

Carbon and oxygen stable isotopes from tree-rings to identify spruce budworm outbreaks in the boreal forest of Québec

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Abstract

The aim of this study was to test the potential of carbon ($\delta^{13}\text{C}$) and oxygen ($\delta^{18}\text{O}$) isotopes as indicators of spruce budworm outbreaks. It was hypothesized that defoliation induced by insects would trigger a ^{13}C enrichment of *Abies balsamea* and *Picea mariana* tree-ring α -cellulose through higher photosynthetic compensatory rate, while $\delta^{18}\text{O}$ would remain constant. The hypothesis was based on observations of increased photosynthetic rate induced by defoliation, as a compensatory mechanism (Little, C.H.A., Lavigne, M.B., Ostaff, D.P., 2003. Impact of old foliage removal, simulating defoliation by the balsam fir sawfly, on balsam fir tree growth and photosynthesis of current-year shoots. For. Ecol. Manag. 186, 261–269), [Lavigne, M.B., Little, C.H.A., Major, J.E., 2001. Increasing the sink:source balance enhances photosynthetic rate of 1-year-old balsam fir foliage by increasing allocation of mineral nutrients. Tree Physiol. 21, 417–426]. Comparison of the two host species, *A. balsamea* and *P. mariana* with a non-host one, *Pinus banksiana*, revealed carbon isotope enrichments during both the 1950s and 1970s spruce budworm outbreaks which did not occur in the non-host species. Carbon and oxygen isotope values showed high synchronicity not only within species but also between species (*A. balsamea* and *P. mariana*) and sites. *P. banksiana* $\delta^{18}\text{O}$ values were also highly synchronous with those of the two other coniferous species. The comparison of host and non-host ring width, $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$ chronologies confirmed the potential of combining these isotope indicators of spruce budworm outbreaks.

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1. Introduction

Spruce budworm (*Choristoneura fumiferana* Clem.) outbreaks are one of the main disturbances of North Eastern boreal forests of North America (Maclean, 1984; Morin 1994). Balsam fir [*Abies balsamea* (L.) Mill.] remains the favourite host species for spruce budworms in the North Eastern boreal forests (Martineau, 1985). However, different coniferous species such as white spruce [*Picea glauca* (Moench) Voss], black spruce [*Picea mariana* (Mill.) B.S.P] or larch [*Larix laricina* (DuRoi) K. Koch] can also be affected by this insect.

Extended records of spruce budworm outbreaks have been assembled through peatland sediment core macrofossil reconstructions (Simard et al., 2002, 2006). While these records provide useful information about past spruce budworm infesta-

tions, their coarse temporal resolution (often greater than 100 years) inaccurately portrays the decadal scale frequency (32 to 34 year intervals) of spruce budworm outbreaks (Royama, 1984). A more accurate but shorter duration chronology (300 to 400 years) of spruce budworm infestations in the North Eastern boreal forest has been reconstructed from living, historical and sub-fossil wood (Blais, 1983; Krause, 1997; Boulanger and Arseneault, 2004). This accurate identification of past insect outbreaks from tree-rings was accomplished through detailed comparisons between host and non-host tree species growth curves (Blais, 1962; Swetnam et al., 1985; Swetnam, 1987). As spruce budworms feed on the foliage they effectively reduce the leaf surface area and photosynthetic capacity of the host trees by varying degrees (heavy, moderate, light) (Kozlowski, 1969). During heavy defoliation, growth loss observed in ring width series exhibits a typical “V” or “U” shape lasting for 5 years or more (Swetnam et al., 1985; Jardon et al., 1994). However, it is highly improbable to find non-host sub-fossil trees in the

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peatland repositories of the North Eastern Boreal forest to compare with host tree records and therefore difficult to further extend the records of spruce budworm infestations beyond the past several hundred years (Simard, 2003). An additional problem arises when attempting to reconstruct past light to moderate spruce budworm infestations, that is, while infestations resulting in only light to moderate defoliations can also induce tree growth loss, the indicative “V” or “U” shaped patterns are usually not observed.

