

Synchrotron x-ray fluorescence and secondary ion mass spectrometry in tree ring microanalysis: applications to dendroanalysis

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Mining operations in the Sudbury district of northern Ontario have resulted in environmental damage caused by acid rain and heavy metal pollution. Remedial liming (Ca/MgCO₃) has been used in an attempt to restore the pH of the lake water and soil to natural levels. The spatial and temporal variation of metal concentrations in the annual growth rings of trees may be a useful indicator of the effects of the pollutants and the liming operation. We used synchrotron radiation-induced x-ray emission (SRIXE) and secondary ion mass spectrometry (SIMS) to examine the distribution of selected metals in the annual growth rings of red pine (*Pinus resinosa*) from an area damaged by acid rain and heavy metal pollution which had been subjected to remedial liming (Ca/MgCO₃). The results show a non-uniform distribution of metals in the stemwood, including seasonal variations and changes in the Ca/Mn ratio consistent with the increased soil pH following liming. Copyright © 2001 John Wiley & Sons, Ltd.

INTRODUCTION

Dendroanalysis is the study of the element distribution in the annual growth rings of trees.¹ In principle, dendroanalysis has the potential to provide a historic record of pollution events and the kinetics of metal uptake by trees, since it might provide a chronology of changes in metal concentration. There has been much debate about the effectiveness of dendroanalysis and conflicting conclusions abound in the literature.^{2,3} Some of the differences may well depend on the analytical techniques used to determine the metal concentrations in the wood samples, since metals are often localized in or near specific physiological structures in stemwood.^{4,5}

Synchrotron radiation-induced x-ray emission (SRIXE) has proved to be useful in environmental analysis, especially when used as a fluorescence microprobe⁶ (XRM). The technique has been extended to dendroanalysis^{7,8} and has been shown to be complementary to secondary ion mass spectrometry (SIMS) in the analysis of stemwood.⁹ In this paper, we report the results of an XRM study of a sample of red pine, (*Pinus resinosa*) collected from a site that had been exposed to both airborne metals and acid rain from local smelters and which had been subsequently subjected to remedial soil liming. Annual rings representative of the period immediately following liming were interrogated with the aim of establishing the spatial distribution of selected metals in the wood

structure and to explore the probable roles of microanalytical techniques in dendroanalysis. The SIMS results were used to examine changes in the Ca/Mn ratio, which has been reported to be sensitive to soil pH.¹⁰ This ratio is expected to rise as a result of liming.

EXPERIMENTAL

Characteristics of study region

Daisy Lake^{11–13} is one of the most polluted lakes in the Sudbury region (Ontario, Canada) since it has been impacted by nickel smelting operations since early in the twentieth century. Studies have reported a lake water pH as low as 4.7 with a maximum in the sediment nickel concentration around 1965. INCO Canada has initiated remedial environmental action that has included aerial distribution of dolomite to raise the soil pH. These liming operations began in 1993.

Samples

The stem wood used in this study was obtained from red pine (*Pinus resinosa*) at chest height (~1.5 m) using a HAGLOF A 558 increment corer supplied by Canadian Forestry Equipment. Cylindrical wood cores 4 mm in diameter and ~10 cm in length were extracted and stored in plastic drinking straws prior to analysis. The specimen from the limed site selected for SRIXE was 15 years old and represented one of the very few specimens at the site. The core was sanded with 400 grit SiO₂ sandpaper to obtain a flat surface and the annual growth rings corresponding to the years 1994, 1995 and 1996 were identified using a low-power light microscope prior to SRIXE analysis using the X26A beamline at the Brookhaven National Laboratory

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Synchrotron Light Source. The years selected represented the period immediately following liming, were remote from the heartwood and did not include the most recent rings.

The same sample as used for SRIXE was used for SIMS; however, since the SIMS analysis is relatively rapid (~8 h for SRIXE vs 0.5–1 h per SIMS scan) and instrument time is more readily available, additional samples from a control site were also analyzed using the latter technique. The control site was about 200 m from the limed site. The trees at this site were 76 years old and were downslope from the limed site in a depression in the rock and had presumably survived because they were sheltered from direct contact with acid aerosols.

