Isotope applications to soil science at the University of Alberta — an historical perspective

Brett C. Feland and Sylvie A. Quideau

Abstract: For the past 70 yr, researchers in the Soil Science/Renewable Resources Department at the University of Alberta have used isotopes to study topics of ecological importance. This review highlights the soil isotope research conducted within our department over this time, including an historical overview of studies of interest. Analytical techniques and advances in instrumentation are discussed, focusing on the measurement of light stable isotope ratios (i.e., for C, H, N, S, and O) using isotope ratio mass spectrometry (IRMS). Early soil isotope work (1950–2000s) focused on agricultural soils and soil fertility issues. These studies included the use of radioactive isotopes such as $^{14}$C and $^{35}$S, and (or) artificially enriched stable isotopes including $^{15}$N-labelled fertilizers. More recently (2000–present), the scope of research widened to include natural-abundance stable isotope ratio studies as higher-sensitivity IRMS systems became more prevalent. Current isotope research topics include N biogeochemistry in natural and managed ecosystems, land management effects on greenhouse gas emissions, carbon cycling in northern landscapes, paleo-reconstruction in peatlands, carbon sequestration in boreal forests, and biodegradation of petroleum hydrocarbons. Further technological progress also enabled new techniques such as compound-specific IRMS analysis, including $\delta^{13}$C and $\delta^2$H measurements of soil $n$-alkanes and phospholipid fatty acids. In conclusion, current IRMS instrumentation presents unparalleled opportunities for multidisciplinary research to track carbon, plant nutrients, and pollutants as they move through soils.

Key words: history of soil science, isotope ratio mass spectrometry, isotope biogeochemistry.

Résumé : Depuis 70 ans, les chercheurs du département des sciences du sol et des ressources renouvelables de l’Université de l’Alberta utilisent des isotopes pour étudier divers sujets présentant de l’importance en écologie. Les auteurs parlent des travaux réalisés avec des isotopes dans ce département au fil des ans. Ils passent notamment en revue les études historiques les plus intéressantes. Il est question d’analyses techniques et des progrès réalisés par l’instrumentation, et les auteurs insistent sur la quantification des rapports des isotopes stables légers (C, H, N, S et O) par la spectroscopie de masse des rapports isotopiques (SMRI). Les premiers travaux sur l’application des isotopes à la pédologie (1950–2000) portaient sur des sols agricoles et des problèmes de fertilité. Ces études comprenaient l’usage d’isotopes radioactifs comme le $^{14}$C et le $^{35}$S ou d’isotopes stables enrichis artificiellement, notamment les engrais marqués au $^{15}$N. Plus récemment (2000–aujourd’hui), les recherches se sont élargies pour inclure des études sur les rapports des isotopes stables qui abondent dans la nature grâce à l’avènement de systèmes SMRI plus sensibles. Au nombre des travaux actuels, mentionnons la biogéochimie de l’azote dans les écosystèmes naturels et aménagés, les conséquences de la gestion des terres sur les émissions de gaz à effet de serre, le cycle du carbone dans le Nord, la paléo-reconstruction des tourbières, la séquestration du carbone par la forêt boréale et la biodégradation des hydrocarbures. D’autres progrès techniques ont engendré de nouvelles technologies comme l’analyse de composés spécifiques par la SMRI, y compris la quantification des n-alkanes et des acides gras phospholipidiques dans le sol avec le $\delta^{13}$C et le $\delta^2$H. En conclusion, les appareils SMRI modernes offrent des possibilités sans précédent pour la recherche multidisciplinaire sur les déplacements du carbone, des oligoéléments et des polluants dans le sol. [Traduit par la Rédaction]

Mots-clés : historique de la pédologie, spectrométrie de masse des rapports isotopiques, biogéochimie des isotopes.

Received 29 November 2019. Accepted 12 February 2020.

B.C. Feland and S.A. Quideau. Department of Renewable Resources, University of Alberta, Edmonton, AB T6G 2R3, Canada.

Corresponding author: Brett C. Feland (email: brett.feland@ualberta.ca).

Copyright remains with the author(s) or their institution(s). This work is licensed under a Creative Commons Attribution 4.0 International License (CC BY 4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author(s) and source are credited.
Introduction

The Renewable Resources Department (formerly the Soil Science Department) at the University of Alberta just celebrated its 100th anniversary in 2019. For nearly 70 of the past 100 yr, soil scientists in that department have been using isotopes, the study of which is incredibly useful in ecological research. For those unfamiliar with isotopes, simply put, the atoms of most individual elements can exist in more than one form, whereas still exhibiting the same (or nearly the same) chemical properties. These different types of atoms of the same element are known as isotopes, and they differ from one another only by the number of neutrons in their nuclei. As a result, these isotopes have different masses (Michener and Lajtha 2007). For example, the three most common isotopes of carbon are carbon-12 (12C), carbon-13 (13C), and carbon-14 (14C). All atoms of carbon contain six protons and six electrons, but the isotopes 12C, 13C, and 14C contain 6, 7, and 8 neutrons each, with atomic mass numbers of 12, 13, and 14, respectively. Approximately 98.9% of the carbon found in nature is 12C, 1.1% is 13C, whereas a trace amount is 14C. Both 12C and 13C are stable, which is to say they do not change over time, whereas 14C is unstable, with a half-life of 5700 yr. It emits β− radiation, gradually decaying to 14N (Garnier-Laplace and Roussel-Debet 2001).

Many ecological studies utilize the stable isotopes of light elements, in particular C, H, N, S, and O. Not only are these elements most frequently involved in biological processes, but the relative percent mass difference for light isotopes with a difference of only one or two atomic mass numbers is greater than for heavier elements (Michener and Lajtha 2007). For example, 13C is 8.3% heavier than 12C. This greater relative mass difference results in greater differences in isotope fractionation (accumulation or depletion of one particular isotope over another) due to naturally occurring mechanisms, thus making them easier to measure than heavy isotopes.

Although the approximate proportions of each naturally occurring isotope in the environment are known, the precise proportions of each isotope in a sample can vary for numerous reasons (Michener and Lajtha 2007). These can include natural enrichment/depletion, an increase/decrease in the relative concentration of one isotope (such as 13C, 2H, 15N, 34S, or 18O) over other isotopes of the same element. Natural enrichment or depletion can occur through biochemical reactions; specifically, in biological systems, the product of any enzymatic reaction (e.g., photosynthesis, nitrification, and denitrification) is more depleted than the corresponding substrate, unless the entire substrate pool gets consumed (Michener and Lajtha 2007). In addition, artificial enrichment/depletion, such as the application of 15N-labelled fertilizer allows tracking of the label as it travels through the soil–plant system. The relative amounts of isotopes in samples are typically measured by isotope ratio mass spectrometry (IRMS) after converting the target elements in the sample to simple gases such as CO2 for C, H2 for H, N2 for N, SO2 or SF6 for S, and CO for O (Michener and Lajtha 2007). Results are generally reported as observed fractions of the rare isotope to the common isotope. The isotope ratio is expressed in delta notation, δ, in units of permil, ‰, relative to the isotope ratio of an internationally accepted standard. The light stable isotope ratios frequently measured by ecological researchers are therefore δ13C/12C, δ2H/1H, δ15N/14N, δ34S/32S, and δ18O/16O.

The isotopic ratios of a sample can be used to determine where certain elements in the sample originated from. IRMS has become an important tool in environmental research, with numerous applications (Chabbi and Rumpel 2012). Researchers can use δ13C isotope ratio data to trace carbon fluxes in the terrestrial carbon cycle (Brüeggermann et al. 2011). In the above-mentioned case of labelled 15N fertilizer application, IRMS can be used to trace where that fertilizer travels, how much is taken up by the target crop, and how much is lost to other parts of the environment (Malhi and Nyborg 1983). More recent developments in soil isotope techniques include the use of compound-specific IRMS analyses to track δ13C ratios in soil microbial biomarkers, i.e., phospholipid fatty acids (PLFAs) (Glaser 2005; Watzinger 2015). The possibilities for isotope ratio research in soil science are endless, and numerous examples of IRMS applications in soil science will be discussed in this review, including the use of natural abundance and enrichment techniques.