Attempts have been made to confirm other indicators of insect infestations using wood anatomy (Harper, 1913; Bailey, 1925; Fillion and Courmoyer, 1995; Krause and Morin, 1995; Liang et al., 1997) and tree growth anomalies (Simard and Payette, 2003). None of these methods provide exclusive evidence of defoliation allowing for assured reconstructions of spruce budworm outbreaks on more extensive temporal scales. The difficulties in identifying and establishing precise onsets of past insect outbreaks using traditional dendrochronological methods has prompted this investigation to determine whether carbon isotopic compositions of host tree annual tree-rings unequivocally provide spruce budworm outbreak “signatures” during two known 20th Century infestation outbreaks (1950s and 1970s). Here, the carbon isotopic compositions are compared to congruent annual ring width and oxygen isotope values in both host and non-host tree species to confirm their utility in tracing spruce budworm outbreaks.

2. Background

From a physiological point of view, various responses may be observed in plants affected by leaf-feeding insects. Lavigne et al. (2001) and Little et al. (2003) studied the effect of defoliation on physiological parameters of *A. balsamea* and showed that the photosynthetic rate of the remaining needles increased, while stomatal activity remained constant. Increase of photosynthetic rate per unit area of residual and/or regrowth foliage as a compensatory mechanism has been observed in other coniferous species as well (Welter, 1989; Reich et al., 1993; Vanderklein and Reich, 1999; Chen et al., 2001). Decreases or no change of photosynthetic rate have also been observed (Welter, 1989).

Discriminations against the heavier carbon isotope (^{13}C) occur during stomatal conductance (g_s) of CO_2 into leaves and during subsequent photosynthetic assimilation (A) of carbon (O’Leary, 1981; Farquhar et al., 1982). As discussed previously, the physiological responses of conifers to insect related defoliation in plants would significantly impact the rate of photosynthetic assimilation (A) and, therefore, carbon isotope analysis of host tree-rings represents a potentially promising method of detecting spruce budworm outbreaks. It is hypothesized that the defoliation induced by leaf-feeding insects would trigger a ^{13}C enrichment of *A. balsamea* tree-ring α -cellulose through a higher compensatory photosynthetic rate. Rather than reflecting changes in photosynthetic capacity, the oxygen isotopic composition ($\delta^{18}\text{O}$) of the tree-ring cellulose is expected to reflect the isotopic signature of source water (Roden and Ehleringer, 1999a,b; Roden et al., 2000), the

isotopic signature of leaf water, consequent of evaporation and the Péclet effect, the biochemical fractionation during biosynthesis of photosynthetic sugars (Sternberg, 1989; Yakir and DeNiro, 1990) and the reequilibrium exchange between the carbohydrate and xylem water during tree-ring xylem cellulose synthesis (Sternberg et al., 1986; Yakir and DeNiro, 1990; Luo and Sternberg, 1992).

Studies on the impact of phytophagous insects on carbon isotope composition of plant materials are very limited (Leavitt and Long, 1986; Ellsworth et al., 1994). Ellsworth et al. (1994) observed that feeding damage induced by piercing–sucking insects on deciduous trees did not provoke compensatory gas exchange, rather a reduction of photosynthesis. In addition, damage induced by feeding insects had no impact on water-use efficiency, WUE (the ratio between photosynthesis and transpiration), or the carbon isotopic composition ($\delta^{13}\text{C}$) of the remaining leaves. However, Leavitt and Long (1986) observed enriched $\delta^{13}\text{C}$ values in *Abies concolor* and *Pseudotsuga menziesii* tree-rings, two host species, during the maximum infestation period of a western spruce budworm outbreak. *Pinus ponderosa*, a non-host species, did not record carbon isotope enrichments.

The objective here was to test the potential of annually resolved tree-ring carbon isotopes as indicators of spruce budworm outbreaks in the North Eastern boreal forest. To verify the effects of defoliation intensity and host species exclusivity on tree-ring carbon isotope compositions, matching carbon isotope analyses were performed on a secondary spruce budworm host species *P. mariana*. Annual tree-ring widths and isotopic analyses (carbon and oxygen) on a control non-host species, *Pinus banksiana* (Lamb.), helped clarify the origin of the carbon and oxygen isotopic variations.

