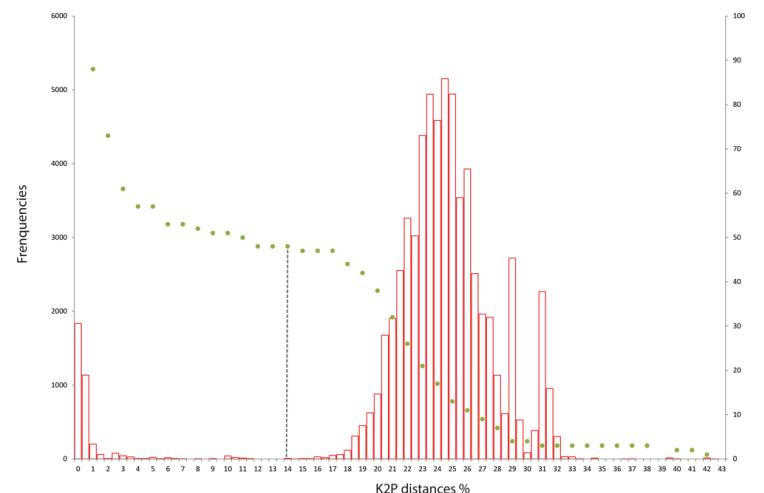


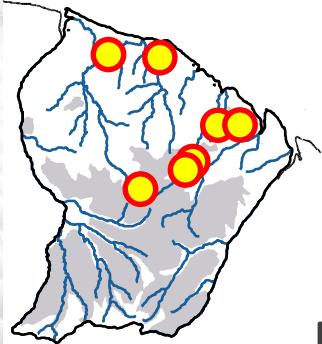
DNA barcoding

- Specimens fixed in 100% ethanol, separated into morphospecies
- Up to 5 specimens / morpho sp / sample for individual-based barcoding
- COI sequencing using Sanger (iBOL funding) or MiSeq technology (APEGE funding)

Barcode gap identification

- Identification of the barcode gap at 14% (Nouragues data)
- MOTU → species richness and community structure





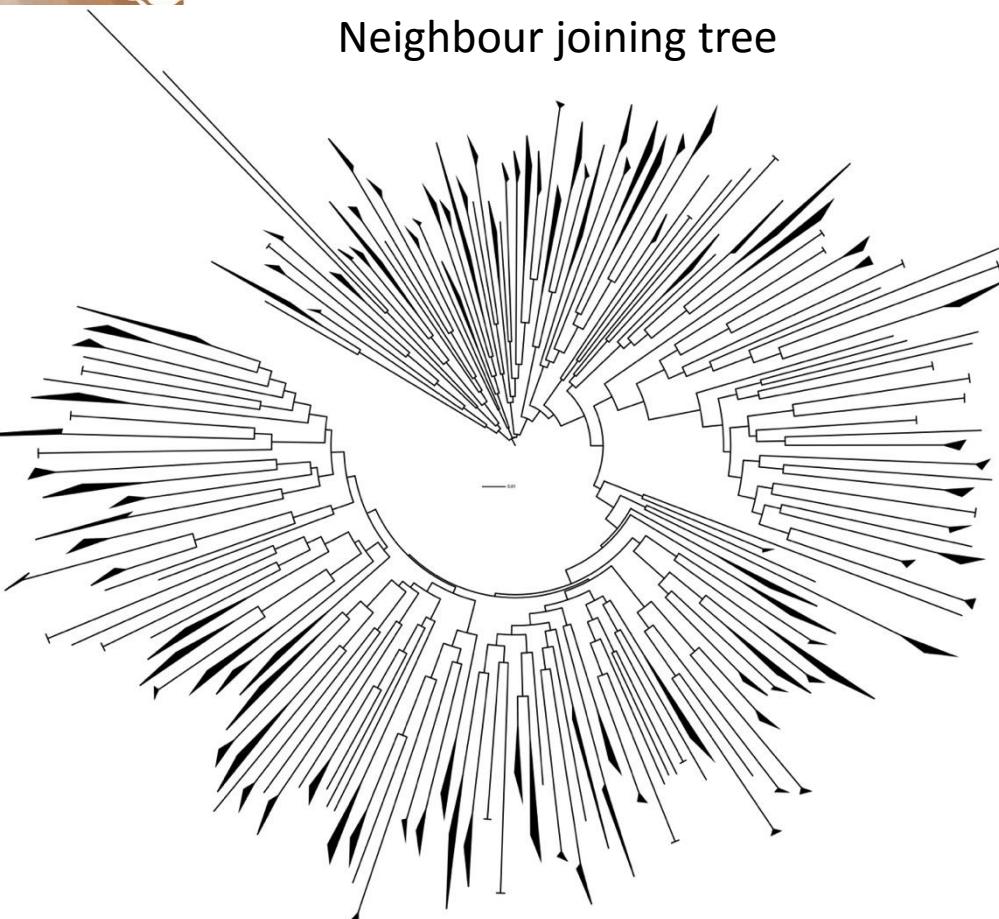
Results

Regional diversity

New results for seven study sites

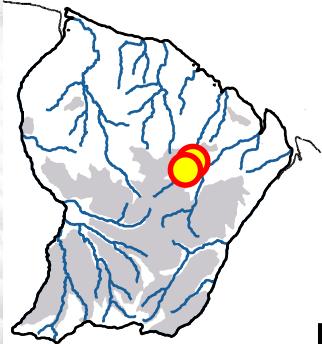
- 2561 specimens barcoded in seven different locations

Neighbour joining tree



	Year	# Specimens	# of MOTUs
Inselberg	2010	440	38
Pararé	2010	213	27
Kaw	2013	227	29
Paracou	2013	389	45
Laussat	2014	97	11
Cacao	2011-13	793	36
Saul	2013-14	402	36
Total		2561	148

- Total of 148 MOTUs
- Good match with morphology (Nouragues data)
- Most are sp new for science
- Dramatic increase compared to the 22 species checklist (Pavlíček & Csuzdi, 2012)
- Highlight a strong deficit



