Natural Resources Canada
Ressources naturelles Canada

Bulletin 85

FRONTLINE EXPRESS

Canadian Forest Service – Great Lakes Forestry Centre

New techniques to measure bioindicators of forest integrity

INTRODUCTION

The Canadian Forest Service is currently developing new techniques to improve our understanding of ecosystem integrity. The presence or absence of "indicator" species in soil and water can provide a way of measuring changes in biodiversity in response to various forest management practices. The assessment of ecosystem integrity is a critical activity within the forest management community. It contributes to the sustainability of Canadian forest management practices, helps maintain domestic and international market access for wood products and supports our social license to harvest wood.

The development of cost-effective, ecologically relevant, sensitive and standardized indicators is an essential component of biomonitoring. The Canadian Forest Service is developing *metabarcoding techniques to address this challenge. Metabarcoding is a DNA-based method of identifying multiple species from a sample of environmental DNA and provides a signature of the organisms that reside or pass through a particular environment. It is particularly effective because DNA can be extracted from a water or soil sample even when there are no intact organisms in the sample. Data from the metabarcoding of soil for example, can provide important information on changes in the soil biodiversity in response to alternative forest management practices.

GREAT LAKES FORESTRY CENTRE (GLFC) ROLE

Stream Biodiversity

GLFC researcher Lisa Venier and colleagues tested this approach in a recent study in Hearst Ontario by comparing species identifications from metabarcoding and from physical characteristics. Aquatic invertebrate samples were collected from streams with a gradient of watershed characteristics. Organisms were collected and identified using physical characteristics and identification keys. All organisms from each sample were then combined and DNA was extracted. The barcode region specific for invertebrates was then amplified (multiple copies were generated) and then sequenced using a new, extremely efficient technique called high throughput Illumina sequencing. Sequences were then matched with known invertebrates in the gene library to attach taxonomic identifications to the samples. The results showed consistency between the metabarcoding and traditional physical approach. In addition, the two approaches indicated similar changes in invertebrate species composition in response to the same key gradients in stream condition (i.e. dissolved oxygen, dissolved organic carbon, total nitrogen and conductivity) linked to watershed size and shifts in forest composition across watersheds. The study demonstrated the potential usefulness of invertebrate DNA metabarcoding to future application in broad-scale biomonitoring across environmental gradients.

***How metabarcoding works**

Metabarcoding uses a short genetic marker in an organism's DNA to identify its taxonomic identity. For invertebrates, a single marker (cytochrome oxidase I) that is found in all invertebrate mitochondria is used. There is a slight difference in the genetic sequence of this marker in each species, which allows for its identification using a gene library based on known species-gene matches.


Processing soil and water samples for biodiversity assessments.

Soil Arthropod Biodiversity

Soil arthropod biodiversity is considered a good indicator of soil health because these organisms are an important component of soil food webs responsible for decomposition and nutrient cycling. Some important methodological questions were tested that could help improve the approach to assessing soil arthropod biodiversity in forest soils. Species identification from specimens is very difficult and limited by available expertise. DNA metabarcoding could simplify the identification and improve repeatability.

The objective of this study was to establish the amount of field sampling required to represent the soil faunal community and understand how much replication of the DNA extraction from soil is required. Results showed that processing individually collected field samples recovered significantly more taxonomic richness than pooling the same number of field samples, but was also more costly in terms of time and money. However, both approaches led to the same ecological conclusions when comparing the similarity of communities between two different ecosystems. There did not seem to be any benefit from conducting three DNA extractions on the same sample (which is the current standard) versus a single DNA extraction. This finding has the potential to increase the efficiency of sample processing.

The gene libraries for these species groups are not very complete, so identifying taxa to species rank was not always possible. More effort is needed to build gene libraries for local areas of interest. However, the unnamed taxonomic sequences were successfully used to assess richness, distinguish among sites and recover site indicators.

Other Projects in Progress

The CFS has several projects in progress that will test these approaches under more complex experimental study designs:

1. Researchers are replicating a study to compare soil fauna across a gradient of biomass removal treatments. The original study used morphological species identification, which will allow for comparison of results with DNA metabarcoding.
2. The sustainability of wood ash amelioration in sites with biomass removal using soil faunal indicators across a national field experiment network of eight study sites is being conducted.
3. A comparison of soil fauna in five harvested, burned and salvage-logged sites representing a chronosequence of development stages across northwestern Ontario is being carried out.
4. The soil faunal recovery of biomass-harvested sites 20 years after treatment using the Long-term Soil Productivity network sites in north-central Ontario is being examined.

This collection of projects will both demonstrate the utility of DNA metabarcoding in the assessment of forest sustainability while also providing useful information in the ongoing development of sustainable forest management policy and guidelines.

Future work

In the future, scientists would like to create standardized protocols for conducting DNA metabarcoding in the context of ongoing and future experiments designed to assess forest sustainability and integrity and compare alternative forest management approaches. Improving the representivity of gene libraries for forest soil species would be a valuable asset to these studies. It is hoped that current and new DNA metabarcoding approaches will be applied to ongoing and future forest sustainability assessments.

CONCLUSION

The assessment of forest sustainability is a critical activity within the forest management community. Ensuring that forest management is sustainable is a legislative requirement that derives from the desire of the public to have forests that continue to provide ecosystem services now and into the future. Sustainable forest management ensures that the forest continues to provide habitat for wildlife, recreational opportunities, clean water, trees for timber and paper and any number of other services. This gives the forest industry the social license to harvest, and ensures market access where consumers demand sustainability. DNA metabarcoding is an important addition to the sustainability toolbox, because it provides a repeatable method for assessing a critical component of forest biodiversity that has been difficult to assess in the past. A broader assessment capability will improve the likelihood of capturing the essence of sustainability and the more success we will have in conserving ecosystem processes.

COLLABORATORS

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SUGGESTED READING

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POLICY PERSPECTIVE

The Canadian Forest Service (CFS) has a mandate to conduct research relating to the protection of the forest resources of Canada under the Forestry Act. The CFS provides a visionary approach to sustainable forest management planning and a science and research-based understanding of the forests.

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*Aussi disponible en français sous les titre : **Nouvelle Express, Bulletin N° 85**.*

ISSN 1496-7847 (Print) ISSN 1496-7855 (Online)