This review focuses on isotope work conducted within the Soil Science Department from the 1950s to 2019 at the University of Alberta, and within the Renewable Resources Department following the merger of the Soil Science and Forestry Departments in 1994. Figure 1 shows the number of articles published using soil isotopes by members of our department over this time span. The list of references used to prepare this figure was compiled from a combination of search results for soil isotope papers from the University of Alberta using the Web of Science and Scopus databases (up until 15 July 2019), along with internal departmental records. Although every effort has been made to include mention of all researchers within the department who have conducted studies using soil isotopes over the last 70 yr, it is possible that some contributors may have been inadvertently left out of this discussion. Additionally, to limit the breadth of this review to a manageable scope, this article focuses on members of the University of Alberta’s Soil Science/Renewable Resources Department. However, other researchers at this university have and continue to conduct environmental research using stable isotopes, including our colleagues in the Department of Chemistry, the Department of Earth and Atmospheric
Sciences, and the Department of Agricultural, Food, and Nutritional Sciences.

Although this review focuses on work at the University of Alberta, several other universities in Canada also have long histories of soil and other environmental stable isotope research. The Ján Veizer Stable Isotope Laboratory (formerly the G.G. Hatch Lab) at the University of Ottawa has been at the forefront of light stable isotope research and education for many years, as has the University of Saskatchewan. The Universities of Calgary, Lethbridge, British Columbia, and Waterloo are all home to researchers involved in soil stable isotopes, and this is by no means a comprehensive list.

Early Soil Isotope Research

Radioactive isotope work

Although the main focus of this article is on light stable isotope research in soil science at the University of Alberta, such a discussion would not be complete without mentioning the numerous soil studies conducted using radioactive isotopes. Below is a brief overview of such research from members of the Soil Science/Renewable Resources Departments.

$^{35}$S

Isotopes have been used in soil science research at the University of Alberta since the 1950s. The earliest such studies were conducted by C.F. Bentley and D.B. Scott, along with Bentley’s graduate students D.V. Cormack and D.J. Hoff (Cormack et al. 1951; Bentley et al. 1955). These researchers used radioactive $^{35}$S-labelled fertilizer to trace uptake in legume and barley crops, using a Geiger–Müller tube to count activity. The test crops were grown at the Breton Plots, a research facility established in 1929 that is still in use by the University of Alberta today. Further, similar research was later continued by Pawluk and Bentley (1964). Additionally, R.A. Drijber, a Ph.D. student supervised by W.B. McGill who graduated in 1993, used $^{35}$S to study the use of sulfonolipids as biomarkers for Cytophaga and Flexibacter bacteria in soils (Drijber and McGill 1994a, 1994b).

$^{14}$C

Radioactive $^{14}$C was used by several soil science researchers at the University of Alberta, the first of whom was W.B. McGill. McGill received his Ph.D. from the University of Saskatchewan in 1971, and along with J.A. Shields and E.A. Paul (McGill et al. 1975), used $^{14}$C and $^{15}$N to trace metabolic pathways in soil microbes. In these studies, $^{14}$C radioactivity was measured, and $^{15}$N content was determined using an Atlas Model GD 150 mass spectrometer. McGill later came to the University of Alberta and continued his research with soil isotopes, much of which will be discussed in the stable isotopes section of this article. However, his research still included radioactive carbon-14. For example, McGill’s Ph.D. student C.M. Monreal used $^{14}$C to study cystine cycling in soil (Monreal and McGill 1989a, 1989b, 1989c, 1989d).

N.G. Juma is another soil scientist who used $^{14}$C in his research, along with his graduate students G.D. Dinwoodie and J.G. Xu. Dinwoodie and Juma used $^{14}$C to study carbon dynamics in barley plots, using a Minaxi β Tri-Carb 4000 series scintillation counter to measure $^{14}$C activity (Dinwoodie and Juma 1988). Later, Xu and Juma used $^{14}$C to study kinetics in root systems (Xu and Juma 1993, 1994, 1995). We will see more of Juma’s contributions to soil isotope research later in this paper.

$^{32}$P and $^{137}$Cs

Although studies including soil phosphorous data were frequently published by University of Alberta
researchers, a few using radioactive $^{32}$P were also conducted. J.A. Robertson and his student P.K. Omanwar used $^{32}$P to study soil exchange kinetics in the 1970s (Omanwar and Robertson 1971). Lastly, the work of M.J. Dudas and C.P. Maulé involving radioactive cesium is worth mentioning. These authors used $^{137}$Cs, produced by nuclear testing in the 1950s and 1960s and deposited in the environment, to trace soil erosion (Maulé and Dudas 1989). These studies followed earlier work by E. de Jong et al. at the University of Saskatchewan (De Jong et al. 1982).

**Stable isotope work**

The earliest soil stable isotope research conducted by members of the University of Alberta Soil Science Department was published in 1968 by R.P. Wellman, F.D. Cooke, and H.R. Krouse (Wellman et al. 1968). In this study, the authors measured the fractionation of $^{15}$N/$^{14}$N during the microbial reduction of nitrate/nitrite to N$_2$. Other early examples include the previously mentioned work of W.B. McGill, E.A. Paul, et al. using $^{15}$N in conjunction with $^{14}$C to study soil microbial pathways (McGill et al. 1975; McGill and Paul 1976).

$^{15}$N stable isotope analysis would become a major theme for several researchers at the University of Alberta. M. Nyborg, a professor in the Department of Soil Science for many years, is one such example, and his isotope-focused research examined soil fertility and fertilizer N and S uptake by plants. Together with S.S. Malhi, who received his Ph.D. in 1978 while studying under M. Nyborg, the two authors published several $^{15}$N papers over the years. In one study, they used $^{15}$N-labelled fertilizer, applied in the fall, to determine how much N was lost due to denitrification, immobilization, and leaching over the winter (Malhi and Nyborg 1983). They determined that fall application of fertilizer resulted in large overwinter losses, which were due to denitrification rather than leaching. Nyborg published further research on this topic with D.J. Heaney (Heaney and Nyborg 1988; Heaney et al. 1992).

In addition to application timing and fertilizer recovery, Malhi and Nyborg’s $^{15}$N fertilizer research included studies of fertilizer placement methods. They also used different types of fertilizer $^{15}$N sources, such as nitrate and urea, and worked with several other students and researchers including E.D. Solberg, R.C Izaaurralde, M. Zhang, and R. Pradhan (Malhi et al. 1989; Nyborg et al. 1990; Malhi and Nyborg 1991; Pradhan et al. 1998; Zhang et al. 2000). This research typically involved the application of isotopically enriched $^{15}$N fertilizer to soil systems and subsequent analysis by IRMS. Much of these analyses were conducted using a Micromass 602C Mass Spectrometer, which was acquired by the Soil Science Department in 1975. This was later replaced by a Stable Isotope Ratio Analyzer (SIRA) 10 from VG Isogas in the UK, acquired in 1987, and often used coupled to an Automatic Nitrogen Analyzer 1500.

