A whole-forest ^{14}C pulse-label study of microbial dynamics and root turnover (EBIS*)

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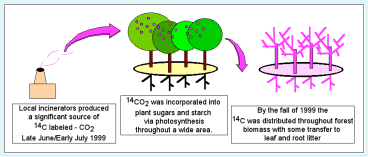
INTRODUCTION

In the summer of 1999, an incinerator released a large pulse of $^{14}CO_2$ near Oak Ridge (OR), Tennessee. The photosynthetic uptake of the $^{14}CO_2$ created a pulse label of ^{14}C in plant biomass of the local forests. This whole-ecosystem isotopic label presents unique opportunities for studying belowground carbon cycling in a temperate forest (Figure 1).
See also poster by Swanston et al.

We address three questions about C pathways from leaf vs.root input to soil organic matter:

1. **How long do fine tree roots live?**
2. **Do ectomycorrhizal fungi decompose dead organic matter or live off root carbon?**
3. **Can we measure ^{14}C in microbial biomass, and what can it tell us about the sources and decay rates of soil organic matter?**

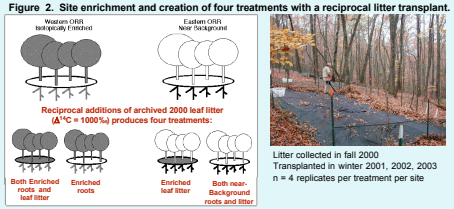
Figure 1. EBIS: Unique ecological opportunity from a ^{14}C release in 1999



EXPERIMENTAL DESIGN

Trees near the incinerator (enriched sites, Western OR in Figure 2) were more labeled with ^{14}C than trees farther away (background sites, Eastern OR in Figure 2). By moving leaves from enriched sites to background sites, and vice versa, the Oak Ridge team created four treatments with different combinations of ^{14}C inputs to soil, as shown in Figure 2.
The sites in this poster have soils formed on highly weathered dolomite.

Figure 2. Site enrichment and creation of four treatments with a reciprocal litter transplant.



Litter collected in fall 2000
Transplanted in winter 2001, 2002, 2003
n = 4 replicates per treatment per site

METHODS

Fine Roots have been harvested each year and separated into live and dead material. New root growth was isolated by harvesting roots that grew through screens in the soil.

Root lifetime was estimated by tracing the radiocarbon label into new roots and through live and dead root populations for the years since the pulse.

Ectomyccorhizal Fungi
We hand-picked ectomycorrhizal tips off roots sampled in September 2001 (6 months after litter transplant).

Microbial Biomass. To isolate microbial biomass carbon from soil, we fumigated soil with chloroform to lyse the cells and extracted with 0.5 M K_2SO_4 (chloroform fumigation-extraction method). Unfumigated (control) soils were also extracted. The control extracts represent the extractable dissolved organic carbon, and the difference between fumigated and control extracts is assumed to be microbial. The extracts were freeze-dried prior to combustion for graphitization. Soils collected in 2002 and 2003.

Radiocarbon content for all samples was determined at the Center for Accelerator Mass Spectrometry, Lawrence Livermore National Laboratory, and reported as Δ(‰) in accordance with Stuiver and Polach (1977). Graphite was prepared by hydrogen reduction.

RESULTS: FINE ROOT LIFETIMES

Radiocarbon provides one of the only ways to directly measure the lifetime of tree roots. Applying radiocarbon, however, requires a model of root growth and population turnover. In particular, we need to know how long photosynthate resides in the tree before allocation to root growth. The EBIS pulse is providing us with this information.

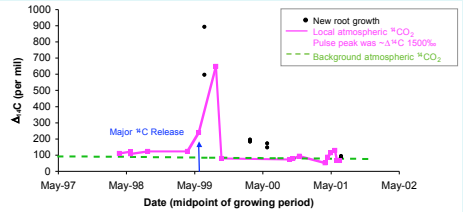


Figure 3. ^{14}C of Atmosphere, Trees, and Fine Roots at Oak Ridge. The change in $\Delta^{14}C$ of new root growth (black circles) over time at Walker Branch, Oak Ridge, TN. The atmospheric $^{14}CO_2$ at Oak Ridge (pink line) shows the major ^{14}C release in 1999. It is based on wood cellulose before July 1999 and direct atmospheric measurements after that.

