Sulphur Fertilization of Lodgepole Pine: A Stable Isotope Tracer Study

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Abstract

Sulphur (S) deficiencies in lodgepole pine are widespread in the B.C. central interior, but there is an insufficient basis for prescribing fertilization treatments with both immediate and sustained growth responses. This project builds on the Ministry of Forests’ fertilization research program and careful pilot studies, to combine area-based fertilization response methods with stable isotope tracing of the uptake and fate of S fertilizers. Both elemental S and more immediately plant-available sulphate-S forms will be compared, at operationally realistic addition rates and in combination with nitrogen fertilization. Using two sites that represent typical site and climate conditions in the Sub-Boreal Spruce zone, we will compare relative uptake of these forms, their mobility and transformations in the soil, and their effects on tree growth. This research will give silviculturists a credible scientific foundation for making better fertilization decisions, and demonstrate serious intent to examine the effects of intensive forest management in B.C.

Keywords

elemental sulphur, sulphate, forest fertilization, lodgepole pine, stable isotopes, tracers, fertilizer response, nitrogen, Sub-Boreal Spruce, forest soils, nutrient cycling
1. Introduction

1.1 Sulphur and lodgepole pine nutrition

The B.C. Ministry of Forests has been actively engaged in forest nutrition and fertilization research in the B.C. interior since the early 1980’s. The earliest of these activities were designed to document the responsiveness of thinned, fire-origin lodgepole pine to N additions (Brockley 1989, 1996). The results from these early trials were variable; some stands responded well to N fertilization and others responded poorly. Through foliar analysis, S was implicated in limiting the effectiveness of N fertilization in many non-responsive stands.

Subsequent research demonstrated that growth responses of lodgepole pine may be enhanced by combining S with N in fertilizer trials, particularly in the Sub-boreal Spruce (SBS) and Sub-boreal Pine Spruce (SBPS) biogeoclimatic zones (Brockley and Sheran 1994; Brockley 1996, 2001a). Examination of S properties of unfertilized soils and foliage from many sites in the B.C. interior revealed that mineral soil S concentrations were among the lowest reported in the world literature (Kishchuk and Brockley 2002).

There is still considerable uncertainty as to which form of sulphur is most effective in stimulating growth response and in sustaining long-term improvement in site S nutrition. Soluble sulphate-S forms (e.g. ammonium sulphate) are rapidly taken up by trees, but may persist in soils and foliage for a relatively short period, and although initial availability of elemental S (S\textsuperscript{0}) is lower, this “slow release” effect may improve the efficiency of S application and may result in long-term improvement in soil S availability. In operational fertilizer prescriptions in B.C., S requirements are currently satisfied by blending urea (46% N) with ammonium sulphate (21% N, 24% S) – a choice based more on convenience and availability than on well-documented research results.

Field evidence for the relative efficacy of various forms of S is both limited and ambiguous: a 1990 fertilizer screening trial found that short-term increases in lodgepole pine needle weight were more influenced by readily available ammonium sulphate than by slowly available elemental S (Brockley, 2002). Differences in subsequent pine radial increment between S sources were less clear, although elemental S created a more lasting increase in total S concentrations in forest floors soils and foliage (Brockley, 2002).

Pre-fertilization foliar levels of N, S, and inorganic sulphate-S have proven useful for predicting whether lodgepole pine will respond significantly to fertilization with either N alone or a combination of N and S, and for estimating the magnitude of the expected responses (Brockley 2000). The availability of these tools significantly improves the likelihood of making the appropriate fertilizer prescription. However, these measures do not address the underlying cause of low S status on these sites, nor the relative effectiveness of various sources of S fertilizer in improving either tree growth or soil S availability.
1.2 The value of stable isotope methods

We are using an innovative approach, not previously employed in North American forest fertilization research, that will yield insights into S behaviour that are unavailable by other methods. Using stable isotope tracing, we will follow the fate of applied sulphur in the forest ecosystem by using carefully selected fertilizers that differ slightly from natural background in their content of the less abundant heavy isotope, $^{34}$S.

Approximately 95% of terrestrial S occurs as the stable isotope, $^{32}$S, while 4.22% occurs as $^{34}$S. If an S source, such as fertilizer or air pollution, differs even slightly from natural background levels in the relative amounts of these two isotopes, it provides a distinctive fingerprint. Differences of only a few parts per thousand can be readily detected, even in materials with low S concentrations. S stable isotope ratios are expressed as $\delta^{34}$S -- the difference in relative abundances of $^{34}$S and $^{32}$S between a sample and a standard, in parts per thousand (‰):

$$\delta^{34}\text{S} (\text{‰}) = \frac{R_{\text{sample}} - R_{\text{standard}}}{R_{\text{standard}}} \times 1000$$

where $R$ is the relative number of $^{34}$S and $^{32}$S atoms.