N.G. Juma, another long-serving professor at the Soil Science Department at the University of Alberta, used $^{15}$N in his soil research. He studied C and N transformation in the soil, developing models of nutrient cycling. Some of his earlier publications involving isotopes, conducted in collaboration with the University of Saskatchewan, include studies of soil N mineralization kinetics and N immobilization in the presence of nitrification inhibitors (Juma and Paul 1983; Juma et al. 1984). P.M. Rutherford, a Ph.D. student of N.G. Juma, used $^{15}$N-enriched urea to compare plant–soil N dynamics in Black Chernozemic and Gray Luvisolic soils cropped to barley (Rutherford and Juma 1989). In subsequent work, Rutherford and Juma (1992) used $^{15}$N (and $^{14}$C) to show that protozoan grazing on soil bacteria induced more significant mineralization of bacterial N and C in coarse-textured soil than in medium- and fine-textured soils. Working with Juma, M.Sc. student K. Haugen-Kozyra studied N dynamics in barley–soil systems under conventional and zero tillage. More $^{15}$N-enriched urea was converted into organic N under zero tillage than conventional tillage suggesting better conservation of N in the former system (Haugen-Kozyra et al. 1993).

Much of the stable isotope ratio research conducted in the 1970s and 1980s by members of the Soil Science Department was done using $^{15}$N. However, G.A. Spiers’ research in clay mineral weathering made use of $^{18}$O. Spiers was supervised by M.J. Dudas and S. Pawluk for his M.Sc. (1982) and Ph.D. (1990), respectively. These researchers, in collaboration with K. Muehlenbachs from the Geology Department (later the Earth and Atmospheric Sciences Department), used the BrF$_5$ method to isolate and measure $^{18}$O in clay separates isolated from soil samples (Spiers et al. 1985).

Some further discussion of W.B. McGill’s research as a professor in the Soil Science/Renewable Resources Department is warranted. McGill’s work can broadly be described in two categories; soil biogeochemistry, and the fate and transport of organic compounds in soil. In 1985, along with colleagues from Duke University and Oregon State University, he published a study using $^{15}$N to trace alder-fixed N in conifers (Binkley et al. 1985). Notably, in this research, the authors measured natural-abundance $^{15}$N in their samples. Up until this point, most isotope ratio studies at the University of Alberta used artificial $^{15}$N enrichment to trace soil N. Another illustrative example of McGill’s use of $^{15}$N in soil research is the work conducted by his Ph.D. student D.C. Jans-Hammermeister studying soil–plant dynamics in barley crops by green manuring with field pea (Jans-Hammermeister et al. 1994).

The works of one additional soil researcher from the University of Alberta, D.J. Pluth, are worth mentioning. In collaboration with McGill, Pluth and his Ph.D. student, G.E. Nason, as well as R.T. Hardin from the Department of Animal Science, studied foliar N dynamics in Douglas-fir with $^{15}$N isotopically enriched.
ammonium nitrate and urea (Nason et al. 1990). Also, Pluth and S.E. Macdonald, the current chair of the Renewable Resources Department, co-supervised Ph.D. student A.G. Mugasha, who studied at the University of Alberta while on leave from the Tanzania Forestry Research Institute. Mugasha and Pluth used $^{15}$N-labelled urea fertilizer to study soil and tree uptake patterns in tamarack and black spruce stands (Mugasha and Pluth 1994a, 1994b). Pluth also collaborated with researchers from the Swedish University of Agricultural Sciences and Forestry Canada to characterize $^{15}$N fractionation in soils and Scots pine (Nömmik et al. 1994). Although the IRMS analysis for this publication was not conducted at the University of Alberta, it is another of the rare, early examples of natural-abundance $^{15}$N isotope ratio studies.

Recent and current soil isotope research (early 2000s–present)

After a lull in publications related to soil isotope research in the late 1990s, new faculty hires, starting with S.X. Chang and S.A. Quideau in the early 2000s, generated a fresh wave of research at the University of Alberta, which resulted in a steady increase in publications since then (Fig. 1). In addition, isotope research evolved from its early focus on soil fertility to encompass broader environmental issues. Current research within the Department of Renewable Resources that utilize isotopic tools includes a wide range of subjects such as: N biogeochemistry in natural and managed ecosystems (S.X. Chang, M.D. MacKenzie, and S.A. Quideau); land management effects on greenhouse gas emissions (G. Hernandez-Ramirez); carbon cycling in northern landscapes (D. Olefeldt); paleo-reconstruction in peatlands (W. Shotyk); carbon sequestration in boreal forests (S.X. Chang and S.A. Quideau); and biodegradation of petroleum hydrocarbons (T. Siddique). In conjunction with a change in focus, research has shifted from using radioactive to stable isotopes, and recent projects have taken advantage of technological advances to measure compound-specific and position-specific isotopic values.

Nitrogen isotopes

Enriched $^{15}$N work

Some of our department’s more current work has continued to take advantage of $^{15}$N-enriched fertilizers to trace the source of N for plant uptake. For example, in the group led by S.X. Chang, $^{15}$NH$_4$NO$_3$ and NH$_4$$^{15}$NO$_3$ were used to study the preferential uptake of NH$_4^+$ vs. NO$_3^-$ by trembling aspen and hybrid aspen seedlings (Choi et al. 2005a). The uptake and recovery of $^{15}$N per tree was higher for the hybrid aspen than for the trembling aspen, and both species exhibited a higher recovery of $^{15}$NH$_4^+$ than $^{15}$NO$_3^-$, which goes against the common assumption that deciduous tree species are better adapted to nitrate than ammonium nutrition. The $^{15}$N pool dilution method has been another commonly used isotopic tool in S.X. Chang and S.A. Quideau’s groups to quantify gross N transformation rates in a range of soil environments and land uses, including forestry and agriculture, as well as in landscapes undergoing reclamation or other land use changes (McMillan et al. 2007; Lang et al. 2010, 2019; Cheng et al. 2012, 2019). In this method, labelled inorganic ammonium and nitrate are applied to the soil to quantify gross ammonification and nitrification rates. In addition, $^{15}$N-enriched litter obtained from plants grown with $^{15}$N-labelled fertilizer can be used to quantify mineralization rates. C. Norris (Ph.D. with S.A. Quideau) used this approach to quantify N cycling in natural and reclaimed soils (Norris et al. 2018).

Natural $^{15}$N abundance

In soils, natural variations in N stable isotopes result from diverging $\delta^{15}$N values of external N sources and internal ecosystem transformations. Many of the internal, microbial-driven N processes (e.g., nitrification, denitrification, and immobilization) result in measurable discrimination between substrate and product, where the product is more depleted in N than the substrate. This causes a progressive $^{15}$N enrichment of the residual soil N pool, which together with plant $\delta^{15}$N values, can be used as an index of the ecosystem N status (Fang et al. 2011). M.D. MacKenzie, a soil scientist studying fertility and N cycling related to disturbance in forest ecosystems, used this natural variation in N stable isotopes to follow the ecological response to N deposition of reclaimed forest stands in the oil sands region of Alberta (Hemsley et al. 2019). The soil, foliar, and root pools of jack pine stands were more enriched in $^{15}$N compared with the trembling aspen stands. Hence in this study, isotopic analysis provided one of the key indicators that pine was more sensitive to the additional N deposition and that these ecosystems may be approaching saturation. Similarly, many of the studies in S.X. Chang’s group have taken advantage of $\delta^{15}$N measurements in foliage and woody tissues to evaluate the effects of land use on plant nutrition. Some of these studies have (i) quantified the effects of soil compaction and forest floor removal on N cycling in lodgepole pine and Douglas-fir stands (Choi et al. 2005b), and trembling aspen (Tan et al. 2006); (ii) explored the relationship between N deposition and tree ring chemistry of jack pine and trembling aspen stands (Jung et al. 2013); and (iii) studied the relationship between soil nitrification and foliar $\delta^{15}$N in trembling aspen and jack pine forests of different stand age (Hu et al. 2014).

