

An integrated approach for advancing the understanding of carbon and nutrient cycling in response to local management and global climate change

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Abstract Long-term impacts of global climate change (GCC) and local forest management on soil and plant processes in carbon (C) and nutrient cycling are complex and difficult to assess, particularly under gradually and continuously rising atmospheric carbon dioxide concentration [CO_2] and warming with multiple limiting factors. In this review, we examine the recent developments and applications of advanced stable isotope, nuclear magnetic resonance (NMR) and bio-molecular techniques, in an integrated approach with innovative rhizosphere and tree ring methods, to improve our understanding and management of above- and below-ground C and nutrient cycling processes in forest ecosystems, particularly in response to GCC and local management practices as well as mitigation / adaptation strategies. The opportunities and limitations of these techniques in investigating C and nutrient cycling processes in forest ecosystems are discussed in the context of both short- and long-term impacts on above- and below-ground processes.

Global climate change and forest management

Over the last century, atmospheric [CO_2] has increased globally by nearly 30% and temperature by approximately 0.6 °C, and these trends are projected to continue and accelerate (e.g. Xu & Chen 2006), particularly with more extreme climatic conditions. The impacts of GCC on the future structure, composition, and cycling of C and nutrients in forest ecosystems deserve particular attention and further research.

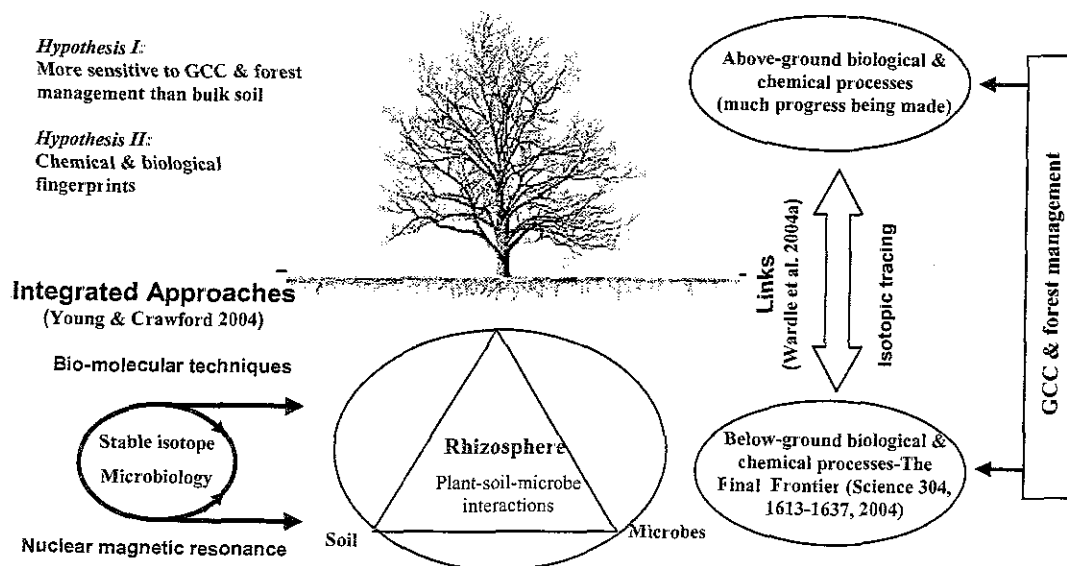
Little is known about the impacts of GCC and forest management on plant-soil-microbe interactions. These interactions mainly occur in the rhizosphere, which is defined as the zone of soil that is affected by the root activity of plants. We view the rhizosphere as the 'hotspot' for plant-soil-microbe interactions - the most chemically and biologically active environment in soil (e.g. Seguin et al. 2004), representing a complex, integrated ecosystem. There is growing need for improving the understanding and management of important below-ground processes. For example, the ecology of below-ground processes and their interactions with above-ground processes has been highlighted in a recent issue of Science: "Soils - The Final Frontier" (Science 304, 11 June 2004).

Understanding rhizosphere C and nutrient cycling processes in relation to rising [CO_2] and temperature is crucial for

predicting the response of forest ecosystems to GCC. The following hypotheses may be advanced to be tested with an integrated approach (see diagram below) (Xu & Chen 2006).

- *The rhizosphere as a 'hotspot' for plant-soil-microbe interactions:* Plant-soil-microbe interactions in the rhizosphere are more sensitive to GCC and forest management than in non-rhizosphere zones, and are closely linked to rhizosphere C and nutrient cycling;
- *Biological and chemical fingerprints in the rhizosphere:* There are biological and chemical fingerprints of GCC and forest management on rhizosphere C and nutrient cycling processes, which are closely linked to above-ground processes.

