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Quantifying the impact of tree-diebacks and salvage logging on mountain forest biodiversity using metabarcoding



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> Background:

Climate change scenarii predict continued rising temperatures and an increase in the intensity and frequency of climate extremes. Extensive tree mortality (forest diebacks) are expected to become more widespread, frequent and severe. Tree diebacks are often followed by removal of dead trees by foresters (salvage logging).



> Main objective:

To measure the impact of climate induced silver fir forest diebacks and associated salvage logging on insect biodiversity using mass trapping and analysis of bulk samples with DNA (meta)barcoding.

> Methodology:

56 experimental plots have been set up in the French Pyrenees silver fir-dominated mountain forests (Pays de Sault and Aure valley).



Environmental measurements:

- Local dieback assessment (ARCHI), tree defoliation index, mistletoe occurrence.
- Remote sensing data.
- Dendrometric measurements, stand structure, deadwood and tree-related microhabitats.

Biomonitoring using mass trapping:

- One Malaise trap and two interception traps per plot deployed from May to September 2017.
- Samples collected once a month (224 Malaise samples and 448 polytrap™ samples in total).



DNA barcoding reference libraries for French saproxylic beetles (PASSIFOR):

- 1571 specimens, 785 COI barcodes, 485 species, 53 families.

Metabarcoding:

- Development of a metabarcoding and standardized bioinformatical pipeline.
- Test of metabarcoding analysis from preservative ethanol



> Deliverables

- Comprehensive DNA barcode reference libraries for saproxylic beetles.
- An innovative workflow and laboratory protocol for biomonitoring forest biodiversity using metabarcoding.
- A complete list of species or species-level OTUs for each collecting event for biodiversity assessments.
- Measures of the difference in taxonomic, phylogenetic and functional diversity for invertebrates communities in forest plots differently impacted by climate change in two régions of the French Pyrenees.



> Metabarcoding & Bioinformatic pipeline:

mtDNA strand

Tagged primers per individual & PCR

ATCGT

TACGA

TACGA

TACGA

ILLUMINA indices per plate

Pooling & Sequencing



Plate sorting with ILLUMINA indices

Individual sorting with tagged primers

3 PCRs to remove jump-tags

Sequence alignment

Individual COI barcode consensus recovery

> Reference:

Marie Lissandra Zepeda-Mendoza, Kristine Bohmann, Aldo Carmona Baez, M. Thomas P. Gilbert. 2016. « DAMe: a toolkit for the initial processing of datasets with PCR replicates of double-tagged amplicons for DNA metabarcoding analyses » *BMC Research Notes*. (9):255. DOI 10.1186/s13104-016-2064-9