3. Methods

3.1. Study area and samples

Five different sites were selected for sampling, three for *A. balsamea*, one for *P. mariana* and one for *P. banksiana*. All sites were located within a 20 km radius, about 100 km north of Chicoutimi (48° 25’N, 71° 4’W), Québec, Canada (Fig. 1; Table 1). They belong to the east balsam fir — white birch domain of the continuous boreal forest (Saucier et al., 1998). Regional climatic conditions for 1942–1990 (Bagotville meteorological station, 48° 20’N, 71° 0’W, 159 m asl) are characterized by a mean temperature of 2.2 °C and a mean annual precipitation of 930 mm, 37% of which fell as snow during this period (Environnement Canada, 1993). Main disturbances in this environment consist of insect outbreaks and fires.

Sampled *A. balsamea* and *P. mariana* were part of *A. balsamea*–*P. mariana* mixed mature stands located on slopes surrounding depressions forming peatlands. The minimal mean age of *A. balsamea* sample stands was 75 years (age at breast height). For the *P. mariana* samples stand, the minimal age was 140 years (age at base height) with a few trees reaching 250 years old (Table 1). All *A. balsamea*–*P. mariana* stands were closed canopies with moss and *Sphagnum* sp. ground

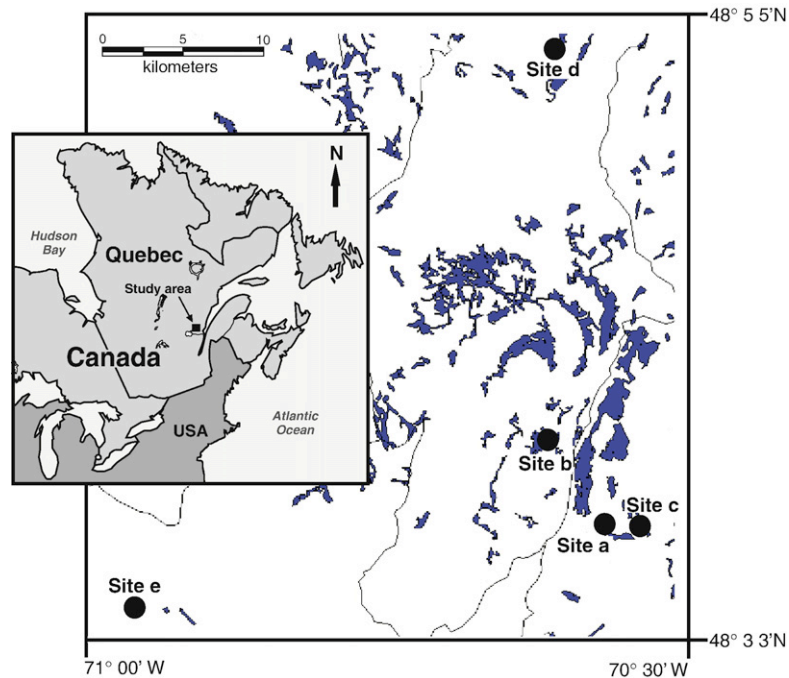


Fig. 1. Locations of the five sample sites.

coverings. *P. banksiana* trees were sampled in a pure *P. banksiana* stand with a mean tree age of 71 years (age at breast height). The landscape was generally flat (negligible slope) and ground vegetation was primarily composed of ericaceous shrubs (*Kalmia angustifolia* L. and *Ledum groenlandicum* Retzius) growing on a sandy soil.

Ten dominant trees per site were selected for carbon and oxygen stable isotope analyses. Two cores, at 180° from each other, were extracted from each tree at breast height with a 0.5 cm diameter Pressler borer. Sample surfaces were prepared with a razor blade and measured using a sliding-stage incremental micrometer (Henson, California, USA) with a precision 0.01 mm (Cook and Kairiukstis, 1990). Visual crossdating was verified using COFECHA program (Holmes, 1983). Standardisation (20 year cubic smoothing spline with a fixed 50% cutoff) was done with the ARSTAN program (Cook, 1985). The five best crossdated trees per site, i.e. showing the highest correlation and also large growth reduction during outbreak periods in the early 1950s and 1970s, were chosen for stable isotope analyses. *P. mariana* samples did not reveal a growth reduction during the 1950s outbreak, although *A. balsamea* growth curves from the same provenance showed the impact of outbreaks in the stand during this period (data not shown).