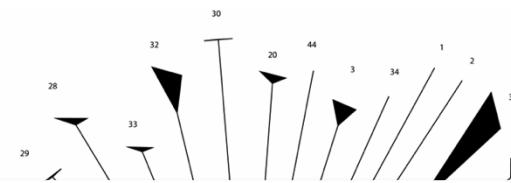
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Regional diversity

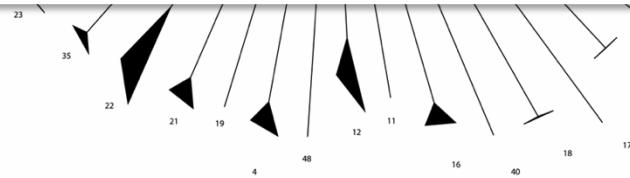


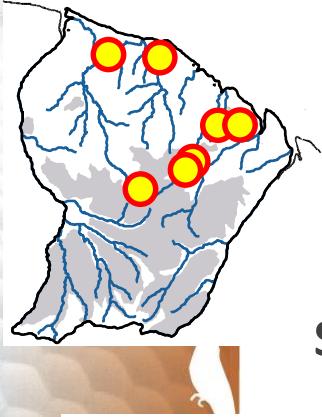
Focus on Nouragues' communities

- 650 specimens barcoded in two different locations (Pararé and Inselberg)
 - 5 families, 12 genera, 48 species level MOTUs
 - Good match with morphology, except in a few cases



- Highest local richness ever recorded worldwide
- Coexistence of most South-American families at a local scale (museum hypothesis)
- Coexistence of clusters of closely related species (cradle hypothesis)

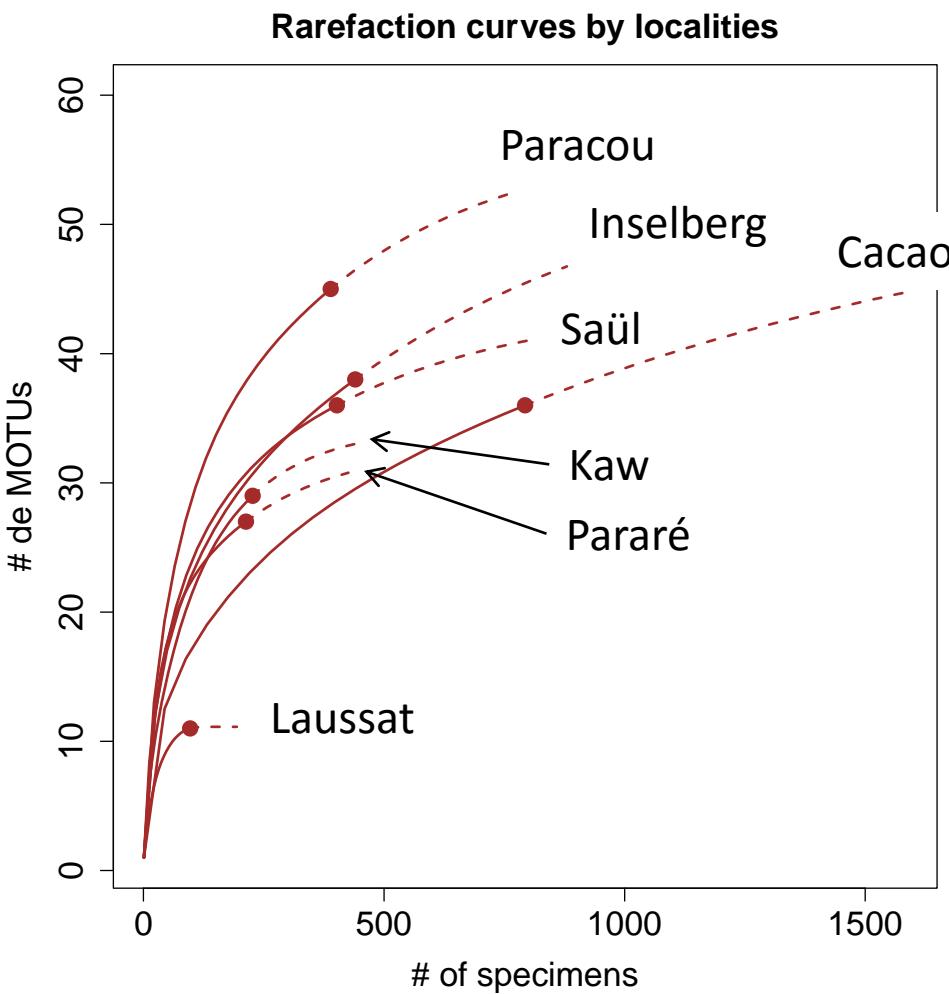




Results

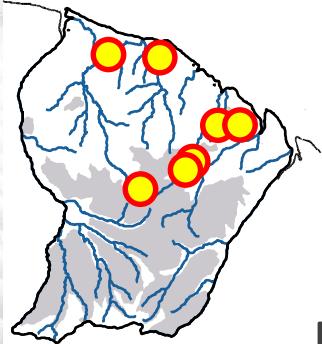
Regional diversity

Sampling completion



	Obs. Richness	% Singletons	Chao 1
Inselberg	38	26	56
Pararé	27	22	33
Kaw	29	26	34
Paracou	45	27	57
Laussat	11	9	11
Cacao	36	33	54
Saul	36	22	44
Total	148	26	-

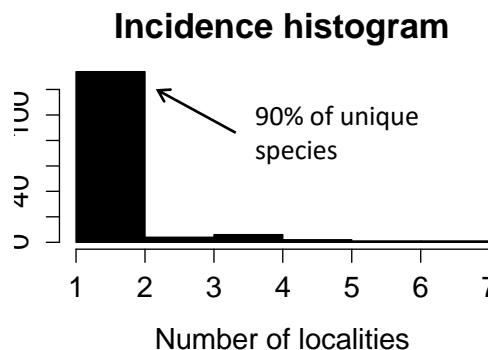
- % of singletons usually > 20%
- Local richness estimates range from 11 to 57 species
- Very high local richness compared with other tropical sites