Mayer et al. (1995) suggested that a difference of 20 ‰ in $\delta^{34}$S values between fertilizer and background was necessary for a viable tracer experiment, but fertilizer S uptake by Norway spruce was detected when this difference was less than 10 ‰ (Rolland et al., 1991; Giesemann et al., 1995). Low soil S levels in the SBS zone, averaging 285 kg total S ha$^{-1}$ (0-50 cm mineral soil) at 6 sites studied by Kishchuk (1998), also mean that operationally realistic S fertilization rates (50 or 100 kg S ha$^{-1}$) will provide a detectable tracer effect.

If no isotopic fractionation occurs through interaction of fertilizer S with soil components and during biological uptake and transformations -- conditions generally met in aerobic soils (Krouse et al., 1996) -- fertilizer S can be followed through the soil-plant system. In the simplest case, where plant uptake occurs from two pools with different $\delta^{34}$S values (native plant-available soil S vs. fertilizer S), the proportion supplied from each source is easily calculated. In our pilot studies, we found that for Kishchuk's (1998) 6 SBS sites, $\delta^{34}$S values in unfertilized forest floors and B horizons averaged + 7.9 ‰ and + 3.3 ‰, respectively. With these background $\delta^{34}$S values and the low S concentrations in our soils, operationally realistic additions of fertilizer materials with $\delta^{34}$S values of + 15-25 ‰ would create a viable tracer experiment.

1.3 Project objectives

To address important remaining uncertainties about the behaviour of S fertilizers in central interior lodgepole pine stands, this study was initiated in 2001, in cooperation with West Fraser Timber Co. Ltd., to answer 3 questions:
• How much of the S added as fertilizer is actually taken up by the crop trees, and how much is retained in the soil (and in which forms) and or lost from the site?
• For single-dose applications, does a slow-release form of S, such as elemental S, provide more or less long-term improvement in S nutrition than a more readily-available form, such as a soluble sulphate salt?
• What is the magnitude of the tree growth responses to these treatments on an area basis?

This project's 2002-03 activities were explicitly intended to be the installation and treatment phases of a 3-year study. Our intended accomplishments for this year consisted of establishment of the experimental plots, and documentation of their pretreatment condition. To meet the stated objectives, and developing those findings into interpretive tools of value to silviculturists, will require the sampling, chemical analysis, and tree measurements planned for the remaining 2 years of this study.

2. Project Design and Methods

2.1 Site Selection

In 2001, we selected two lodgepole pine sites for our installations: (1) “Holy Cross” (HC) – km 138, Holy Cross Forest Service Road, Vanderhoof Forest District (SBSdk), and (2) “Kenneth Creek” (KC) – km 84.5, Beaver-Bowron Forest Service Road, Prince George Forest District (SBSwk1) (Figures 1-4). Our criteria included: absence of serious forest health problems, representative soil and climatic conditions, adequate stocking, age of approximately 20 years, and good all-weather access. The Kenneth Creek site had the additional advantage of having an adjacent fertilization trial established by Rob Brockley in 1993 (Ministry of Forests Experimental Project 886.13), and significant ancillary data (quantification of soil nutrient pools, fertilization response, litterfall, and litter decomposition rates).
2.2 Experimental Design and Treatments

The HC site has uneven stocking and we could only establish 16 circular fixed area plots (10 m radius—sufficient to contain 25 crop trees per plot). In the more uniform but lower density stand at KC, we created 24 plots with a larger 11.5 m radius. With these numbers of plots and trees, we can have 4 treatments (X 4 replicates of each) at both sites, with 2 additional treatments at KC.

All S additions were at 100 kg S / ha, and N (as urea) at 300 kg N / ha, rates that are typical of operational and research fertilization treatments in the central interior. Pre-testing of 24 commercial S products from across North America identified 3 promising candidate tracer materials:

- K$_2$SO$_4$ (IMC Kalium, Evergro Fertilizers, Delta, B.C.): $\delta^{34}$S = 19.2 ‰
- elemental S – S$^0$ (Shell Canada, Cremona, Alta.): $\delta^{34}$S = 22.3 ‰
- “Sulfur 95” - S$^0$ (Agrimax Corporation, Calgary, Alta.): $\delta^{34}$S = 19.3‰

These fertilizers differ from background $\delta^{34}$S values at both sites by > 10 ‰, making them suitable tracers at the planned addition rates.