3.2. Sample preparation and stable isotope analyses

Since the two most recent outbreak periods in the studied area occurred in the 1950s and 1970s (Blais, 1983; Morin and Laprise, 1990), annual rings from the 1940–1990 period were separated from each core with a scalpel, under a binocular microscope, at the earlywood–latewood border. This period was chosen in order to obtain representative pre- and post-epidemic sections for both outbreak periods. Within each site, each same year ring (between

1940 and 1990; 51 years) from the five sampled trees were pooled together to create 51 tree-ring samples per site. Rings were ground using a steel ball mill (MM200; Retsch, Haan, Germany). Holo-cellulose was isolated by delignification in an acetic-acid-acidified sodium chlorite solution, after first removing oils and resins with toluene–ethanol and ethanol Soxhlet extractions (Leavitt and Danzer, 1993). Holo-cellulose was then converted to alpha-cellulose in sodium hydroxide. Approximately 1.0±0.1 mg of α-cellulose was packed in tin capsules for carbon combustion. Samples for carbon isotopic analysis were converted to CO₂ with an elemental analyzer (ECS 4010, Costech Analytical, Valencia, CA) and analyzed with a continuous flow isotope ratio mass spectrometer (Delta PlusXP, ThermoFinnigan, Bremen). Carbon isotopic results are reported in per mil (‰) and expressed in relation to the Vienna Peedee Belemnite (VPDB) standard. For oxygen pyrolysis, approximately 0.30±0.05 mg of α-cellulose was packed in silver cups. Samples for oxygen isotopic analysis were converted to CO with a pyrolysis elemental analyzer (TC/EA, ThermoFinnigan, Bremen) and also analyzed with a continuous flow isotope ratio mass spectrometer (Delta PlusXP,

Table 1
Locations and descriptions of the trees sampled

Site	Tree species	Location	Mean diameter (cm)	Mean age
Site a	<i>Abies balsamea</i>	48° 37' 28.44"N –70° 33' 55.65"W	23.2	69
Site b	<i>Abies balsamea</i>	48° 40' 28.98"N –70° 36' 49.74"W	17.4	75
Site c	<i>Abies balsamea</i>	48° 37' 26.65"N –70° 32' 04.41"W	17.3	80
Site d	<i>Picea mariana</i>	48° 54' 06.96"N –70° 36' 34.47"W	15.2	140
Site e	<i>Pinus banksiana</i>	48° 34' 29.96"N –70° 58' 47.64"W	14.7	71

Thermofinnigan, Bremen). Oxygen isotopic results are reported in per mil (‰) relative to the Vienna Standard Mean Ocean Water (VSMOW) standard. The isotope composition (δ) of carbon and oxygen is expressed as a deviation from the standard:

$$\delta^{xx}E = (R_{\text{sample}}/R_{\text{standard}} - 1) \times 1000$$

where E is the element of interest (carbon or oxygen), xx represents the heaviest isotope in the element (^{13}C or ^{18}O) and R is the isotope ratio ($^{13}\text{C}/^{12}\text{C}$ or $^{18}\text{O}/^{16}\text{O}$).

The precision and accuracy of mass spectrometric analyses were 0.2‰ (Acetanilide) and 0.03‰ (Acetanilide) for carbon and 0.2‰ (IAEA-602) and 0.005‰ (Hekatek Benzoic acid) for oxygen, respectively. Carbon analyses were carried out at the Idaho Stable Isotope Laboratory (University of Idaho, ID, USA) and oxygen analyses at the Stable Isotope Core Laboratory (Washington State University, WA, USA).