Variations in soil N isotopes linked to changes in environmental conditions may be too small or short lived to be useful as diagnostic tools. In contrast, one systematic variation has been linked to trophic levels, with the lower positions being relatively less enriched than the higher trophic levels, and this consequently has been recognized as a powerful tool for reconstructing trophic relationships...
in soil food webs (Scheu and Falca 2000). Irma Diaz (Ph.D. in S.A. Quideau’s group) used the $\delta^{15}N$ values of soil mite bodies to position them along a trophic gradient in the forest floors of coniferous and deciduous stands of western Canadian boreal mixedwood forest. In addition to mature (control) stands, she also studied the potential effect of clear-cutting on these trophic guilds (Diaz-Aguilar and Quideau 2013). The difference of $\delta^{15}N$ values in mites placed them in three distinct trophic guilds: detritivores, omnivores, and predators. Furthermore, the trophic position of these forest floor mites stayed constant regardless of their habitat.

**Optical spectroscopic techniques**

Globally, nitrous oxide represents more than half of the agriculture-derived greenhouse gas emissions. However, wide spatial and temporal variation of soil $N_2O$ fluxes brings high uncertainty to estimates of rates and sources. Two formation pathways for $N_2O$ have been identified, denitrification by anaerobic bacteria, or as a by-product of aerobic nitrification (Baggs 2008). The development of cavity ring-down spectroscopy, which allows measurements of site-specific isotopic composition as well as bulk $\delta^{15}N$ in $N_2O$, now allows separation of the two pathways. Namely, the difference in $\delta^{15}N$ values between the central and terminal positions of $N_2O$ can be used to differentiate between the nitrification (mean value of 33‰) and denitrification (mean value of 0‰) pathway (Ibraim et al. 2019). This approach is currently in use by G. Hernandez-Ramirez to determine the effects of various organic amendments on nitrous oxide emissions from agricultural soils (unpublished results).

**Carbon isotopes**

**Natural abundance $^{13}C$**

Elucidating humification processes responsible for the transformation of plant residues into soil organic matter is a key to understanding global carbon biogeochemistry and the controls of carbon accumulation in soils. Peatlands and other organic soils are unique in that their organic matter is not stabilized by interactions with the soil mineral matrix as is the case with other soil types. Yet, peatland organic matter can be preserved for centuries and sometimes millennia, and as such, can serve as a valuable archive of past environmental conditions. W. Shotyk, a soil and water geochemist interested in the cycling of trace metals, has been using sphagnum moss and peat bogs to reconstruct their natural and anthropogenic sources to the atmosphere. Numerous experimental approaches and instrumental tools have been proposed and utilized to study peat formation processes. In regard to the specific use of light stable isotopes, researchers in W. Shotyk’s group have investigated the value of $\delta^{13}C$ and $\delta^{15}N$ measurements as indices of peat humification extent (Zaccone et al. 2018).

One of the most common themes in the use of the natural abundance $^{13}C$ technique is to use $\delta^{13}C$ data from plant samples to infer the effect of different factors (management or edaphic) on plant water use efficiency (see for example, the following work from S.X. Chang: Choi et al. 2005b, 2007; Tan et al. 2006; Matsushima et al. 2014). Additionally, as soil organic matter becomes progressively enriched in $^{13}C$ with increasing humification, its isotopic composition can be a useful tool to explore decomposition processes and the extent of microbial processing (Quideau et al. 2003). This approach has been successfully applied to the study of organic matter processes in reclaimed (Turcotte et al. 2009), arboreal (Enloe et al. 2010), riparian (Card et al. 2010), and agricultural (Kipps 2015) soils, and harvesting effects in forest soils (Hannam et al. 2005; Sewell 2018).

**Enriched $^{13}C$**

Carbon dioxide ($^{13}CO_2$) pulse labelling of plants is a powerful method to trace the fate of carbon fixed by photosynthesis. This technique was used by C. Arevalo (Ph.D. with S.X. Chang), who measured $^{13}C$ incorporation in different plant tissues (leaves, stems, and roots) of two hybrid poplar clones, and studied how that incorporation was affected by plant age (Arevalo et al. 2010). In addition, C. Norris and S.A. Quideau combined $^{13}C$-pulse labelling with characterization by solid-state $^{13}C$ nuclear magnetic resonance (NMR) spectroscopy to track the incorporation of the $^{13}C$ label into aspen leaf and root macromolecules (Norris et al. 2012). This work was conducted in conjunction with R.E. Wasylischen’s solid-state NMR research group in the Department of Chemistry. Combining C and O isotope composition ($^{13}C$, $^{14}C$, and $^{18}O$) to molecular characterization by NMR allowed the quantitative comparison of cellulose preparation techniques (Gaudinski et al. 2005). Lastly, combining soil physical fractionation and isotopic and NMR analyses allowed Norris et al. (2011) to assess how soil carbon stabilization under pine vegetation varied along a climosequence from Saskatchewan to Manitoba.

D. Olefeldt works on carbon cycling in boreal to arctic landscapes, with a focus on impacts of permafrost thaw, wildfire, droughts, and human activities on carbon storage. Northern permafrost peatlands store more carbon per unit area than any other biome on earth. Increased air temperature, and associated increased fire frequency, are both contributing to the rapid thawing of these permafrost soils, with the concomitant release of carbon to the atmosphere and possibly to aquatic systems. By combining measurements of $^{14}C$ of dissolved organic carbon and $CO_2$ released from northern permafrost peatlands to $^{13}C-CO_2$ analyses, D. Olefeldt and his group were able to co-currently determine the age and sources of $CO_2$ to streams and to differentiate between $CO_2$ released from weathering vs. soil organic matter degradation (Burd et al. 2018; Estop-Aragonés et al. 2018). In addition, $^{13}C-CH_4$ chamber measurements were used to assess whether methane was produced via acetoclastic or hydrogenotrophic pathways in wetland soils of
different permafrost conditions and from thermokarst ponds in peatlands (Hutchins et al. 2020). Lastly, $^{13}$C-labelled glucose was added to peat during anaerobic incubations to test hypotheses of priming — again to study whether peat following permafrost thaw is vulnerable to rapid decomposition (unpublished results).

**Compound specific isotopic analysis (CSIA)**

Stable isotope probing (SIP) of microbial PLFAs, lipids found in microbial membranes, is a method that tracks the incorporation of labelled substrates into distinct microbial groups found in the soil environment. The label can be added as $^{13}$C-enriched CO$_2$ and followed from root exudates into rhizospheric communities (Béasse 2012). The label can also be added as a simple substrate (e.g., $^{13}$C-enriched glucose) or as more complex $^{13}$C-enriched plant materials. For instance, Norris et al. (2016) used double-labelled aspen litter ($^{13}$C and $^{15}$N) to compare soil microbial activity and function in aspen and spruce stands, which are typical of the boreal mixed-wood landscape of western Canada. Similarly, Lloret and Quideau (2015) incubated soils sampled under both aspen and spruce to study how microbial communities processed different $^{13}$C-labelled substrates (glucose, leaves, and roots). Carbon assimilation by microbes was tracked by isotope probing of PLFAs, and measurement of evolved $^{13}$C-CO$_2$ allowed quantification of the percent of CO$_2$ coming from each added substrate. Incubation of $^{13}$C-labelled aspen litter in spruce soils mimics the effects of anticipated future vegetation shifts, and results suggested that shifting from spruce to aspen in the boreal forest may increase microbe-driven carbon stabilization. More recently, Lejoly et al. (2020) used a similar approach to compare carbon processing in natural and reclaimed sandy soils.