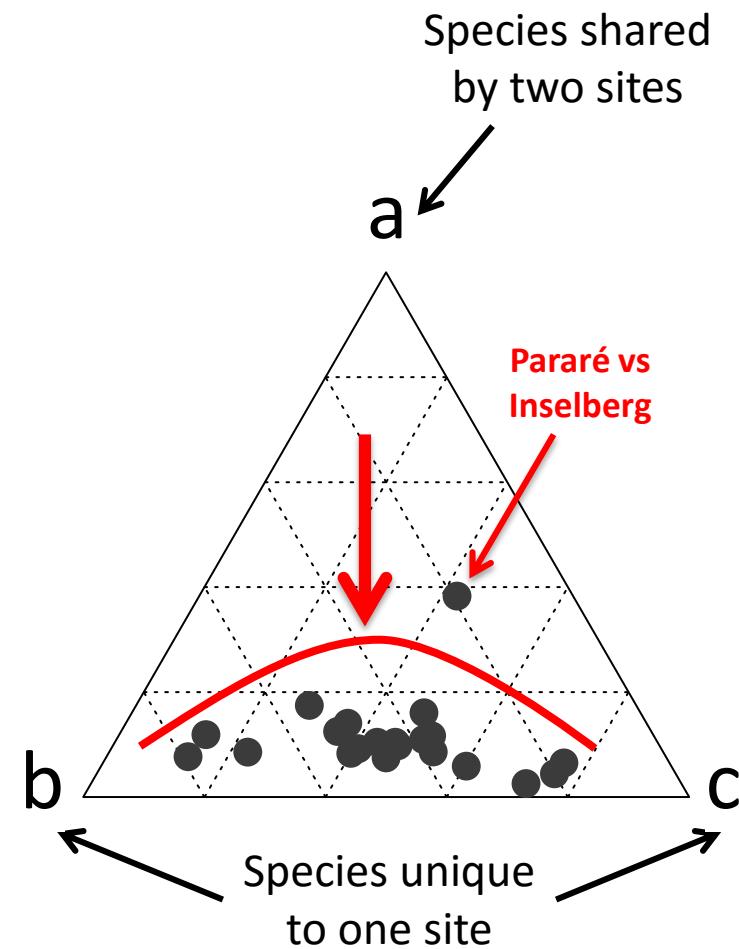
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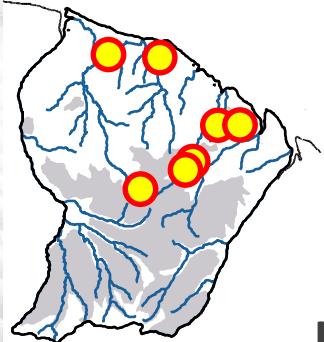
Regional diversity

Beta-diversity



- 134 MOTUs out of 148 (i.e. 90%) were found in a single locality
- Mean pairwise Sorensen's dissimilarity = 81%
- % of shared MOTUs in pairwise comparisons < 20% except in one case

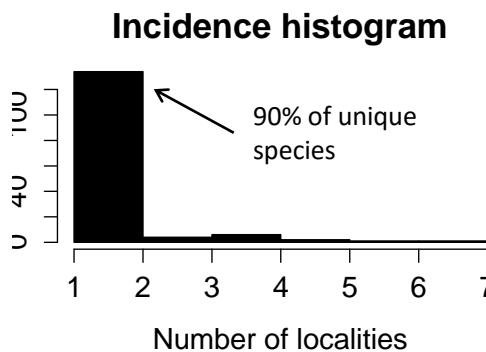




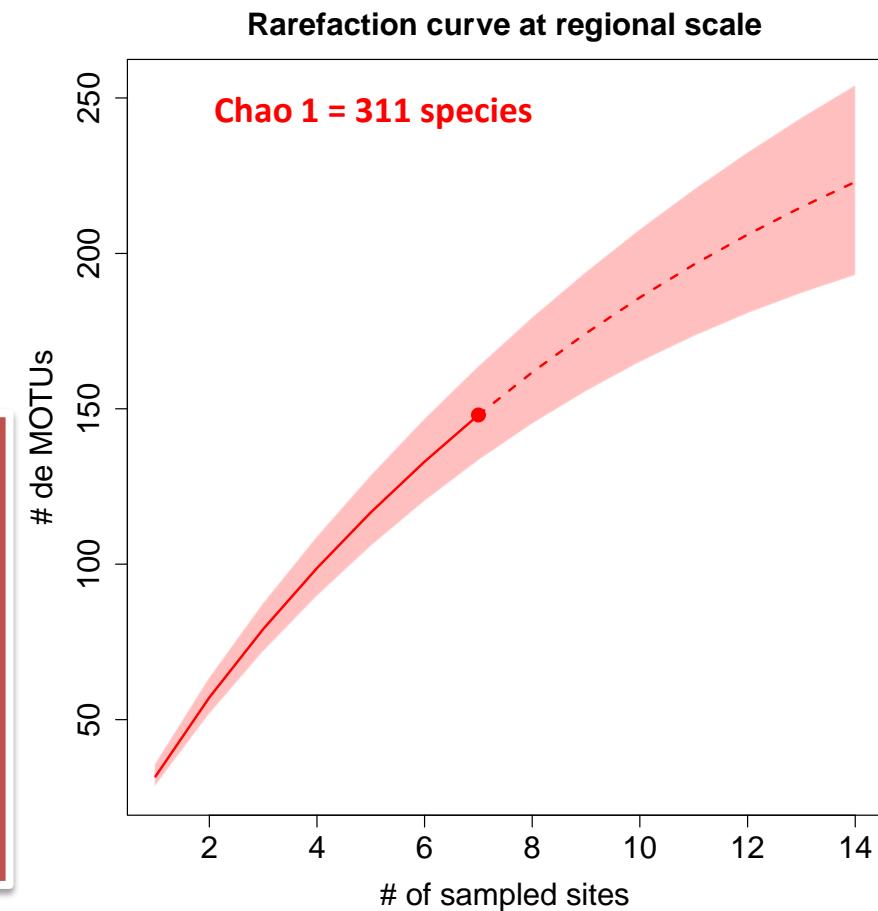
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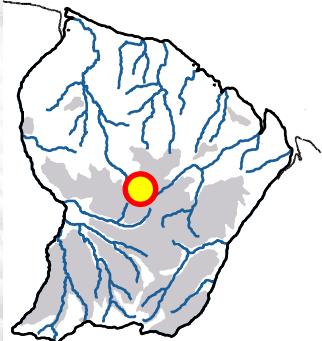
Regional diversity

How many species in French Guiana?



