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Intramolecular isotope analysis reveals plant ecophysiological signals covering multiple timescales

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Akademisk avhandling

som med vederbörligt tillstånd av Rektor vid Umeå universitet för avläggande av filosofie doktorsexamen framläggs till offentligt försvar i lärosal N440, Naturvetarhuset, onsdagen den 23 januari, kl. 09:00. Avhandlingen kommer att försvaras på engelska.

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Abstract

Our societies' wellbeing relies on stable and healthy environments. However, our current lifestyles, growth-oriented economic policies and the population explosion are leading to potentially catastrophic degradation of ecosystems and progressive disruption of food chains. Hopefully, more clarity about what the future holds in store will trigger stronger efforts to find, and adopt, problem-focused coping strategies and encourage environmentally friendly lifestyles.

Forecasting environmental change/destruction is complicated (*inter alia*) by lack of complete understanding of plant-environment interactions, particularly those involved in slow processes such as plant acclimatisation and adaptation. This stems from deficiencies in tools to analyse such slow processes. The present work aims at developing tools that can provide retrospective ecophysiological information covering timescales from days to millennia.

Natural archives, such as tree-rings, preserve plant metabolites over long timescales. Analyses of intramolecular isotope abundances in plant metabolites have the potential to provide retrospective information about metabolic processes and underlying environmental controls. Thus, my colleagues and I (hereafter we) analysed intramolecular isotope patterns in tree rings to develop analytical tools that can convey information about clearly-defined plant metabolic processes over multiple timescales. Such tools might help (*inter alia*) to constrain plants' capacities to sequester excess amounts of anthropogenic CO_2; the so-called CO_2 fertilisation effect. This, in turn, might shed light on plants' sink strength for the greenhouse gas CO_2, and future plant performance and growth under climate change.

In the first of three studies, reported in appended papers, we analysed intramolecular $^{13}C/^{12}C$ ratios in tree-ring glucose. In six angiosperm and six gymnosperm species we found pronounced intramolecular $^{13}C/^{12}C$ differences, exceeding 10‰. These differences are transmitted into major global C pools, such as soil organic matter. Taking intramolecular $^{13}C/^{12}C$ differences into account might improve isotopic characterisation of soil metabolic processes and soil CO_2 effluxes. In addition, we analysed intramolecular $^{13}C/^{12}C$ ratios in a *Pinus nigra* tree-ring archive spanning the period 1961 to 1995. These data revealed new ecophysiological $^{13}C/^{12}C$ signals, which can facilitate climate reconstructions and assessments of plant-environment interactions at higher resolution; thus providing higher quality information. We proposed that $^{13}C/^{12}C$ signals at glucose C-1 to C-2 derive from carbon injection into the Calvin-Benson cycle via the oxidative pentose phosphate pathway. We concluded that intramolecular $^{13}C/^{12}C$ measurements provide valuable new information about long-term metabolic dynamics for application in biogeochemistry, plant physiology, plant breeding, and paleoclimatology.

In the second study, we developed a comprehensive theory on the metabolic and ecophysiological origins of $^{13}C/^{12}C$ signals at tree-ring glucose C-5 and C-6. According to this theory and theoretical implications of the first study on signals at C-1 to C-3, analysis of such intramolecular signals can provide information about several metabolic processes. At C-3, a well-known signal reflecting CO_2 uptake is preserved. The glucose-6-phosphate shunt around the Calvin-Benson cycle affects $^{13}C/^{12}C$ compositions at C-1 and C-2, while the $^{13}C/^{12}C$ signals at C-5 and C-6 reflect carbon fluxes into downstream metabolism. This theoretical framework enables further experimental studies to be conducted in a hypothesis-driven manner. In conclusion, the intramolecular approach provides information about carbon allocation in plant leaves. Thus, it gives access to long-term information on key ecophysiological processes, which could not be acquired by previous approaches.

The abundance of the hydrogen isotope deuterium, δD, is important for linking the water cycle with plant ecophysiology. The main factors affecting δD in plant organic matter are commonly assumed to be the δD in source water and leaf-level evaporative enrichment. Current δD models incorporate biochemical D fractionations as constants. In the third study we showed that biochemical D fractionations respond strongly to low ambient CO_2 levels and low light intensity. Thus, models of δD values in plant organic matter should incorporate biochemical fractionations as variables. In addition, we found pronounced leaf-level δD differences between α-cellulose and wax n-alkanes. We explained this by metabolite-specific contributions of distinct hydrogen sources during biosynthesis.

Overall, this work advances our understanding of isotope distributions and isotope fractionations in plants. It reveals the immense potential of intramolecular isotope analyses for retrospective assessment of plant metabolism and associated environmental controls.

Keywords

NMR spectroscopy, tree ring, isotope ratio, isotope effect, intramolecular $^{13}C/^{12}C$ signal, carbon allocation, acclimation, plant performance, climate reconstruction, plant ecophysiology

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