More than half of the total carbon in forest soils is stored below a depth of 20 cm. Although forest floor carbon is known to turn over quite rapidly, the stability of carbon stored deeper in forest soil profiles still remains uncertain. Quideau et al. (2018) compared carbon stability in Luvisolic and Podzolic B horizons by measuring total soil respiration and explored the potential of measuring natural $^{13}$C abundance of the respired CO$_2$ to differentiate between sources. The natural abundance $^{13}$C composition of microbial PLFAs was observed to differ between rhizosphere and bulk soil; namely, rhizosphere PLFAs showed $^{13}$C depletion compared with bulk forest floor, indicating that rhizosphere microbes were accessing more recently fixed carbon than in bulk soil (Thacker 2018). Lastly, a multi-isotope approach may be the best strategy to constrain soil processes and elemental fluxes. For instance, Paul and Quideau (2020) used $^{13}$C and $^2$H analyses of n-alkanes to contrast humification processes in reconstructed, peat-dominated soils, and natural, native forest soils.

T. Siddique, an environmental soil chemist in the Department of Renewable Resources, has been using DNA-SIP to identify the specific microorganisms involved in the biodegradation of petroleum hydrocarbons. In particular, labelled toluene was used as a model compound for BTEX in biodegradation study under methanogenic conditions (Zamir et al. 2012). Finally, the biodegradation of naphtanic acids, contaminants found in tailing ponds of Northern Alberta’s oil sands, was followed using $^{13}$C-labelled surrogates. More specifically, $^{13}$C-PLFA analysis allowed the authors to differentiate microbial uptake of $^{13}$C-enriched surrogates and $^{13}$C-depleted biogenic methane (Ahad et al. 2018).

**IRM Instrumentation**

**Early instrumentation**

In 1975, the Soil Science Department at the University of Alberta obtained its first IRMS system, a Micromass 602 Mass Spectrometer. Figure 2a shows a photo of this instrument in operation by technician C. Nguyen in 1985. Early IRMS instruments generally operated as dual-inlet systems, alternating between the analysis of sample gas and reference gas. Such systems are still in use today, as the dual inlet approach has several advantages, including high precision (Michener and Lajtha 2007) and low consumption rates of reference gases. However, continuous-flow IRMS systems eventually grew in popularity. They permitted the analysis of sample gases like CO$_2$ and N$_2$ immediately after preparation, such as from the outlet of a combustion elemental analyzer (i.e., continuous flow elemental analyzer – isotope ratio mass spectrometry, EA–IRMS). This allowed for the rapid, automated measurement of isotope ratios from weighed solid samples, including soils and plant material. The Micromass unit in the Soil Science Department was later replaced in 1987 by a VG Isogas SIRA 10 Continuous Flow IRMS, interfaced to a Carlo-Erba NA1500 Strumentazione combustion elemental analyzer. These instruments were used for $\delta^{13}$C and $\delta^{15}$N stable isotope ratio analysis.

As mentioned previously, early studies in soil isotope ratios were primarily conducted using samples isotopically enriched in $^{15}$N and (or) $^{13}$C. As instruments were designed and built with higher sensitivity and precision, such research began to include measurements on natural-abundance samples, with no isotopic enrichment. In 2003, IRMS capabilities within the Renewable Resources Department continued to expand with the acquisition of a ThermoFinnigan Delta+ Advantage IRMS purchased from Isomass Scientific. This unit was a workhorse in the Soil Science/Renewable Resources Department for many years. It was interfaced to a Costech 4010 Elemental Analyzer through a ConFlo III continuous-flow system for EA–IRMS. Like the VG Isogas unit, this was used for bulk $\delta^{13}$C and $\delta^{15}$N analysis of soils, plant material, and other ecological samples, but the higher sensitivity of the ThermoFinnigan instrument allowed for other applications as well. It was also interfaced to a gas chromatography–combustion (GC–C)
unit, which, for the first time, enabled researchers at the University of Alberta to conduct compound-specific δ\(^{13}\)C isotope ratio analysis of soil extracts. Though no longer used for EA–IRMS or GC–C–IRMS, the ThermoFinnigan unit is still used today, interfaced to a GasBench II system for δ\(^{13}\)C analysis of CO\(_2\).

**Current configuration**

In 2016, the acquisition of a suite of new instruments and peripherals at the Renewable Resources Department established the Stable Isotope Facility for Ecosystem Research (SIFER). A photograph of this facility is shown in Fig. 2b. This laboratory is currently equipped with three isotope ratio mass spectrometers. The first is the ThermoFinnigan Delta+ Advantage IRMS from 2003 discussed earlier. In 2016, this IRMS was connected to a total organic carbon (TOC) interface from Isomass Scientific which, along with an OI 1030W TOC analyzer, permits the δ\(^{13}\)C analysis of total or dissolved organic or inorganic carbon in water samples. The rest of the systems currently used in SIFER were all obtained in 2016.

For bulk EA–IRMS analysis, we use a Thermo Scientific Delta V Advantage IRMS with ConFlo IV continuous-flow interface. A Thermo Scientific Flash 2000 HT Plus Elemental Analyzer Unit completes this system. When operated in combustion mode, this allows for bulk δ\(^{13}\)C and δ\(^{15}\)N analysis in solid samples, much like with the previous ThermoFinnigan and Costech systems, but with better sensitivity. However, when used in pyrolysis mode, bulk analysis of δ\(^{2}\)H and δ\(^{18}\)O in solid samples is also possible. A liquid autosampler and injection system permits δ\(^{2}\)H and δ\(^{18}\)O analysis of water samples as well.

The last IRMS system currently housed in SIFER is our highest sensitivity instrument, a Thermo Scientific Delta V Plus IRMS. This instrument is equipped with two different chromatography systems and is used for compound-specific isotope ratio studies. A Thermo Scientific Trace 1310 GC and GC Isolink II enable compound-specific analysis of GC samples for δ\(^{12}\)C and δ\(^{15}\)N (combustion mode), or δ\(^{2}\)H (high-temperature conversion mode). This system has numerous potential applications, allowing isotope ratio analysis of not just bulk samples, but individual chemical species within each sample. So far, we in SIFER have analyzed δ\(^{13}\)C in PLFA extracts from soil samples, and δ\(^{2}\)H in n-alkanes in soil extracts. For liquid chromatography samples, a Thermo Scientific Ultimate 3000 LC system with LC Isolink permits compound-specific δ\(^{13}\)C analysis in aqueous matrices. Finally, for greenhouse gas isotope ratio studies, SIFER is equipped with a Picarro G2201-i cavity ring down spectroscopy system. This instrument allows for the rapid analysis of δ\(^{13}\)C in CO\(_2\) and CH\(_4\) in gas samples and is field portable.

**IRMS technicians**

Numerous technicians and support staff members in the Soil Science/Renewable Resources Department were responsible for the operation and maintenance of IRMS instrumentation at the University of Alberta, including C. Figueiredo, C. Nguyen, and J. Khatkar. A. Harms, currently the manager of the department’s Natural Resources Analytical Laboratory, has been with the department since 1997 and continues to operate and maintain these systems today.

**Challenges and Opportunities**

Major advances in instrumentation, especially with regard to the continuous-flow isotope ratio mass spectrometers (CF-IRMS) interfaced to a range of peripherals have allowed high-precision measurements of...
increasingly small sample sizes (Crotty et al. 2013). Coupling of CF-IRMS to gas and liquid chromatographs in the mid-80s opened the door to the isotopic analysis of individual molecules (i.e., CSIA). Analyses of some metabolites that are well suited to gas chromatography, such as $\delta^{13}$C values of soil microbial phospholipids, are now mostly routine in isotope laboratories. Analytical precision while measuring natural isotopic abundance in individual PLFAs has improved to the point where it is possible to differentiate between carbon sources with little isotopic separation. Such was the case for PLFAs extracted from rhizospheric and bulk soil samples (Thacker 2018). Other emerging applications include $\delta^{15}$N analysis in individual amino acids (Ohkouchi et al. 2017) and combined analysis of multiple isotopes, such as $\delta^{13}$C and $\delta^{2}$H measurements in soil n-alkanes (Paul and Quideau 2020). CSIA is particularly well suited to soil metabolomics, and it is a powerful tool to trace natural metabolites and pollutants alike. Newer technology is now in place to follow molecules that are best analyzed with liquid chromatography, such as soil carbohydrates and amino acids. However, while this analytical approach possesses tremendous potential, applications to date have been limited, mostly due to the cost and the rarity of CSIA instrumentation dedicated to environmental research.