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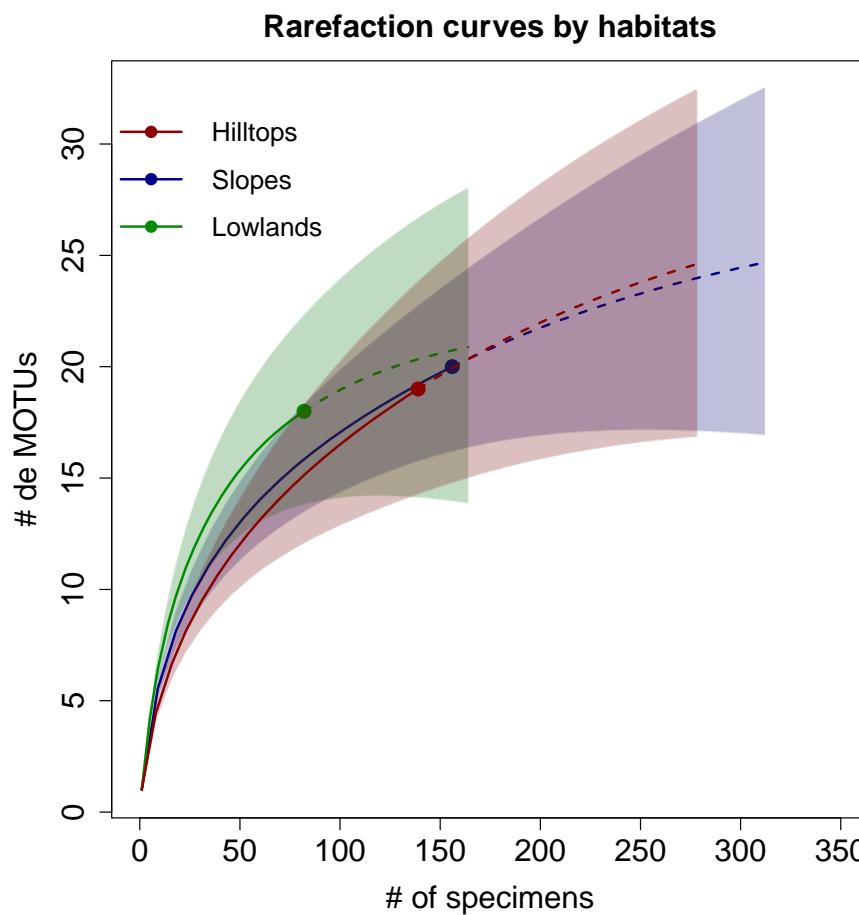




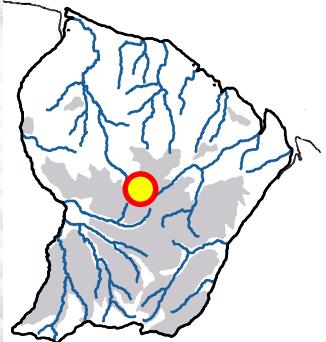
Results

Local community structure

Example: community structure in three habitats at Saül



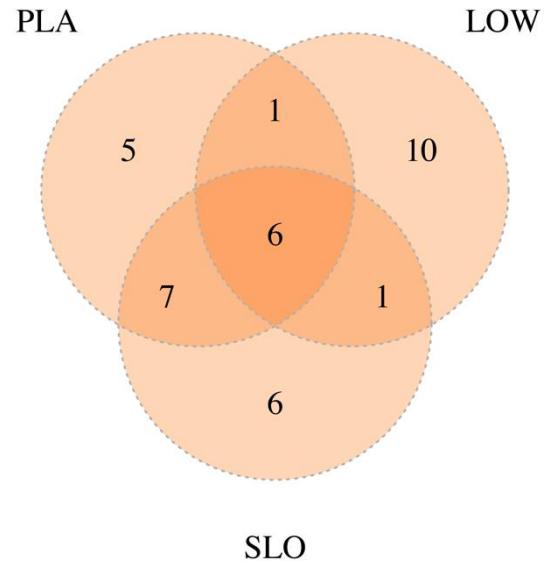
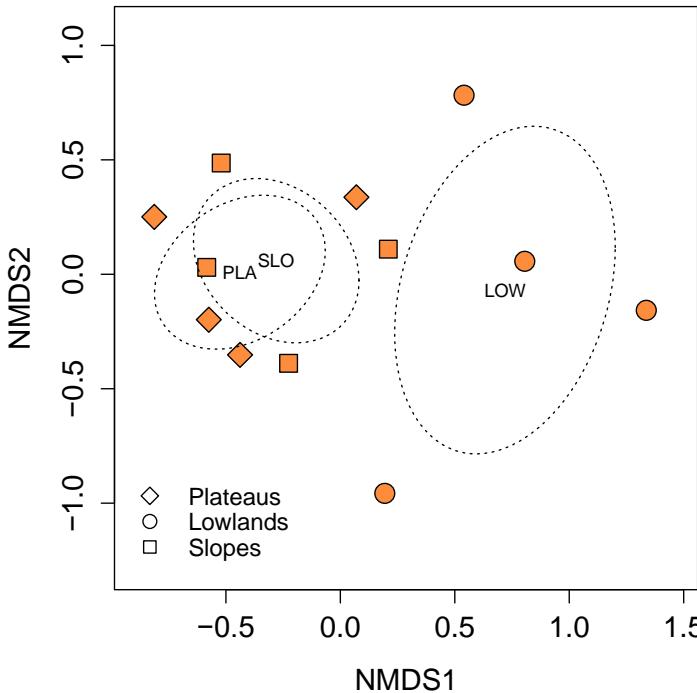
- Species richness lower in lowland forests than in the other two habitats
- Alpha diversity patterns identical in slope and plateau forests