The treatments at both sites compare either sulphate-S or elemental S (added in combination with N), with N-only and unfertilized controls. N-only and N + elemental S treatments included KCl to remove any confounding effect of K in K$_2$SO$_4$.

Treatments 1-4 are at both sites, with 5-6 at KC only, and were assigned in a completely randomized design:

1. Control
2. N + KCl
3. N + K$_2$SO$_4$
4. N + Shell S\(^0\) + KCl
5. N + Agrimax S\(^0\) + KCl
6. Agrimax S\(^0\)

2.3 Data Collection and Analysis

Pre-treatment soil sampling (September/October, 2001) consisted of litter, F & H horizons, and 0-20 cm mineral soil, each composited from 15 random locations/plot. At KC, the low gravel content allowed us to sample the 20-40 cm mineral soil. Current-year lodgepole pine foliage was sampled in October 2002 before fertilization and composited from 10 crop trees per plot. The full sampling protocol will be repeated 1 and 2 years after fertilization.

Standard soil (total C, N, S, extractable sulphate-S, pH, exchangeable cations, mineralizable N, available P) and foliar (total N and S, microwave digestion, ICP analysis of macro- and micronutrients) analyses will be performed by the Ministry of Forests Analytical Laboratory. S isotope analyses on foliage and soils will be performed in the Stable Isotope Laboratory at the University of Calgary. Concentrations and $\delta^{34}$S values for total S, inorganic sulphate-S, and total sulphate-S will be determined directly, and other fractions (organic S, organic sulphate-S, and carbon-bonded S) will be calculated, using methods discussed by Krouse et al. (1996) and Mayer et al. (1992).

The diameter (dbh), total height, and height to live crown of all crop trees will be measured at fertilization, and again after 2 years. Foliage biomass, measured by needle weights, is a good index of potential radial increment response to fertilization (Brockley and Sheran, 1994), and will be assessed in 2003. Longer-term remeasurements after this project would be at 3- or 5-year intervals.

Fertilizer uptake and fate will be assessed by measuring shifts over time in $\delta^{34}$S values in plant tissues, soil horizons, and soil S fractions. As fertilizer S uptake occurs, the plant tissues will show a shift in $\delta^{34}$S values toward those of the fertilizer, allowing us to determine which of the fertilizers is providing a more rapidly-available supply of S. This tracer signal will also be detectable in the soil, allowing us assess the relative mobility of these S sources, and the soil S forms (organic or inorganic) in which they will ultimately be sequestered.

Treatment effects on lodgepole pine growth and on soil and foliar variables will be examined by one-way analysis of variance (ANOVA), using the general linear model procedure (SAS Institute Inc. 1989). Mensurational data may be subjected to covariance analysis, using appropriate initial measurements as covariates. Three *a priori* questions will be tested using single degree-of-freedom contrasts:

1. Is control different than fertilized treatments?
2. Is N alone different than N+S?
3. Is N+S (sulphate-S) different than N+S(S\(^0\))?
3. Activities

3.1 Research

Despite a late start to field work, we were able to complete all of the time-sensitive field operations by mid-November, 2002, in approximately three weeks after approval of funding. Plot layout at Holy Cross was substantially completed in 2001, and only pretreatment crop tree measurements needed to be done in 2002. At Kenneth Creek, only plot centres had been chosen in 2001, so the following operations needed to occur before fertilizer treatments could be applied: selection and tagging of healthy crop trees, culling of excess trees, marking of plot centres and boundaries, and pretreatment measurement of trees.

Fertilizers were purchased, pre-weighed for each plot, and hand-applied before any significant snow accumulation occurred.

Current-year foliage was sampled before fertilizer application, and analyzed at the Ministry of Forests laboratory. S stable isotope analysis of 2001 pretreatment litter, forest floor, and mineral soils was continued at the University of Calgary.

3.2 Extension

Paul Sanborn gave a presentation, entitled "Forest Fertilization Research: The Next Generation" to an audience of approximately 150 silviculturists at the winter meeting of the Northern Silviculture Committee in Prince George on January 22\textsuperscript{nd} (See abstract in Appendix.)

3.3 Operational variances

All work was completed within the planned budget. Further details are provided in notes accompanying the detailed financial statements.

4. Results and Discussion

Highlights of the pretreatment stand, foliar, and chemical conditions are as follows. (Full documentation of all pretreatment data will be contained in the establishment report manuscript, identified as an extension product for 2003-04.)