Apparatus

The synchrotron measurements were carried out at the Brookhaven National Synchrotron Light Source (information describing the facility can be found at <http://www.nsls.bnl.gov>). The x-ray storage ring was operated at 2.5 GeV with a 200 mA injection current. Bending magnet radiation at beamline X26A was collimated to produce a beam size of $\sim 10 \times 15 \mu\text{m}$ on the sample. A diagram of the x-ray microprobe apparatus is shown in Fig. 1. This apparatus may be used with or without the monochromator; in this work, a white beam was used in order to minimize the time required for data acquisition. The sample was scanned in 500 μm steps in the x direction and 50 μm steps in the y direction to cover a total area of $\sim 2.5 \times 12 \text{ mm}$. The x-ray fluorescence intensities for Ca, Mn, Fe, Ni, Zn, Cr and Cu were measured using an Si(Li) x-ray detector at each point in the scan. The escape depths for the x-ray emission from these elements is sufficient to ensure that surface contamination (if present) makes a negligible contribution to the fluorescence yield. The sample thickness was greater than the escape depths for the x-rays from the elements of interest.

SIMS analyses were carried out in the step-scanning mode using the Cameca IMS/3f instrument at Surface Science Western (University of Western Ontario). The samples were mounted in metal-free epoxy resin and gold coated for 5 min using a Hummer VI sputter coater to reduce surface charging during analysis. A 100 nA O^- primary ion beam

with a kinetic energy of 17 keV/ion was used to pre-sputter a $250 \times 250 \mu\text{m}$ area for 50 s prior to analysis, effectively removing any surface contamination. The same raster area was used during analysis while positive secondary ions were collected from a central spot 150 μm in diameter. A 4500 V extraction voltage was used with a 150 μm step size to move the primary ion beam across the annual growth rings. Additional analytical parameters varied for the individual elements as follows: ^{12}C and ^{16}O secondary ions were counted for 1 s with no offset voltage on the sample holder, a 100 V offset was used for the remaining elements with count times of 5 s for ^{40}Ca and 20 s for ^{55}Mn . The 100 V offset effectively suppresses molecular secondary ions.

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RESULTS AND DISCUSSION

The distributions of the x-ray fluorescence intensities obtained from Ca, Mn, Zn and Ni are displayed in Fig. 2. No effort was made to quantify the results in terms of element concentrations both because of the difficulties inherent in each analytical technique and the focus on relative element distributions rather than concentration.

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Ca, bound to functional groups in the cell walls, helps to make the cells rigid and serves as a major structural element in trees and thus has the highest concentration of all metals present in stemwood. The location of the annual rings shown in Fig. 2 as observed by a low-power optical microscope is reflected in the Ca distribution. This suggests a variable uptake throughout any growing season. Seasonal variation in metal content within individual rings has often been reported in the literature.^{3,6}

The Mn distribution closely matches that of Ca. This suggests that Mn serves the same structural function as Ca and competes with it for cation-exchange sites in the cell walls. Ca and Mn, although not displaying a completely uniform distribution, were the most evenly distributed of the metals examined in this study. Zn and Ni show apparent seasonal variations in uptake similar to those obtained for Ca and Mn.