Annual tree ring width measurements can be used to study tree growth indices of different species at scales from years to centuries in seasonally-distinguished growth environments, particularly in response to climate change and human influences (e.g. Büntgen et al. 2007). Together with tree ring growth measurements, tree ring stable isotope compositions (e.g. Treydte et al. 2006) and nutrient concentrations (e.g. Elhani et al. 2003) may be used to reconstruct past, long-term climate change in different regions of the world, and to assess human influences (e.g. acid deposition and fertilization) on the long-term structure, function and productivity of forest ecosystems.



Rhizosphere study techniques

The quantitative understanding of rhizosphere processes is poor, since the rhizosphere is a difficult system to physically sample and manipulate (e.g. Xu & Chen 2006). Currently there are two commonly-used methodologies to physically separate rhizosphere soil from bulk soil. One is the hand-shaking method (e.g. Seguin et al. 2004). The second approach involves direct (*in situ*) sampling of soil adjacent to roots by thin sectioning and/or placement of different sized mesh materials around roots (Xu & Chen 2006). It is challenging, but necessary to develop sampling techniques and protocols building on the promising hand-shaking method, which takes into account the spatial and temporal variability in the rhizosphere of forest ecosystems.

Microbiological methods

Soil microbial properties, such as microbial biomass C, N and phosphorus (P), respiration, metabolic quotient and enzyme activity, can be very sensitive to GCC and forest management (e.g. Xu & Chen 2006). However, information about the impacts of GCC and forest management on soil microbial properties is rather limited (e.g. Chen et al. 2004, Bastias et al. 2006, Burton et al. 2007). Conventional culture-dependent methods have been used for the measurement of soil microbial composition for more than a hundred years. Nevertheless, only 0.1–1% of soil microorganisms are accessible by these approaches.

Biomolecular techniques

Recent advances in bio-molecular techniques make it possible to apply culture-independent and DNA/RNA nuclear acid-based techniques to analyze the targeted sequences of bacterial or fungal DNA directly extracted from soil (e.g. Xu & Chen 2006). The determination of 16S ribosomal RNA (rRNA) genes and 18S rRNA genes has proved most useful for investigating the diversity and composition of bacteria and fungi, respectively, since these molecules are composed of highly conserved regions and also of regions with considerable sequence variation.

Stable isotope and NMR techniques

Stable isotope techniques are considered a critical component in studies of GCC (e.g. elevated $[CO_2]$) and forest management on soil C and N dynamics (e.g. Xu & Chen 2006) because they are very powerful tools for advancing the understanding of important C and N cycling processes in terrestrial ecosystems. Recent applications of stable isotope techniques to soil biological studies have resulted in significant advances in the understanding of soil microbial processes regulating C and N cycling in terrestrial ecosystems. The combined use of stable isotope and bio-molecular techniques holds great promise, as it allows for the identification of specific microorganisms that are actively involved in particular metabolic processes (e.g. Xu & Chen 2006).

NMR techniques have been increasingly used in soil science, geochemistry and environmental science (e.g. Johnson et al. 2005, Xu & Chen 2006). In particular, ^{13}C NMR has been widely used to improve the understanding of soil organic matter (SOM) quality and composition in relation to terrestrial C and N cycling processes. Natural abundance ^{15}N CP/MAS NMR spectra of SOM have indicated that almost all of the NMR signal intensity lies in the chemical shift region assigned to peptide/amide N (Knicker et al. 1993). In a rare application of ^{14}N -NMR to soils, Mao et al. (2002) have discovered the existence of nitrate-N in soil HA, with the HA nitrate-N closely related to soil N availability and rather responsive to ecosystem management. Advanced NMR techniques need to be assessed for their potential in improving the understanding of rhizosphere C and nutrient

cycling, particularly when combined with stable isotope and bio-molecular techniques (e.g. Knicker 2002).

Tree ring techniques

Recent tree ring research (Xu et al. 2007) shows that rising $[CO_2]$ during 1830s–1970s reduced tree ring width of even-aged European beech and oak forests in central and high Belgium, due to $[CO_2]$ -induced warming and the associated decrease in growing season rainfall. In this 140-year period, acid deposition, closely-coupled with rising $[CO_2]$, resulted in a further decrease in tree ring width under increasing water-limiting conditions in central Europe, possibly due to reduced long-term tree water use efficiency as reflected in tree ring carbon isotope composition ($\delta^{13}C$). The long-term decreases in tree growth and water use efficiency differ between species and between regions. This highlights the likely worsening impact of future climate change on the long-term productivity and biodiversity of beech and oak forests in central Europe, with increasing water stress and more frequent summer heatwaves.

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