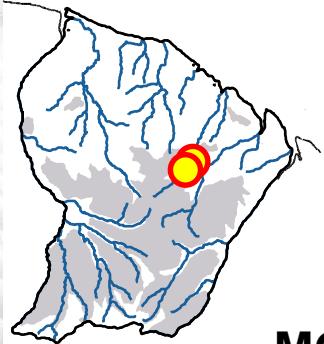
Results

Local community structure

Example: community structure in three habitats at Saül



- Strong habitat signal
- Lowland forests differentiated from the other two habitats
- 58% of MOTUs found in a single habitat

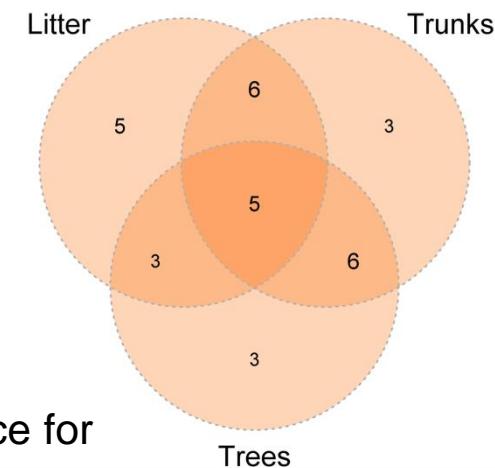
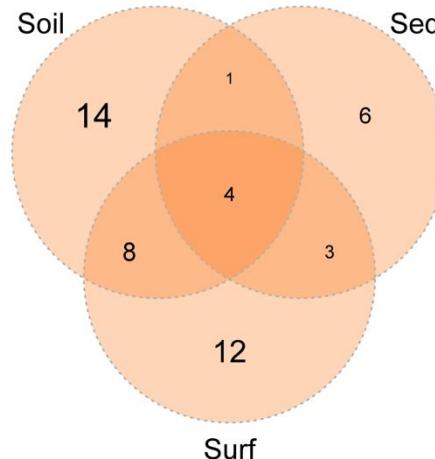


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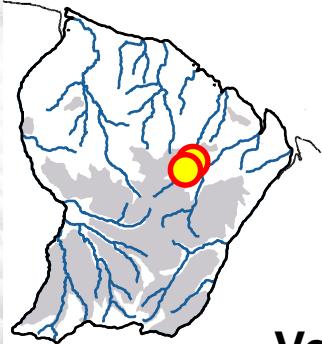
Local community structure

MOTU distribution among microhabitats

- Dominance of specialist species for a given broad type of microhabitat



- Surface species show little preference for litter, decaying trunks or epiphytic soils



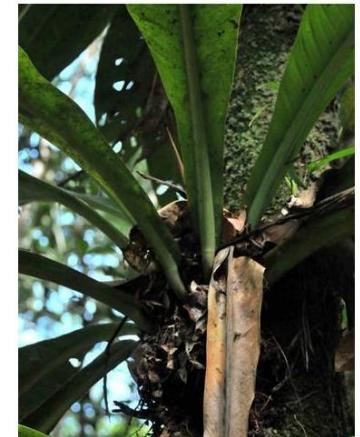
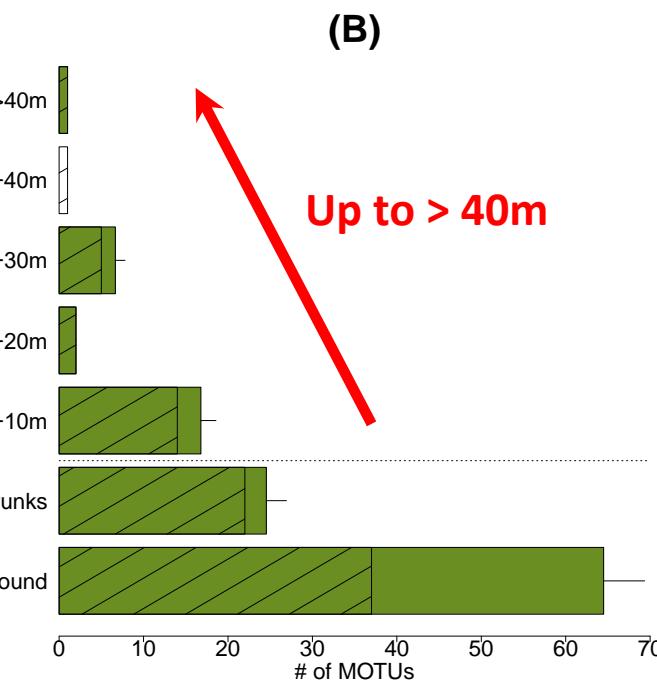
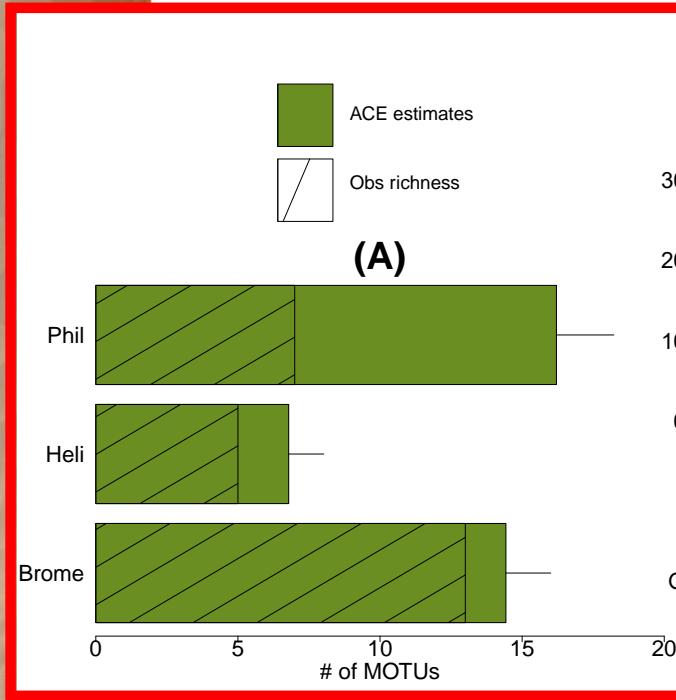
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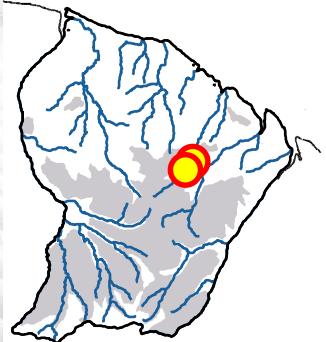
Local community structure



Vertical stratification

- 17 MOTUs in epiphytic soils
- Only 3 only found in these microhabitats
- In Bromeliads, other epiphytic plants, *Heliconia* flowers...