4. Results

4.1. Ring width analysis

The tree-ring time series cover two different known spruce budworm outbreaks (1950s and 1970s; Morin, 1994). Fig. 2 reveals markedly lower *A. balsamea* ring width index values between 1949–1954 (1955 for site a) and 1975–1980, (the years 1951–52 and 1978 represent the minimum ring widths). Full recovery (to average ring width index values: 1.04) occurred between 1954 and 1955, and in 1980 for the most recent outbreak period. The ring index for *P. mariana* suggests that this host species was not affected by the 1950s spruce budworm outbreak (Fig. 2). However, during the 1970s outbreak, the *P. mariana* ring width index reduced on cue with the host *A. balsamea* ring width index although its recovery to normal growth required two additional years (Fig. 2).

Only the most recent outbreak period was covered by *P. banksiana* data. A light radial growth decrease is observed at the beginning of the mid 1970s epidemic period although this decrease does not appear to be anomalous (Fig. 2).

4.2. Carbon and oxygen isotopes

Year to year variation of tree-ring carbon and oxygen isotope values shows a high degree of synchronicity both within and between species (Fig. 3b, c). Carbon isotope enrichments commenced for both tree species in 1947–48 and then again in 1976. Maximum enrichment values in 1950–51 and 1977 were followed by depletion minimum values in 1954 and 1980, respectively (Fig. 3b). A comparison between Fig. 3a and b shows that the most enriched carbon isotope values for *A. balsamea* (–22.4 in 1950–51 and –21.4 in 1977) occurred 1 year before the maximum growth reduction occurred (1951–52 and 1978). For *P. mariana* the 1951 carbon isotope enrichment peak did not coincide with any growth reduction whereas the 1977 enrichment peak occurred 1 year earlier than maximum ring growth reduction (Fig. 3a, b). Carbon isotope depletion troughs directly coincide with complete growth recovery rings for *A. balsamea* (1954 and 1980) but not for *P. Mariana* (Fig. 3a, b). During the 1950s outbreak, the carbon isotope enrichments seemed to precede ring width index decreases by 1 (sites a and d) to 2 (sites b and c) years. However, carbon isotope enrichments commence one year before ring width index reductions, for all sites, during the 1970s spruce budworm outbreak. Additionally, ring width index increases coincide with carbon isotope enrichments, for both species and both outbreak periods, prior to growth reductions. The carbon isotopic composition of *P. banksiana* did not vary during the mid 1970s defoliation period in comparison with the distinct carbon isotope enrichments observed for *A. balsamea* and *P. Mariana* (Fig. 3b).

The oxygen isotope time series for *A. balsamea*, *P. mariana* and *P. banksiana* show regular amplitude variations that are quite consistent through time, revealing no particular trends (Fig. 3c). During both outbreak periods (1950s and the 1970s), but particularly the 1970s period, an oxygen isotope enrichment was observed that is coincident with the carbon isotope enrichment (Fig. 3b, c). For the mid 1970s period, the most enriched oxygen isotope value occurred in 1976 followed by an immediate and sharp depletion in 1977 (Fig. 3c).