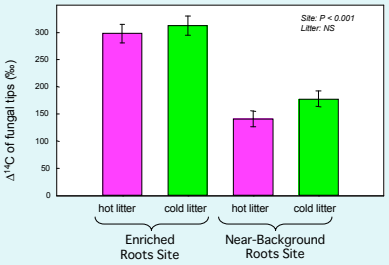
RESULTS
New root growth contains about 10–20% of the ^{14}C signature from the previous year's growth (Figure 3), showing that roots grow from a mixture of recent photosynthate and stored carbon. This result is helping us parameterize models of root growth, and to estimate fine-root turnover based on atmospheric trends in $^{14}CO_2$.

Our data from this and other temperate forests in North America and Europe suggest that fine roots (i.e., roots < 2mm diameter) of trees live 2-8 years--about 5 times longer than previously thought (data not shown).

RESULTS: ECTOMYCORRHIZAL FUNGI

Ectomycorrhizal (ECM) fungi grow on roots and they can get carbon from the host root or by decomposing organic matter. It is not known what proportion of their carbon they typically get from these different sources.

They are often the most abundant decomposer group in forest soils.



RESULTS

ECM fungi were enriched in ^{14}C only in sites with enriched roots. There is no evidence for ^{14}C tracer entering from litter or soil organic matter inputs. Thus, over the year, the ECM did not switch between mycorrhizal and saprotrophic functions (i.e. did not appear to decompose dead organic matter but instead grew on live root C).

The ECM fungi may use their decomposing enzymes to mineralize the nitrogen and phosphorus held in organic matter, without taking up any of the organic carbon. This mineralization by fungi can increase nutrient availability to plant roots.

RESULTS: MICROBIAL BIOMASS ^{14}C

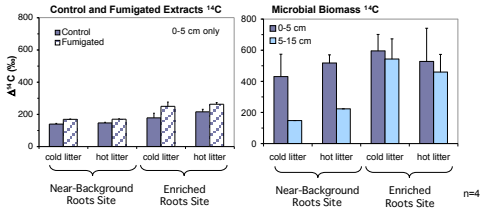
Methods Development Results
The challenges of measuring microbial or dissolved soil ^{14}C are to achieve complete combustion and maintain tube integrity while (I) minimizing the pretreatment needed for the K_2SO_4 extract, and (2) combusting enough material to obtain ~ 1mg C.

We were able to combust the salt extracts directly after freeze-drying by double-tubing to prevent devitrification, and by adding extra silver powder to form AgS precipitate to prevent oxygen consumption by SO_2 formation.

By comparing the CO_2 yield of replicate samples combusted with different sample or reagent weights, we determined that complete combustion was not sensitive to, and could be achieved with a range of, sample and reagent weights. We also confirmed complete combustion by comparing the C yield from a total carbon analyzer (Shimadzu TIC/ TOC) with C yield from combustion.

For results presented here, we combusted the equivalent of 10 ml of extract (approx. 750 mg of freeze dried salt-extract) at 900 °C with approximately 200 mg CuO and 10 mg Ag.

To calculate microbial ^{14}C, where F=fumigated, c=controls: Microbial C $\propto$ $C_F - C_c$ *and* $C_F\Delta_F = C_m\Delta_m + C_c\Delta_c$



- Microbial biomass at 0-5 cm was enriched in all treatments, but was most enriched in sites of enriched root litter as compared to leaf litter. Deeper in the soil, only plots with enriched roots had significantly enriched microbial biomass.
- The rapid enrichment (~500 ‰ in 2002) is consistent with conceptual models of microbes as an active carbon pool that decomposes root, litter, and dissolved organic material.
- The similarity to heterotrophic respiration values (~400–600 ‰; data not shown), suggests that we may be able to use microbial biomass ^{14}C to estimate the season-integrated signature soil-respired $^{14}CO_2$.

CONCLUSION

- **The ^{14}C signature of roots reflects the fact that new roots are ~80% current year's photosynthate and ~20% C stored from the previous two years.**
- **Ectomycorrhizal fungi acquired all C from live roots, rather than by decomposing organic matter.**
- **Chloroform-fumigation provides an accurate, moderately-simple measurement of ^{14}C microbial biomass.**
- **Microbial biomass was rapidly enriched and matched the signature of heterotrophic soil respiration. Microbial biomass ^{14}C in different treatments over time may be a useful tracer of the cycling of root vs. litter inputs to respiration and soil organic matter.**

ACKNOWLEDGMENTS

This research was supported by the Office of Science, Biological and Environmental Research, U.S. Department of Energy under Contract No. DE-AC03-76SF00098. The Enriched Background Isotope Study (EBIS) is led by Paul Hanson, Oak Ridge National Lab.