The development of non-mass spectrometry approaches (i.e., optical spectroscopic techniques) has literally brought isotopic analyses to the field. Compared with traditional IRMS analyses, spectroscopic instruments present some advantages, including lower cost, ease of use, and maybe more importantly portability. These infrared (IR) absorption laser spectrometers utilize either the more sensitive (but less robust) mid-IR technology or the most commonly used, cheaper near-IR technology. Combined with cavity ring down technology, near-IR absorption spectroscopy lasers are particularly well adapted to the measurements of greenhouse gases under various field conditions, including remote locations.

Technical advances in the area of isotope geochemistry have been nothing short of phenomenal since the creation of the first isotope mass spectrometers in the first half of the 20th century. The first published work on carbon stable isotopes, using what was considered at the time to be a “mass spectrometer of high sensitivity and high resolving power” quoted a precision of 0.5% for the $^{12}$C/$^{13}$C ratio (Nier and Gulbransen 1939). By contrast, modern IRMS systems can easily reach $\delta^{13}$C/$^{12}$C precisions of ±0.2‰, and many systems routinely achieve precisions much better than that. Today, isotopes of light elements (C, H, N, O, and S) provide environmental science researchers the means to trace the origins and track the fates of individual metabolites, nutrients, and chemical pollutants as they move through soils, plants, food webs, soil and surface waters, and the atmosphere.

Acknowledgements

The authors thank J.A. Robertson for his valuable input and assistance in finding early department records, as well as University of Alberta librarian J. Thorlakson for her help in conducting database searches. We also thank all current Renewable Resources Department faculty members who contributed information about their ongoing soil isotope research. This article would not have been possible without the dedicated efforts of all past and present members of the department involved in soil isotope research.

References

Ahad, J.M.E., Pakdel, H., Gammon, P.R., Siddique, T., Kuznetsova, A., and Savard, M.M. 2018. Evaluating in situ biodgradation of $^{15}$N-labelled naphthenic acids in groundwater near oil sands tailings ponds. Sci. Total Environ. 643: 392–399. doi:10.1016/j.scitotenv.2018.06.159. PMID:29940450.

Arevalo, C.B.M., Bhatti, J.S., Chang, S.X., and Sidders, D., 2010. Distribution of recent photosynthates in saplings of two hybrid poplar clones. Commun. Soil Sci. Plant Anal. 41: 1004–1015. doi:10.1080/00103620103646089.

Baggs, E.M. 2008. A review of stable isotope techniques for $^{15}$N source partitioning in soils: recent progress, remaining challenges and future considerations. Rapid Commun. Mass Spectrom. 22: 1664–1672. doi:10.1002/rcm.3456. PMID:18435506.

Béasse, M. 2012. Microbial communities in organic substrates used for oil sands reclamation and their link to boreal seedling growth. M.Sc. thesis, Department of Renewable Resources, University of Alberta, Edmonton, AB, Canada. 96 pp.

Bentley, C.F., Hoff, D.J., and Scott, D.B. 1955. Fertilizer studies with radioactive sulphur II. Can. J. Agric. Sci. 35: 264–281. doi:10.4141/agsci-1955-0036.

Binkley, D., Sollins, P., and McGill, W.B. 1985. Natural abundance of nitrogen-15 as a tool for tracing alder-fixed nitrogen. Soil Sci. Soc. Am. J. 49: 444–447. doi:10.2136/sssaj1985.03615995004900020034x.

Brüegemann, N., Gessler, A., Kayler, Z., Keel, S.G., Badeck, F., Barthel, M., et al. 2011. Carbon allocation and carbon isotope fluxes in the plant-soil-atmosphere continuum: a review. Biogeosciences, 8: 3457–3489. doi:10.5194/bg-8-3457-2011.

Burd, K., Tank, S.E., Dion, N., Quinton, W.L., Spence, C., Tanentzap, A.J., and Olefeldt, D. 2018. Seasonal shifts in export of DOC and nutrients from burned and unburned peatland-rich catchments, Northwest Territories, Canada. Hydrol. Earth Syst. Sci. 22: 4455–4472. doi:10.5194/hess-22-4455-2018.

Card, S.M., Quideau, S.A., and Oh, S.W. 2010. Carbon characteristics in restored and reference riparian soils. Soil Sci. Soc. Am. J. 74: 1834–1843. doi:10.2136/sssaj2009.0466.

Chabbi, A., and Rumpel, C. 2012. Preface to the Special Issue on “Challenges and limits of stable isotopes in environmental research”. Org. Geochem. 42: 1437–1439. doi:10.1016/j.orggeochem.2011.12.003.

Cheng, Y., Cai, Z.C., Zhang, J.B., Lang, M., Mary, B., and Chang, S.X. 2012. Soil moisture effects on gross nitrification difference between adjacent grassland and forested soils in central Alberta, Canada. Plant Soil. 352: 289–301. doi:10.1007/s11104-011-0997-2.

Cheng, Y., Wang, J., Chang, S.X., Cai, Z., Müller, C., and Zhang, J. 2019. Nitrogen deposition affects both net and gross soil nitrogen transformations in forest ecosystems: a review.
Cormack, D.V., Bentley, C.F., and Scott, D.B. 1951. Fertilizer stud-

Choi, W.-J., Chang, S.X., Curran, M.P., Ro, H.-M., Kamaluddin,

Estop-Aragonés, C., Czimczik, C.I., Heffernan, L., Gibson, C.,

Enloe, H.A., Quideau, S.A., Graham, R.C., Sillett, S.C., Oh, S.W.,

Drijber, R.A., and McGill, W.B. 1994a. Sulfonilipid content of

Cytophaga and Flexibacter species isolated from soil and cul-
tured under different nutrient and temperature regimes.
Can. J. Microbiol. 40: 132–139. doi:10.1139/m94-021.

Drijber, R.A., and McGill, W.B. 1994b. Sulfonilipoids as a bio-
marker to monitor the population dynamics of the genera
Cytophaga and Flexibacter in soil worked by the earthworm
Aporrectodea turgida. Soil Biol. Biochem. 26: 1395–1403.
doi:10.1016/0038-0717(94)90223-2.

Enloe, H.A., Quideau, S.A., Graham, R.C., Sillett, S.C., Oh, S.W.,
and Wasylishen, R.E. 2010. Soil organic matter processes in
old-growth redwood forest canopies. Soil Sci. Soc. Am. J. 74:
161–171. doi:10.2136/sssaj2009.0031.

Estop-Aragonés, C., Czimczik, C.I., Heffernan, L., Gibson, C.,
Walker, J.C., Xu, X., and Olefeldt, D. 2018. Respiration of aged
soil carbon during fall in permafrost peatlands enhanced by
active layer deepening following wildfire but limited following
thermokarst. Environ. Res. Lett. 13(8). doi:10.1088/1748-
9326/aad550.

Fang, H., Yu, G., Cheng, S., Zhu, T., Zheng, J., Mo, J., et al. 2011.
Nitrogen-15 signals of leaf-litter-soil continuum as a possible
indicator of ecosystem nitrogen saturation by forest succes-
sion and N loads. Biogeochemistry, 102: 251–263.
doi:10.1007/s10533-010-9438-1.

Garnier-Laplace, J., and Roussel-Debet, S. 2001. Radionuclide fact-
sheet: carbon and the environment. IRSN, Fontenay-aux-
Roses, France. [Online]. Available from https://www.irsn.fr/EN/
Research/publications-documentation/radionuclides-sheets/
environment/Documents/CarbonE_UK.pdf [17 Nov 2019].