Figure 3 repeats the distribution of Ca for reference in addition to those obtained for Fe, Cu and Cr. The latter show the least regular distributions of the metals in this

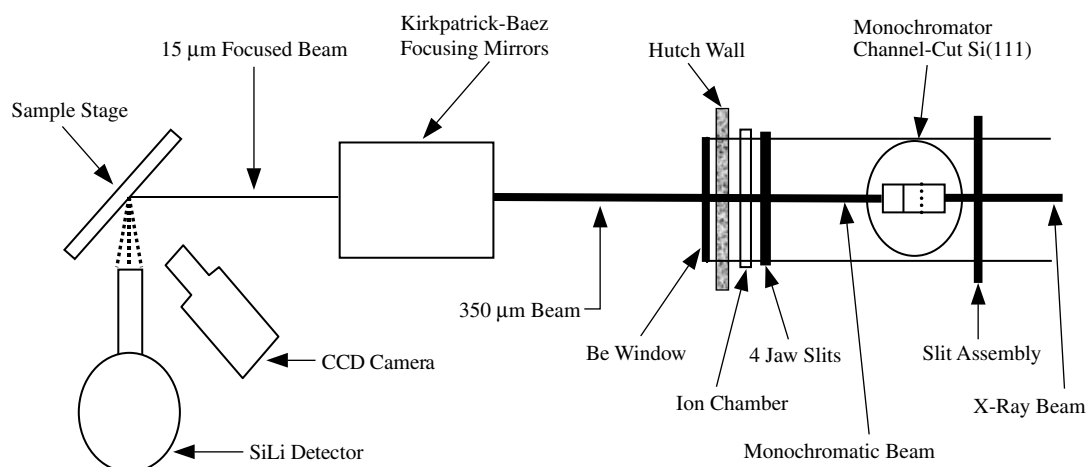


Figure 1. Schematic diagram of the X26A microprobe beamline.