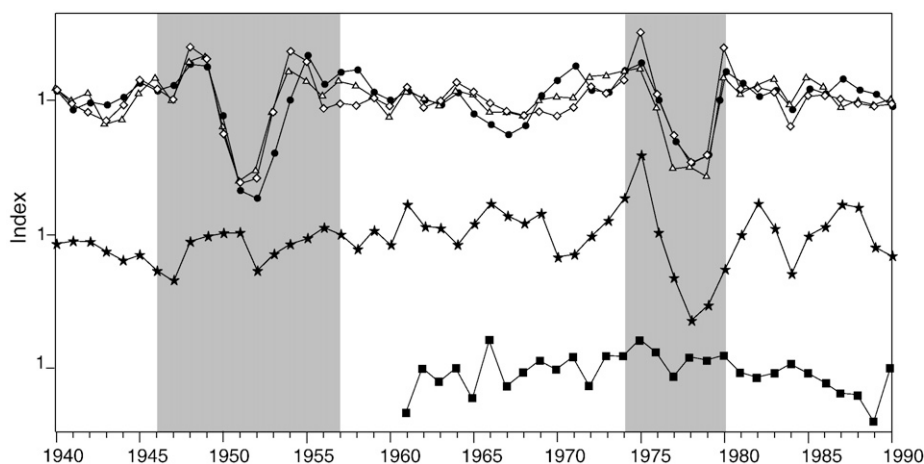


Fig. 2. Tree-ring indices of *Abies balsamea* (● site a, △ site b, ◇ site c), *Picea mariana* (★ site d) and *Pinus banksiana* (■ site e). Shaded areas indicate aerial survey observations of defoliation caused by the spruce budworm in the region (Hardy et al., 1985) covering the period 1930–80.

5. Discussion

The carbon isotopic compositions of both *A. balsamea* and *P. mariana* enrich in response to defoliation during the 1950s and 1970s spruce budworm outbreaks (Fig. 3b). Similarly, Leavitt and Long (1986) recorded carbon isotopes enrichments in host species affected by western spruce budworm outbreaks. While hypothetical at present, the mechanisms accounting for carbon isotope enrichments coincident with these budworm defoliation periods must be related to one or all of the well known effects on carbon isotope compositions in plants. In general, carbon isotope enrichments in plants can be the result of reduced stomatal conductance, increased photosynthetic capacity or a combined change in both (Francey and Farquhar, 1982). Previous studies have shown that the different levels of defoliation on *A. balsamea* (Piene, 1980; Lavigne et al., 2001; Little et al., 2003), and also on other coniferous species (Reich et al., 1993; Vanderklein and Reich, 1999; Chen et al., 2001), induced an increase in photosynthetic rates. An increase in leaf conductance was also observed by Reich et al. (1993). Lavigne et al. (2001) suggested that the increased photosynthetic rate observed in *A. balsamea* resulted from an increased allocation of mineral nutrients to 1-year old foliage enhancing their amount and/or activity, as well as increasing chlorophyll concentration.

Supporting the hypothesis of increased photosynthetic rate in the early stages of defoliation, carbon isotope enrichments coincide with an increase in ring width for *A. balsamea* at the beginning of the 1950 and 1970s outbreaks, as well as for *P.*

mariana at the onset of the latest outbreak (Fig. 3a, b). Therefore, despite the loss of foliar biomass, photosynthetic compensation may have surpassed the initial rate of carbohydrate production in these cases. Reich et al. (1993) also observed overcompensation in terms of whole plant growth due to shifts in allocation and enhanced photosynthesis. Fig. 3a and b shows carbon isotope enrichments occurring for a few years more after tree-ring widths started to decrease. For trees to survive heavy defoliation periods, it is necessary for them to make use of their starch reserves (Kozłowski et al., 1991). Starch reserve carbon isotopic compositions are relatively more enriched than tree-ring cellulose (Brugnoli et al., 1988; Le Roux et al., 2001; Helle and Schleser, 2004) and, the use of these reserves during periods of heavy defoliation may partially explain the enriched carbon isotope values coinciding with the two spruce budworm outbreak periods. Carbon isotope depletions coincided with ring width increases at the end of both outbreaks indicate a return to non-compensatory conditions (Fig. 3a, b).

During the 1950s outbreak, the ring width measurements for *P. mariana* indicate that they were not or only slightly affected by the spruce budworm defoliation. Nealis and Regnière (2004) showed that *P. mariana*, although vulnerable to spruce budworm attacks during infestation, is generally less defoliated than other host species, such as *A. balsamea*. This was explained by a temporary reduction in susceptibility of the tree species resulting from a lack of synchronism of spring larval emergence and bud burst. This mechanism was particularly important during the less severe 1950s outbreak