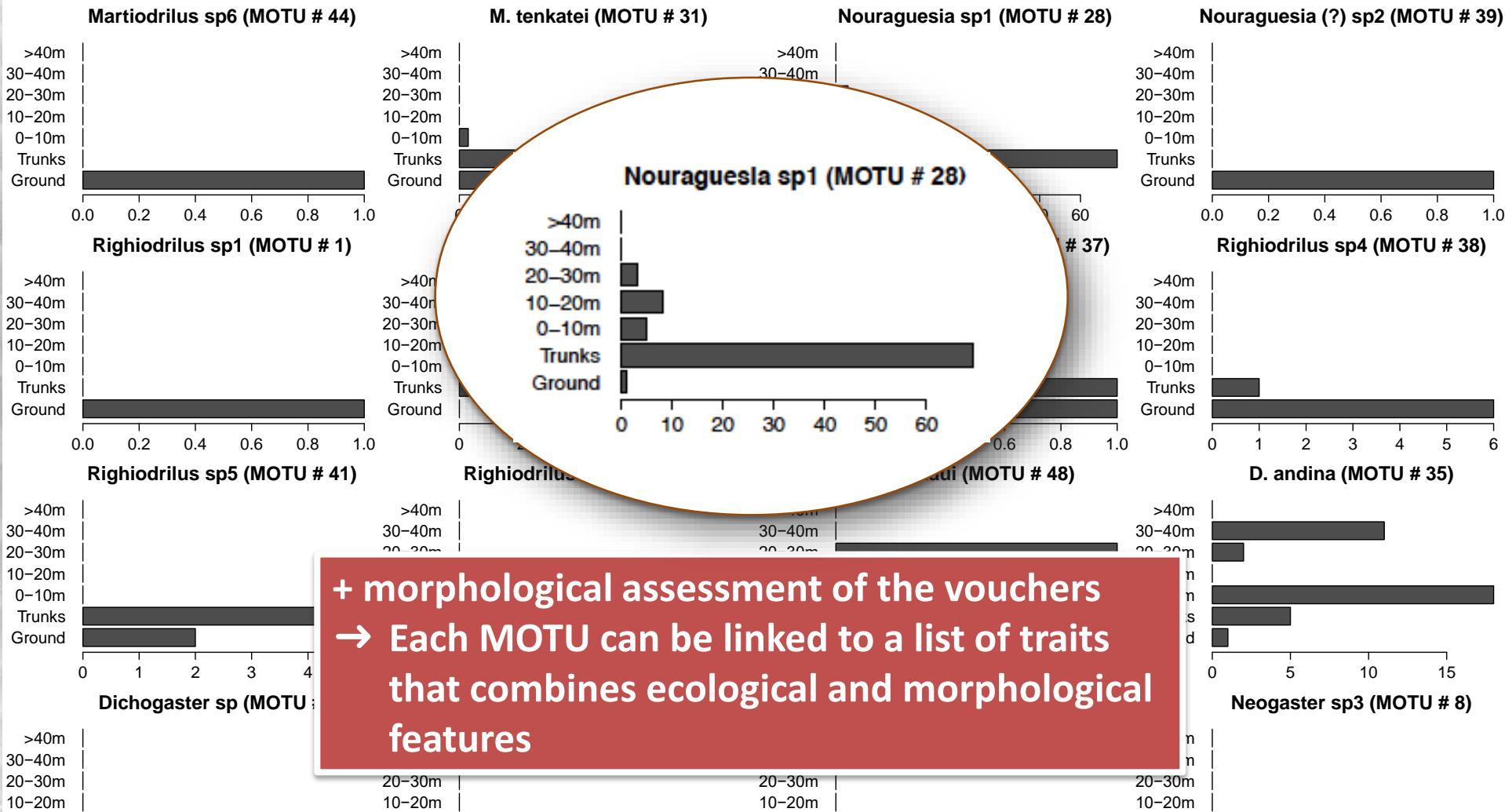




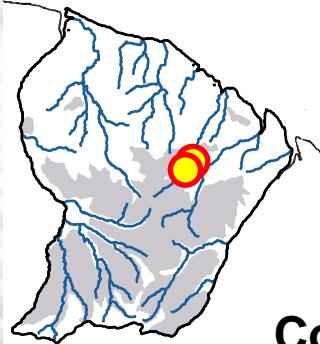
Results

Species ecology

Example: species micro-habitat affinities in Nouragues' forests



+ morphological assessment of the vouchers
 → Each MOTU can be linked to a list of traits
 that combines ecological and morphological features