Gaudinski, J.B., Dawson, T.E., Quideau, S., Schuur, E.A.G.,
Roden, J.S., Trumbore, S.E., et al. 2005. Comparative analysis of
cellulose preparation techniques for use with C-13,
C-14, and O-18 isotopic measurements. Anal. Chem. 77:
7212–7224. doi:10.1021/ac050548u. PMID:16285668.

Glaser, B. 2005. Compound-specific stable-isotope (delta C-13)
analysis in soil science. J. Plant Nutr. Soil Sci. 168: 633–648.
doi:10.1007/s00174-005-0517-5.

Hannam, K.D., Quideau, S.A., Kishchuk, B.E., Oh, S.W.,
and Wasylishen, R.E. 2005. Forest-floor chemical properties are
altered by clear-cutting in boreal mixedwood forest stands
dominated by trembling aspen and white spruce. Can. J.
For. Res. 35: 2457–2468. doi:10.1139/x05-104.

Haugen-Kozyra, K., Juma, N.G., and Nyborg, M. 1993. Nitrogen
partitioning and cycling in barley-soil systems under conven-
tional and zero tillage in central Alberta. Can. J. Soil Sci. 73:
183–196. doi:10.4141/css93-021.

Heaney, D.J., and Nyborg, M. 1988. Overwinter transformations
of nitrate derived from soil and 15N-labeled potassium nitrate.
Soil Sci. Soc. Am. J. 52: 667–671. doi:10.2136/sssaj
1988.03615995005200030018x.

Heaney, D.J., Nyborg, M., Solberg, E.D., Malhi, S.S., and
Ashworth, J. 1992. Overwinter nitrate loss and denitrification
potential of cultivated soils in Alberta. Soil Biol. Biochem. 24:
877–884. doi:10.1016/0038-0717(92)90009-M.

Hemsley, T.L., MacKenzie, M.D., and Quideau, S.A. 2019.
Ecophysiologically based response of aspen (Populus tremuloides)
and jack pine (Pinus banksiana) to atmospheric nitrogen deposi-
tion on reconstructed boreal forest soils in the Athabasca
oil sands region. Sci. Total Environ. 696: 133544. doi:10.1016/
j.scitotenv.2019.07.350. PMID:31465928.

Hu, Y.-L., Yan, E.-R., Choi, W.-J., Salifu, F., Tan, X., Chen, Z.C., et
al. 2014. Soil nitrification and foliar 15N declined with stand
age in trembling aspen and jack pine forests in northern
Alberta, Canada. Plant Soil. 376: 399–409. doi:10.1007/s11104-
2013-1704-4.

Hutchins, R.H.S., Tank, S.E., Olefeldt, D., Qunton, W.L., Spence,
C., Dion, N., et al. 2020. Fluvial CO2 and CH4 patterns across
wildfire disturbed ecosoues of subarctic Canada: current status
and implications for future change. Glob. Change Biol. 26(4):
2304–2319. doi: 10.1111/gcb.14960.

Ibrahim, E., Wolf, B., Harris, E., Gasche, R., Wei, J., Yu, L., et al.
2019. Attribution of N2O sources in a grassland soil with laser
spectroscopy based isotopocule analysis. Biogeosciences, 16:
3247–3266. doi:10.5194/bg-16-3247-2019.

Jans-Hammermeister, D.C., McGill, W.B., and Jensen, T.L. 1994.
Dynamics of 15N in two soil-plant systems following incorpo-
ration of 15N bloom and full bloom field pea. Can. J. Soil Sci. 74:
99–107. doi:10.4141/css94-013.

Juma, N.G., and Paul, E.A. 1983. Effect of a nitrification inhibitor
on N immobilization and release of 15N from nonexchange-
able ammonia and microbial biomass. Can. J. Soil Sci. 63:
167–175. doi:10.4141/jcss83-018.

Juma, N.G., Paul, E.A., and Mary, B. 1984. Kinetic analysis of
net nitrogen mineralization in soil. Soil Sci. Soc. Am. J. 48:
753–757. doi:10.2136/sssaj1984.03615995004800040001x.

Jung, K., Choi, W.-J., Chang, S.X., and Arshad, M.A. 2013. Soil and
ring chemistry of Pinus banksiana and Populus tremuloides
stands as indicators of changes in atmospheric environments
in the oil sands region of Alberta, Canada. Ecol. Indic. 25:
256–265. doi:10.1016/j.ecolind.2012.10.006.

Kipps, K. 2015. Laboratory-measured soil organic carbon miner-
alization in soil samples from six long-term crop rotations in
Alberta as a function of sample disturbance. M.Sc. thesis,
Department of Renewable Resources, University of Alberta,
Edmonton, AB, Canada. 125 pp.

Lang, M., Cai, Z.-C., Mary, B., Hao, X., and Chang, S.X. 2010.
Land-use type and temperature affect gross nitrogen trans-
formation rates in Chinese and Canadian soils. Plant Soil, 334:
377–389. doi:10.1007/s11104-010-0389-z.

Published by NRC Research Press
Lang, M., Li, P., Ti, C., Zhu, S., Yan, X., and Chang, S.X. 2019. Soil gross nitrogen transformations are related to land-uses in two agroforestry systems. Ecol. Eng. 127: 431–439. doi:10.1016/j.ecoleng.2018.12.022.

Lejoly, J., Quideau, S.A., and Rees, F. 2020. Microbial response to carbon and nutrient additions in boreal forest soils and cover soils used during post mining reclamation. Can. J. Soil Sci. 100(1): 69–80. doi:10.1139/CJSS-2019-0088.

Lloret, E., and Quideau, S.A. 2015. Microbial processing of leaf- and root-derived organic matter in the boreal forest. EGU General Assembly Conference Abstracts 17, 1950.

Malhi, S.S., and Nyborg, M. 1983. Field-study of the fate of 15N-labelled urea: tillage, time of application and method of placement. Proc. Annual Alberta Soil Science Workshop. pp. 70–75.

Matsushima, M.Y., Choi, W., and Chang, S.X. 2014. Canada bluejoint foliar delta N-15 and delta C-13 indicate changed soil N availability by litter removal and N fertilization in a 13-year-old boreal plantation. Soil Sci. Plant Nutr. 60: 208–215. doi:10.1080/00380717.2013.869762.

Maulé, C.P., and Dudas, M.J. 1989. Preliminary identification of soil separates associated with fallout 137Cs. Can. J. Soil Sci. 69: 171–175. doi:10.4141/cjss89-016.

McGill, W.B., and Paul, E.A. 1976. Fractionation of soil and 15N nitrogen to separate the organic and clay interactions of immobilized N. Can. J. Soil Sci. 56: 203–212. doi:10.4141/cjss76-029.

McGill, W.B., Shields, J.A., and Paul, E.A. 1975. Relation between carbon and nitrogen turnover in soil organic fractions of microbial origin. Soil Biol. Biochem. 7: 57–63. doi:10.1016/0038-0717(75)90032-2.

McMillan, R., Quideau, S.A., MacKenzie, M.D., and Biryukova, O. 2007. Nitrogen mineralization and microbial activity in oil sands reclaimed boreal forest soils. J. Environ. Qual. 36: 1470–1478. doi:10.2134/jeq2006.0530. PMID:17766826.

Michener, R., and Lajtha, K. (eds.). 2007. Stable isotopes in ecological and environmental science. 2nd ed. Blackwell Publishing, Malden, MA, USA.

Monreal, C.M., and McGill, W.B. 1989a. The dynamics of free cystine cycling at steady-state through the solutions of selected cultivated and uncultivated chernozemic and luvisolic soils. Soil Biol. Biochem. 21: 689–694. doi:10.1016/0038-0717(89)90065-5.