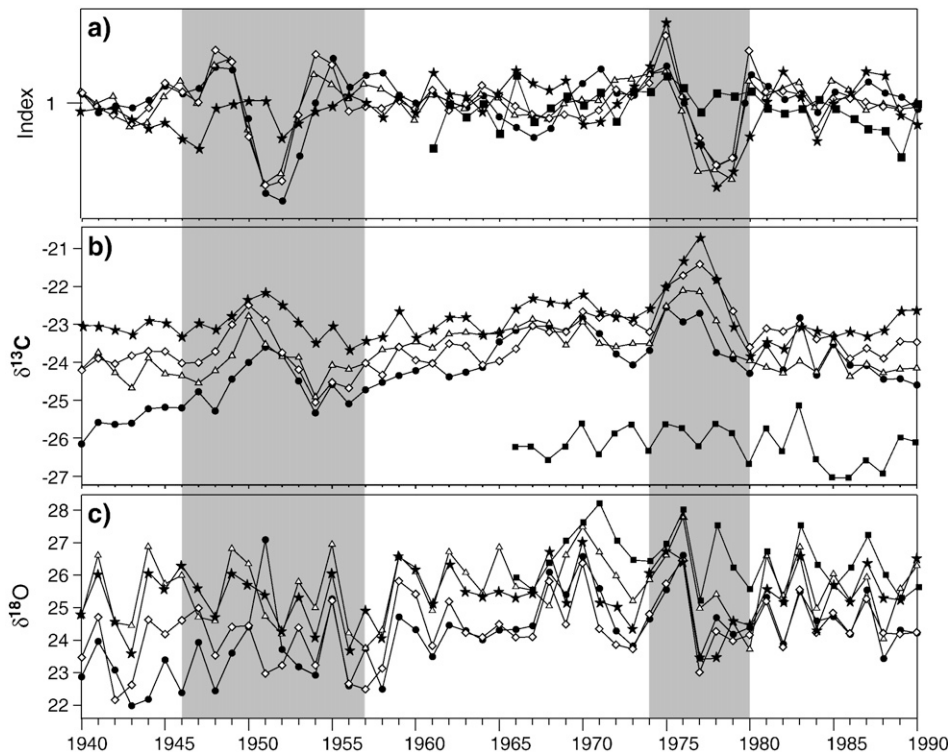


Fig. 3. *Abies balsamea* (● site a, Δ site b, ◇ site c), *Picea mariana* (★ site d) and *Pinus banksiana* (■ site e) a) tree-ring indices, b) α -cellulose carbon isotope composition and c) α -cellulose oxygen isotope composition. Shaded areas indicate aerial survey observations of defoliation caused by the spruce budworm in the region (Hardy et al., 1985) covering the period 1930–80.

(Jardon, 2001; Bouchard et al., 2006). In spite of the absence of radial growth reduction signs, the carbon isotope values reacted (enriched) in the same way as in *A. balsamea* (Fig. 3a). Two explanations for the carbon isotope enrichment in *P. mariana* during the 1950s outbreak are suggested. First, photosynthetic compensation to defoliation (as discussed previously) may be partially responsible for the carbon isotope enrichment. The second hypothesis is related to the tree stand reaction. The 1950s outbreak was not as severe as the 1970s one and the budworm population, in this mixed *P. mariana*–*A. balsamea* stand, could feed mainly on *A. balsamea* (growth reduction similar to *A. balsamea* from sites a, b and c; data not shown), potentially leaving *P. mariana* less affected. A defoliated canopy could have increased light availability. Increased light reception coupled with foliar nutrient concentration has been known to increase the WUE and enrich carbon isotope values in tree-rings (Leavitt and Long, 1991; Guehl et al., 1995; Yakir and Israeli, 1995). Canopy opening and increased radiation might also have contributed to the carbon isotope enrichment of *A. balsamea*.