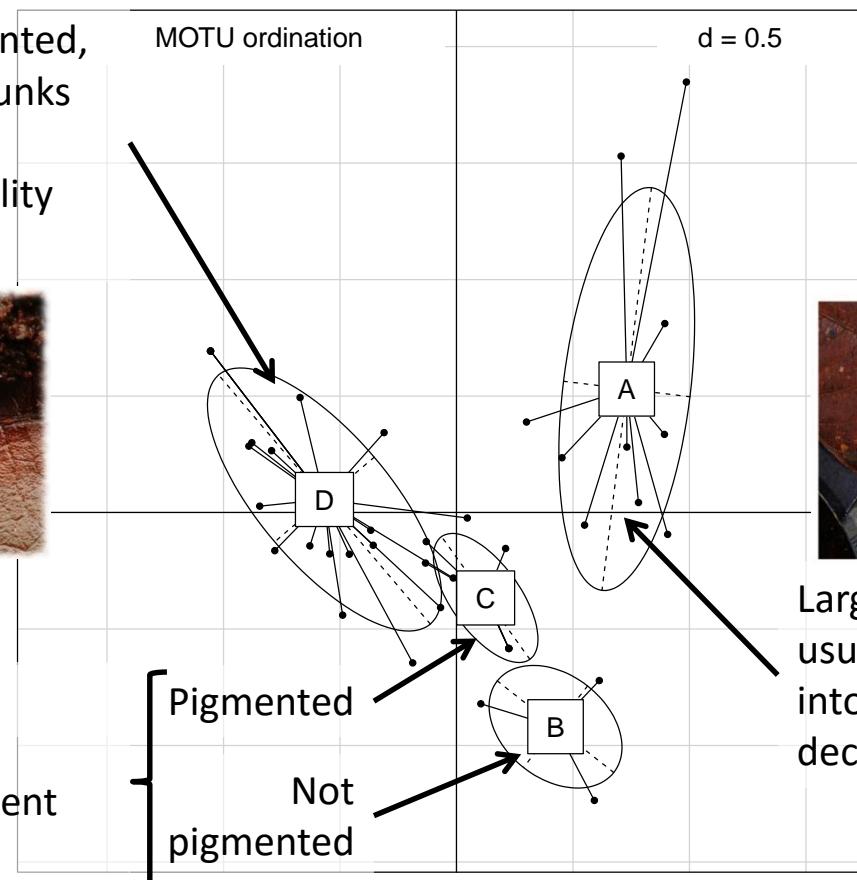
Results

Community functional ecology

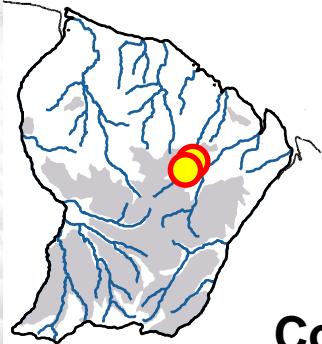


Large to medium, soil/sediment dwellers

Small usually pigmented, living in decaying trunks and epiphytic soils, feeding on high quality substrates



Large to very large, usually pigmented, living into the soil and/or decaying trunks

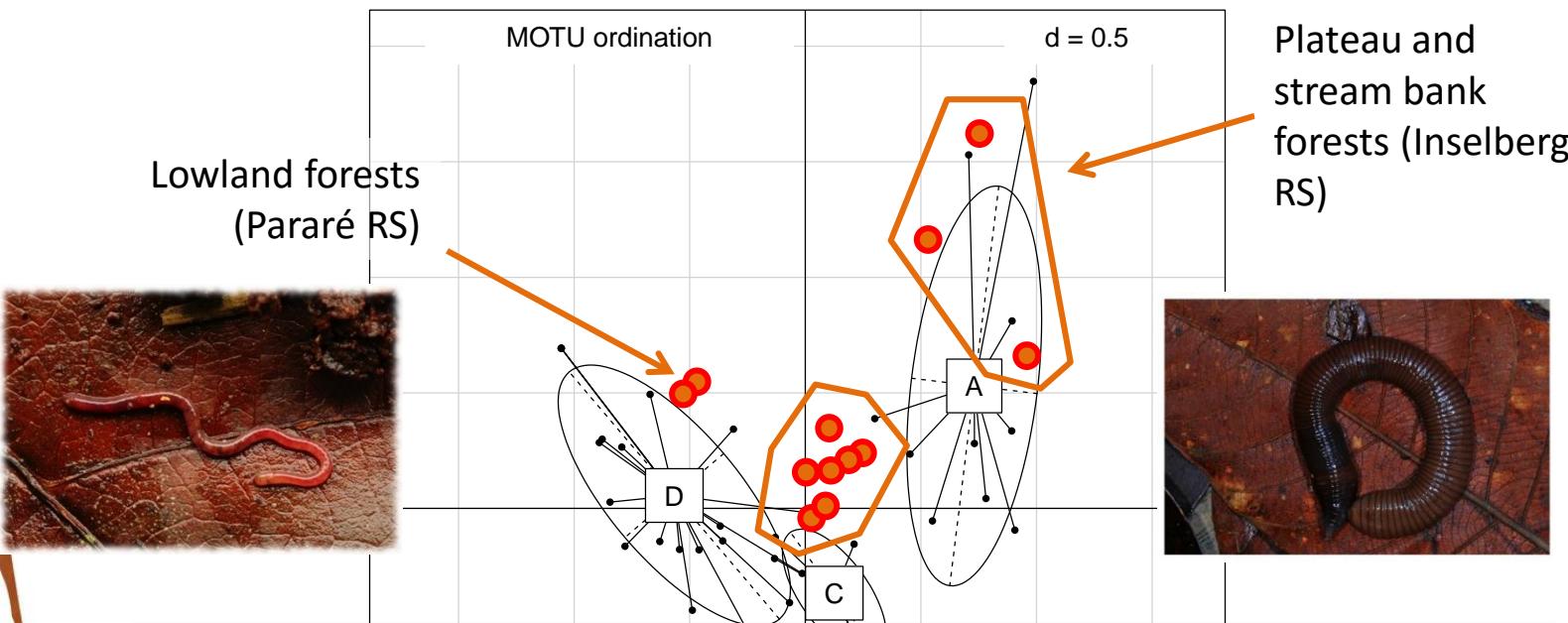


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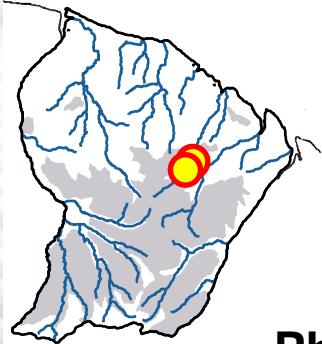
Community functional ecology