Monreal, C.M., and McGill, W.B. 1989b. The effects of soil amendments on the dynamics of free cystine cycling at steady-state through the solutions of a black chernozemic and an andept soil. Soil Biol. Biochem. 21: 695–701. doi:10.1016/0038-0717(89)90066-7.

Monreal, C.M., and McGill, W.B. 1989c. Kinetic analysis of cystine cycling through the solution of a gray luvisol and an andept soil. Soil Biol. Biochem. 21: 671–679. doi:10.1016/0038-0717(89)90063-1.

Monreal, C.M., and McGill, W.B. 1989d. Kinetic analysis of soil microbial components under perturbed and steady-state conditions in a gray Luvisol. Soil Biol. Biochem. 21: 681–688. doi:10.1016/0038-0717(89)90064-3.

Mugasha, A.G., and Pluth, D.J. 1994a. 15N-labelled urea fertilization of a tamarack/black spruce mixed stand on a drained minerotrophic peatland. 15N in soil and tree uptake. For. Ecol. Manage. 68: 339–351. doi:10.1016/0378-1127(94)90055-8.

Mugasha, A.G., and Pluth, D.J. 1994b. Distribution and recovery of 15N-urea in a tamarack/black spruce mixed stand on a drained minerotrophic peatland. For. Ecol. Manage. 68: 353–363. doi:10.1016/0378-1127(94)90056-6.

Nason, G.E., Pluth, D.J., Hardin, R.T., and McGill, W.B. 1990. Dynamics of foliar N in Douglas-fir after spring and fall application of ammonium nitrate and urea. Can. J. For. Res. 20: 1515–1523. doi:10.1139/x90-201.

Nier, A.O., and Gulbransen, E.A. 1939. Variations in the relative abundance of the carbon isotopes. J. Am. Chem. Soc. 61: 697–698. doi:10.1021/ja01872a047.

Nömmik, H., Pluth, D.J., Larsson, K., and Mahendrappa, M.K. 1994. Isotopic fractionation accompanying fertilizer nitrogen transformations in soil and trees of a Scots pine ecosystem. Plant Soil, 158: 169–182. doi:10.1007/BF00009442.

Norris, C.E., Quideau, S.A., Bhatti, J.S., and Wasylissen, R.E. 2011. Soil carbon stabilization in jack pine stands along the Boreal Forest Transect Case Study. Global Change Biol. 17: 480–494. doi:10.1111/j.1365-2486.2010.02236.x.

Norris, C.E., Quideau, S.A., Landhäusser, S.M., Bernard, G.M., and Wasylissen, R.E. 2012. Tracking stable isotope enrichment in tree seedlings with solid-state NMR spectroscopy. Sci. Rep. 2: 719. doi:10.1038/srep00719. PMID:23056911.

Norris, C.E., Quideau, S.A., and Oh, S.W. 2016. Microbial utilization of double-labeled aspen litter in boreal aspen and spruce soils. Soil Biol. Biochem. 100: 9–20. doi:10.1016/j.soilbio.2016.05.013.

Norris, C.E., Quideau, S.A., Landhäusser, S.M., Drozdowski, B., Hogg, K.E., and Oh, S.W. 2018. Assessing structural and functional indicators of soil nitrogen availability in reclaimed forest ecosystems using 15N-labelled aspen litter. Can. J. Soil Sci. 98: 357–368. doi:10.1139/cjss-2018-0021.

Omanwar, P.K., and Robertson, J.A. 1971. A micro and rapid method for measuring P32 in plant samples. Plant Soil, 34: 225–227. doi:10.1007/BF00372776.

Paul, A., and Quideau, S.A. 2020. Carbon and hydrogen isotopes of n-alkanes in soils reconstructed after mining disturbance. J. Environ. Qual. doi:10.1002/eq.20069 (in press).

Pawluk, S., and Bentley, C.F. 1964. The uptake of S35 by crops from variable depths in the soils. Can. J. Soil Sci. 44: 261–263. doi:10.1016/0008-7188(64)90018-x.
Scheu, S., and Falca, M. 2000. The soil food web of two beech forests (*Fagus sylvatica*) of contrasting humus type: stable isotope analysis of a macro- and a mesofauna-dominated community. Oecologia, **123**: 285–296. doi:10.1007/s004420051015. PMID:28308733.

Sewell, P. 2018. Seeing the forest for the soil: topographic controls on soil carbon dynamics in the boreal mixedwood forest. M.Sc. thesis, Department of Renewable Resources, University of Alberta, Edmonton, AB, Canada. 122 pp.

Spiers, G.A., Dudas, M.J., Muehlenbachs, K., and Pawluk, S. 1985. Isotopic evidence for clay mineral weathering and authigenesis in Cryoboralfs. Soil Sci. Soc. Am. J. **49**: 467–474. doi:10.2136/sssaj1985.03615995004900020039x.

Tan, X., Kabzems, R., and Chang, S.X. 2006. Response of forest vegetation and foliar $\delta^{13}$C and $\delta^{15}$N to soil compaction and forest floor removal in a boreal aspen forest. For. Ecol. Manage. **222**: 450–458. doi:10.1016/j.foreco.2005.10.051.

Thacker, S. 2018. Rhizosphere microbial response to predicted vegetation shifts and changes in rhizodeposition in boreal forest soils. M.Sc. thesis, Department of Renewable Resources, University of Alberta, Edmonton, AB, Canada. 152 pp.

Turcotte, I., Quideau, S.A., and Oh, S.W. 2009. Organic matter quality in reclaimed boreal forest soils following oil sands mining. Org. Geochem. **40**: 510–519. doi:10.1016/j.orggeochem.2009.01.003.

Watzinger, A. 2015. Microbial phospholipid biomarkers and stable isotope methods help reveal soil functions. Soil Biol. Biochem. **86**: 98–107. doi:10.1016/j.soilbio.2015.03.019.

Wellman, R.P., Cook, F.D., and Krouse, H.R. 1968. Nitrogen-15: microbiological alteration of abundance. Science, **161**: 269–270. doi:10.1126/science.161.3838.269. PMID:5657330.

Xu, J.G., and Juma, N.G. 1993. Above- and below-ground transformation of photosynthetically fixed carbon by two barley (*Hordeum vulgare* L) cultivars in a typic Cryoboroll. Soil Biol. Biochem. **25**: 1263–1272. doi:10.1016/0038-0717(93)90223-X.

Xu, J.G., and Juma, N.G. 1994. Relations of shoot C, root C and root length with root-released C of two barley cultivars and the decomposition of root-released C in soil. Can. J. Soil Sci. **74**: 17–22. doi:10.4141/cjss94-002.

Xu, J.G., and Juma, N.G. 1995. Carbon kinetics in a Black Chernozem with roots in situ. Can. J. Soil Sci. **75**: 299–305. doi:10.4141/cjss95-043.

Zaccone, C., Plaza, C., Ciavatta, C., Miano, T.M., and Shotyk, W. 2018. Advances in the determination of humification degree in peat since Achard (1786): applications in geochemical and paleoenvironmental studies. Earth Sci. Rev. **185**: 163–178. doi:10.1016/j.earscirev.2018.05.017.

Zamir, S., Foght, J.M., and Siddique, T. 2012. DNA-SIP to characterize microbial community involved in toluene degradation in oil sand tailings enrichment cultures. Poster presentation. International Society for Microbial Ecology (ISME) 14, Copenhagen, Denmark, 19–24 Aug. 2012.

Zhang, M., Nyborg, M., Malhi, S.S., and Solberg, E.D. 2000. Localized root growth in soil induced by controlled-release urea granule and barley nitrogen uptake. J. Plant Nutr. **23**: 413–422. doi:10.1080/0190416009382027.

Published by NRC Research Press