4.1 Stand characteristics

Initial measurements indicated that the slightly older KC site (21 vs. 18 years) has mean crop tree heights and diameters approximately 50% larger than at HC, consistent with the higher productivity expected in a moister subzone (Table 1).
Table 1. Lodgepole pine crop tree measurements, November 2002.

<table>
<thead>
<tr>
<th></th>
<th>Dbh (mm)</th>
<th>Height (dm)</th>
<th>Height to Live Crown (dm)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Holy Cross</strong></td>
<td>Mean</td>
<td>93.7</td>
<td>74.2</td>
</tr>
<tr>
<td><em>(n = 400)</em></td>
<td>Std Dev</td>
<td>18.1</td>
<td>9.1</td>
</tr>
<tr>
<td><strong>Kenneth Creek</strong></td>
<td>Mean</td>
<td>145.9</td>
<td>109.1</td>
</tr>
<tr>
<td><em>(n = 600)</em></td>
<td>Std Dev</td>
<td>18.7</td>
<td>9.5</td>
</tr>
</tbody>
</table>

4.2 Soil chemical properties

Site means for selected soil chemical properties indicate that these sites are within the normal range for forest soils in the SBS zone, and in particular, have the very low total and sulphate S concentrations that are typical of the central interior (Table 2). The lower nutrient concentrations in the mineral soil at Kenneth Creek is consistent with the coarser soil textures at that site.

Table 2. Selected pre-treatment soil chemical properties (2001): means of plot composite samples.

<table>
<thead>
<tr>
<th></th>
<th>%</th>
<th>ppm</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total C</td>
<td>Total N</td>
</tr>
<tr>
<td><strong>Holy Cross</strong></td>
<td>L</td>
<td>53.29</td>
</tr>
<tr>
<td><em>(n = 16)</em></td>
<td>FH</td>
<td>40.98</td>
</tr>
<tr>
<td><strong>Kenneth Creek</strong></td>
<td>L</td>
<td>53.56</td>
</tr>
<tr>
<td><em>(n = 24)</em></td>
<td>FH</td>
<td>40.85</td>
</tr>
<tr>
<td></td>
<td>Mineral (0-20 cm)</td>
<td>2.25</td>
</tr>
<tr>
<td></td>
<td>Mineral (20-40 cm)</td>
<td>1.14</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.47</td>
</tr>
</tbody>
</table>

1 Min N = mineralizable N (anaerobic incubation)
2 Avail P = available P (Bray P1 extraction)

4.3 Lodgepole pine foliar analyses

Macro- and micronutrient concentrations of the pretreatment current-year foliage indicated potential concerns with only N and S, based on criteria compiled by Brockley (2001b) (Table 3).

<table>
<thead>
<tr>
<th></th>
<th>Holy Cross</th>
<th>Kenneth Ck</th>
</tr>
</thead>
<tbody>
<tr>
<td>Macronutrients (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>N</td>
<td>1.248</td>
<td>1.125</td>
</tr>
<tr>
<td>P</td>
<td>0.135</td>
<td>0.119</td>
</tr>
<tr>
<td>S</td>
<td>0.0704</td>
<td>0.0685</td>
</tr>
<tr>
<td>Ca</td>
<td>0.146</td>
<td>0.148</td>
</tr>
<tr>
<td>K</td>
<td>0.509</td>
<td>0.412</td>
</tr>
<tr>
<td>Mg</td>
<td>0.099</td>
<td>0.090</td>
</tr>
<tr>
<td>Micronutrients (ppm)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>8.7</td>
<td>7.5</td>
</tr>
<tr>
<td>Cu</td>
<td>2.6</td>
<td>2.4</td>
</tr>
<tr>
<td>Fe</td>
<td>58.6</td>
<td>27.6</td>
</tr>
<tr>
<td>Mn</td>
<td>300.0</td>
<td>372.1</td>
</tr>
<tr>
<td>Zn</td>
<td>44.9</td>
<td>39.2</td>
</tr>
</tbody>
</table>

4.4 S isotopic abundance

Stable isotope analyses of the total S of foliage, forest floor and mineral soils confirmed the results of grab samples collected during site selection in 2001, and indicate that natural abundance $\delta^{34}\text{S}$ values differ from those of the fertilizers by at least 10 ‰ at both sites (Table 4). At the fertilizer addition rates used in the treatments, this will provide sufficient “leverage” to give us a viable tracer experiment.

Table 4. Pretreatment natural abundance $\delta^{34}\text{S}$ values (‰) of total S (site means).