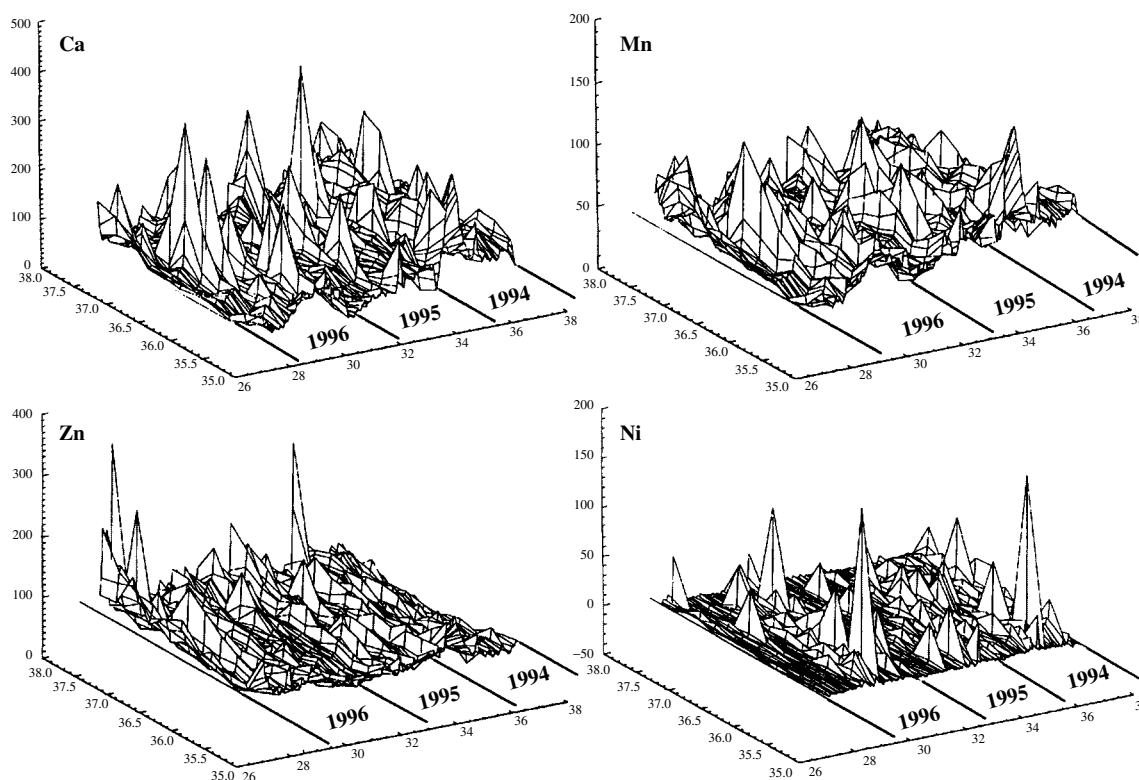


Figure 2. The distribution patterns generated by x-ray fluorescence from Ca, Mn, Zn and Ni. The x - y plane represents the wood surface, the axes measure the movement of the sample past the x-ray beam (~ 12 mm in the x direction and 2.5 mm in the y direction). The x-ray fluorescence intensities, in arbitrary units, for each element are plotted on the z axes. The boundaries of the growth rings are shown with the dates corresponding to their year of deposition.

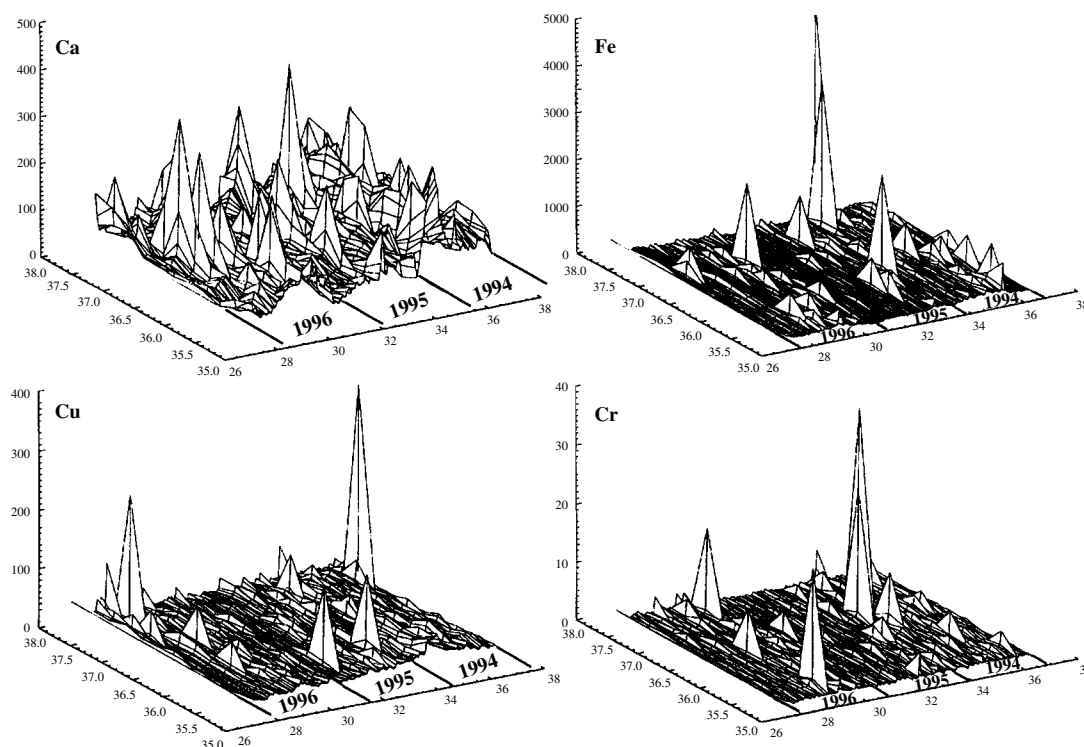


Figure 3. The distribution patterns generated by x-ray fluorescence from Ca, Cu, Fe and Cr. The x - y plane represents the wood surface, the axes measure the movement of the sample past the x-ray beam (~ 12 mm in the x direction and 2.5 mm in the y direction). The x-ray fluorescence intensities, in arbitrary units, for each element are plotted on the z axes. The boundaries of the growth rings are shown with the dates corresponding to their year of deposition.