Simultaneous analyses of tree-ring cellulose carbon and oxygen isotopes may help discriminate whether changes observed in the carbon isotope values originated from a modification of photosynthesis or stomatal conductance since the oxygen isotope composition of the congruent tree-rings is not expected to reflect changes in photosynthetic capacity (Craig and Gordon, 1965; Flanagan et al., 1991). Barbour et al. (2002) indicate that the response of tree-ring oxygen isotopes to changes in coniferous tree leaf conductance of water vapour (g_s) is expected to be rather small compared to broad-leaf trees. Furthermore, Scheidegger et al. (2000) report a negative relationship between oxygen isotope values and stomatal conductance. Roden and Ehleringer (1999a,b) and Roden et al. (2000) demonstrated that the source water oxygen isotopic composition has influenced tree-ring cellulose oxygen compositions. Spruce budworm outbreaks preceded by dryer early summers have previously been observed (Wellington et al., 1950; Pilon and Blais, 1961). A change in the precipitation regime and warmer temperature might account for an enriched source water oxygen isotopic composition and also explain the slight negative growth index observed in *P. banksiana* during the 1970s outbreak (Fig. 3a, c). Dryer conditions (low relative humidity) and consequent higher vapour pressure deficits (VPD) might also have contributed to an enriched oxygen isotopic composition at the leaf water level (Roden and Ehleringer, 1999a,b; Roden et al., 2000; Barbour et al., 2002). Without additional details on the environment at the time of defoliation, it is difficult to give confident details on the origin of the isotope composition variations observed. A comparison between the rather synchronous oxygen isotope patterns of host and non-host species (Fig. 3c) suggests that these variations are most likely related to climatic changes rather than spruce budworm outbreaks.

It is known that defoliation usually starts 1 to 4 years before the first year of important growth reduction (Blais, 1958, 1962; Krause et al., 2003). Defoliation might in fact have started between 1946–1949 and 1972–1975 for the earlier and later

outbreaks, respectively. Based on the results presented here, the carbon isotope composition of balsam fir α -cellulose may be a more immediate indicator of the beginning of defoliation since carbon being incorporated into the photosynthates reflects an immediate response of the physiological processes discussed previously. According to these results, the onset for the first epidemic period would be in 1948 (sites b, c and d) and 1949 (site a). The beginning of the second period would be in 1974 (sites a and d) and 1975 (sites b and c). Those dates fall within the large scale aerial observations of defoliation over Eastern North America (Hardy et al., 1985).

6. Conclusions

In order to test the potential of carbon and oxygen isotopes as indicators of spruce budworm outbreaks, host and non-host coniferous species were studied. Carbon and oxygen isotopic compositions of annual tree-ring cellulose showed high synchronicity not only within species but also between species (*A. balsamea* and *P. mariana*) and sites. *P. banksiana* oxygen isotope values were also highly synchronous with those of the two other coniferous species suggesting regional climate variations were primarily responsible for the oxygen isotope compositions.

Comparisons between host and non-host ring width and carbon and oxygen isotope chronologies reveal their potential as indicators of spruce budworm outbreaks. The carbon isotope enrichments observed in *A. balsamea* and *P. mariana* in conjunction with the outbreak periods was not present in *P. banksiana*. However, the oxygen isotope chronologies of the three species varied in the same manner, indicating a influencing factor other than climate for enriched *A. balsamea* and *P. mariana* carbon isotope compositions. Enriched oxygen isotope compositions in direct relation to outbreak period enriched carbon isotope values is thought to reflect dryer climatic conditions prior to and during those outbreak periods. While the oxygen isotope results did not totally negate the possibility of reduced stomatal activity contributing to enriched carbon isotope values during the spruce budworm epidemic periods, comparisons of host and non-host species isotope compositions in direct relation to ring widths strongly suggest that the primary mechanism behind the carbon isotope enrichments observed in host species is defoliation.

Carbon isotope values were observed to enrich 1 (sites a and d) or 2 years (sites b and c) before ring widths decreased. Carbon isotope enrichment and ring width decrease synchronicity strongly supports our hypothesis of photosynthetic compensation induced by defoliation. Even though the 1950s outbreak was undetectable with *P. mariana* ring width measurements, probably due to light defoliation, it was evident in carbon isotope measurements. This bodes well for carbon isotope detection of even light spruce budworm defoliation periods in future studies.

At this point, it is difficult to explain with certainty the mechanisms behind carbon and oxygen isotope variations observed during outbreak periods. Further research on physiological aspect of defoliation in conjunction with isotope

measurements is required to address these important questions. Despite uncertainties regarding the mechanisms involved, tree-ring carbon and oxygen isotopes analyses enabled the reconstruction of the two last spruce budworm outbreaks in the North Eastern boreal forest.

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