<table>
<thead>
<tr>
<th></th>
<th>Holy Cross</th>
<th>Kenneth Ck</th>
</tr>
</thead>
<tbody>
<tr>
<td>Foliage</td>
<td>5.6</td>
<td>8.2</td>
</tr>
<tr>
<td>L</td>
<td>4.5</td>
<td>7.2</td>
</tr>
<tr>
<td>FH</td>
<td>4.9</td>
<td>8.0</td>
</tr>
<tr>
<td>Mineral (0-20 cm)</td>
<td>4.3</td>
<td>7.4</td>
</tr>
<tr>
<td>Mineral (20-40 cm)</td>
<td>n.s.</td>
<td>6.6</td>
</tr>
<tr>
<td>n.s. = not sampled</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

5. Conclusions

Despite late approval of funding, the outstanding efforts of our contractors, suppliers, and collaborators allowed us to meet our project goals for 2002-03. This marks the successful completion of the establishment phase of this project. As such, these accomplishments will now permit us to do the monitoring and interpretation of treatment responses needed to obtain practical, management-oriented insights into the behaviour of sulphur fertilizers in central interior lodgepole pine stands.
References


Appendix:

Abstract of presentation to Northern Silviculture Committee, January 22, 2003, Prince George, B.C.

Forest Fertilization Research: The Next Generation

Paul Sanborn, Rob Brockley, and Bernhard Mayer

Almost 20 years of tree nutrition research by Ministry of Forests and university researchers have greatly improved understanding of lodgepole pine nutritional status and fertilization response in the B.C. interior. Although the greatest emphasis has been on nitrogen, evidence has accumulated for regionally significant deficiencies of other elements, most notably sulphur. Several fertilization trials in the central interior have shown growth responses to N + S fertilization that significantly exceed those from N-only treatments.

Total S concentrations in soil parent materials in central interior B.C. are among the lowest observed in the glaciated temperate and boreal zones. Environmental factors have further compounded this tendency to low S levels in forest ecosystems in this region: fires cause S loss by volatilization from forest floors and biomass, natural S inputs from the marine atmosphere are blocked by the Coast Mts., and the limited scale of industrial activity has not created an acid rain problem.

Fertilization research to date has compared lodgepole pine growth responses to various forms and levels of S addition, combined with a range of N fertilization rates. Generally, the more soluble sulphate-S forms give a more immediate response, both in foliar S concentrations and subsequent growth rates, than does elemental S. The latter must undergo microbial oxidation to sulphate before becoming available for plant uptake -- a process than can considerably delay or limit fertilization response, especially given the much greater availability of N fertilizers.

The differing behaviour of various S fertilizers requires further study, in particular, to determine quantitatively their uptake by crop trees, and their ultimate fate in the ecosystem. For example, what proportion of the added S fertilizer actually gets taken up by the crop trees? How much is leached below the rooting zone of the soil? In which form(s) is fertilizer S retained in the forest floor and mineral soil, and at what depths?

Such questions are most effectively answered by the stable isotope tracer methods that have been successfully applied in fertilizer N research for decades. Application of fertilizers artificially enriched with the stable isotope $^{15}$N has allowed many otherwise obscure aspects of the soil N cycle to be elucidated.

Unfortunately, the same methods have been much more difficult to apply in S fertilizer research due to the prohibitive cost of materials artificially enriched in $^{34}$S. However, advances in analytical methods have allowed environmental scientists to track the
movement of pollutant-derived S in ecosystems, using tracer methods that exploit relatively small differences in $^{34}\text{S} / ^{32}\text{S}$ ratios between the artificial input and natural background. Our next generation of studies is designed to use similar naturally-occurring differences in stable isotopic signatures to help unravel the fate and transformations of fertilizer S in central interior lodgepole pine stands.

In 2001 and 2002, we established replicated plots at two sites in our region, in the SBSdw3 and SBSwk1, in 18- and 21-year-old PI stands, respectively. Our treatments included N-only (300 kg/ha) and N + S (100 kg/ha) treatments involving both readily-available $\text{K}_2\text{SO}_4$ and elemental S. These S fertilizers have $^{34}\text{S} / ^{32}\text{S}$ ratios that are approximately 10-15 parts per thousand above background levels in soils and biomass at these sites. These differences are large enough, given the magnitude of fertilization rates in relation to the size of existing soil S reserves, to allow us to trace the subsequent fate and transformations of these S inputs.

Acknowledgements

Brian Walker (Fraser Lake Sawmills) provided indispensable assistance throughout the planning and installation of these installations, and we are grateful for his interest and support. Funding was provided by Forest Renewal B.C. (MYA) and Forest Innovation Investment.