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study. Any technique which carries out analysis on a sub-millimeter scale will be subject to large errors when used to determine the bulk concentration of metals having a similarly variable distribution. Small area analysis, especially for less abundant species, is better suited to studying the physiological activity of metals when their distributions can be correlated to identifiable wood components such as ray cells or resin ducts. Naturally, if sufficient measurements are carried out it should be possible to infer bulk concentrations. This latter criterion is likely to place unacceptably large time constraints on any analysis. Based on these data alone it might be suggested that bulk analysis could be reliably inferred from SRIXE for Ca but that similar inferences for metals having a more diffuse spatial distribution, such as for Cu, would be more problematic.

The SIMS results were used to examine changes in the Ca/Mn ratio after liming. The results in Table 1 show a statistically significant increase in the ratio following liming. This result is consistent with an immediate positive response to remedial action. There is no statistically significant difference between the control and limed sites in 1991, two years prior to liming. Although the data were collected for only a single specimen at each site, they are consistent with a rapid change in bioavailability of metals with increase in soil pH. A large number of individual points within each annual ring were interrogated to improve the reliability of the result for the single specimen.

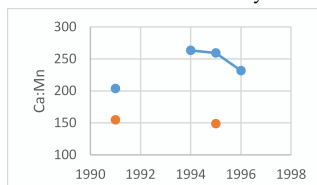
CONCLUSIONS

SRIXE analysis of selected metals in tree rings shows that inferences of bulk metal concentrations from analytical techniques that interrogate regions in the sub-millimeter range will give poor estimates of bulk concentrations unless sufficient individual analyses are completed to produce a statistically significant result. Analysis of small areas will be better suited to establishing the physiology of metals in specific wood structures. In this case, for instance, the results are consistent with cell-wall sequestration of both Ca and Mn as structural elements. Cr, Zn, Ni, Fe and Cu are confined to very small regions that appear to be

Table 1. Ca/Mn secondary ion yield ratios^a

| Ca/Mn ratio | Standard deviation | Year | Site type | No. of measurements |
|-------------|--------------------|------|------------|---------------------|
| 203.6 | 75.3 | 1991 | Limed 1993 | 50 |
| 263.5 | 61.1 | 1994 | Limed 1993 | 17 |
| 259.4 | 74.2 | 1995 | Limed 1993 | 35 |
| 231.5 | 86.1 | 1996 | Limed 1993 | 25 |
| 154.7 | 97.5 | 1991 | Control | 16 |
| 148.8 | 72.0 | 1995 | Control | 20 |

^a When ratios are compared with the value obtained before liming at the study site, only the values obtained at the limed site in 1994 and 1995 yield differences that are statistically different at the 95% confidence level.



distributed randomly on the surface. The roles of these metals in the wood structure will be better understood if these regions can be identified more closely with the elements of the wood structure, such as ray cells. It is interesting that these elements are often associated with enzyme activity. There is a strong indication of seasonal uptake for all the metals with an apparent maximum for Ca, Mn and Zn in the middle of the growing season.

The SIMS results, which represent repeated analysis with the rings and which rely on the most regularly distributed metals, show a rapid increase in the Ca/Mn ratio following liming of the soil. This observation represents direct evidence of changes in metal bioavailability with increase in pH, provides support for liming as an effective remedial action and suggests that Ca/Mn ratios may provide a surrogate measure of changes in soil pH.

In this work, SRIXE and SIMS were used as complementary techniques. SIMS has the advantage of high surface sensitivity, an ability to detect virtually all elements and is more readily available than SRIXE. However, it requires high vacuum, is more difficult to use for quantitative analysis than SRIXE and is sensitive to surface charging. The spatial resolution of SRIXE is poorer than that of SIMS when the latter is used in the imaging mode but new synchrotron facilities with better x-ray focusing are becoming available. On balance, no one technique is likely to provide all the information required for a complete analysis, especially when working with difficult samples.

Acknowledgments

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This looks good, actually, with few data points.

All the more impressive because a dendrochem

signal is apparent with so few data. *X-Ray Spectrom.* 2001; **30**: 338–341

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