Community functional structure

- Projection of community data as supplementary columns on the reference typology



→ Guild proportionality (functional stability) is observed in most habitats
→ Except in a few ones where ecological constraints modify the proportion of specific functional groups



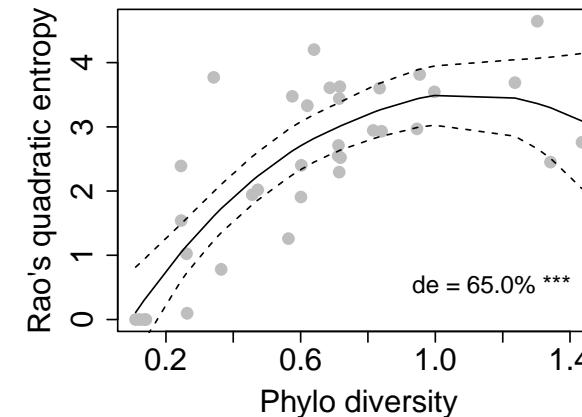
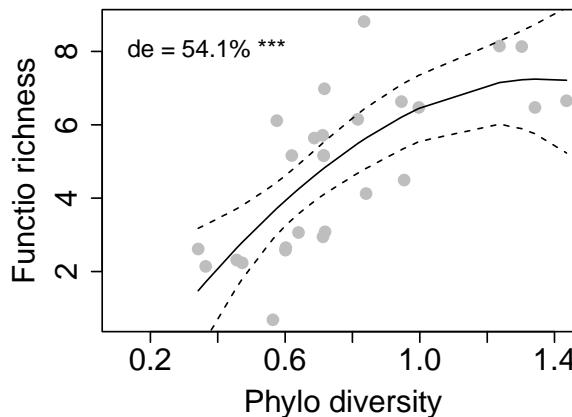
Results

Community eco-phylogenetics

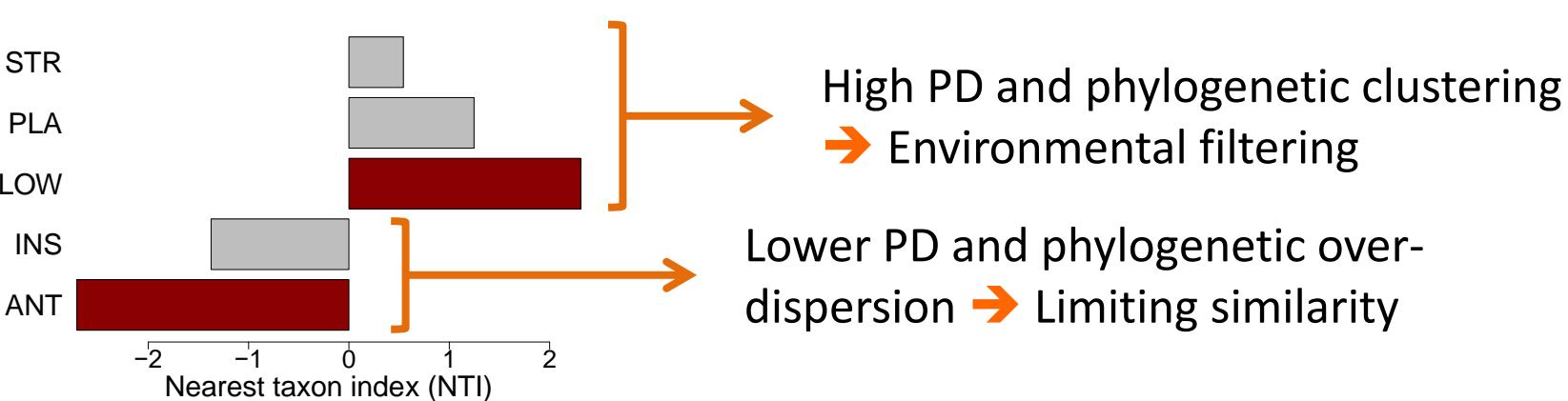


Phylogenetic diversity patterns among habitats and microhabitats

• Phylogenetic and functional diversity are significantly correlated



→ Inferring assembly rules from community phylogenetic patterns





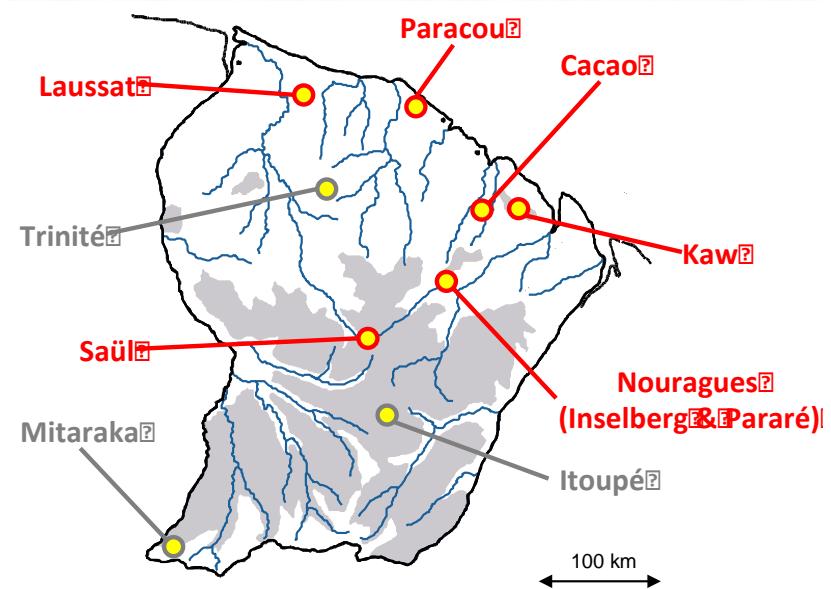
Conclusions

Highlights

- High levels of local diversity
- High levels of spatial turnover
- Consistent functional structure among communities

Perspectives

- Extend the sampling coverage
- Develop a comprehensive functional trait database
- Beta diversity patterns for TD, FD, PH
- Historical biogeography and diversification processes
- DNA barcode libraries for other studies using environmental DNA





Thanks for your attention