

TREE-RING CHEMISTRY RESPONSE IN BLACK CHERRY TO AMMONIUM SULFATE FERTILIZATION AT TWO WEST VIRGINIA SITES

David R. DeWalle¹, Jeffrey S. Tepp², Callie J. Pickens², Pamela J. Edwards³, and William E. Sharpe¹

Abstract: The chemical element content of black cherry (*Prunus serotina* Ehrh.) tree rings showed significant changes related to annual ammonium sulfate treatments on one watershed (Fernow WS-3) which exhibited a significant increase in streamflow N export due to treatment. However, tree-ring, soil and streamflow chemistry did not respond to the same treatment on another watershed (Clover Run WS-9). On WS-3, tree-ring concentrations of P, K, Ca, Mg, B, Zn, S and Sr were higher and concentrations of Mn, Fe, Cu and Al were lower in wood formed either before or after treatment. Lack of response to treatment on WS-9 was largely due to soil retention of N, lack of nitrate leaching and possibly the location of the sampled trees near the watershed mouth. Overall, black cherry tree-ring chemistry was consistent with streamflow chemistry changes caused by treatment and the method shows promise as an index to soil chemistry changes. Sapflow in rings formed prior to treatment and/or radial translocation prevented an exact determination of the year of treatment initiation.

INTRODUCTION

Study of the chemical element content of tree rings, or dendrochemistry, is receiving increased attention as a method of determining changes in the soil chemical environment. Several studies have been conducted which showed, or attempted to show, a link between tree-ring chemistry and soil chemistry. A study by Bondietti and others (1990) with red spruce (*Picea rubens* Sarg.) suggested a link between atmospheric deposition and altered soil chemistry. The relationship between soil pH and tree-ring chemistry was examined by Guyette and others (1992) with Eastern redcedar (*Juniperus virginiana* L.) and DeWalle and others (1991) with a variety of species. Effects of liming were studied by Kashuba-Hockenberry and DeWalle (1994) with scarlet oak (*Quercus coccinea* Muenchh.), DeWalle and others (in press) with red oak (*Q. rubra* L.) and McClenahan and others (1988) with tulip-tree (*Liriodendron tulipifera* L.). No known studies have focused on tree-ring response to soil treatment with known quantities of ammonium sulfate. In addition, concern about the effects of radial translocation or sapflow in xylem raises doubt about the utility of using tree rings to measure the timing of soil changes.

Experiments at two research watersheds maintained by the USDA Forest Service, Northeastern Forest Experiment Station in north central West Virginia (Adams and others 1993) provided an opportunity to determine the dendrochemical response to soil fertilization with ammonium sulfate in black cherry (*Prunus serotina* Ehrh.) trees. In earlier studies with black cherry, DeWalle and others (1991) showed that cation concentrations, such as Mn and Sr, in sapwood were sensitive to soil pH variations. In this paper, we compare the black cherry tree ring chemistry on control and treated areas and in wood formed before and after treatment to determine possible effects of ammonium sulfate treatment and radial translocation/sapflow.

¹Professors of Forest Hydrology, School of Forest Resources and Environmental Resources Research Institute, The Pennsylvania State University, University Park, PA 16802.

²Graduate Research Assistants, School of Forest Resources and Environmental Resources Research Institute, The Pennsylvania State University, University Park, PA 16802.

³Hydrologist, U. S. Forest Service, Northeastern Forest Experiment Station, Parsons, WV 26287.

STUDY AREAS

Experiments were conducted on watersheds maintained by the USDA Forest Service, Northeastern Forest Experiment Station on or near the Fernow Experimental Forest near Parsons in north central West Virginia (39°3'15"N, 79°41'15"W). Treatment and control watersheds on the Fernow were WS-3 and WS-7, respectively. In the Clover Run area, watershed WS-9 and a nearby control area were sampled. WS-9 and the control area are on the Monongahela National Forest near Parsons. At both the Fernow and Clover Run sites, the treatment and control areas are adjacent to one another. Land use, current and past experimental treatments, and dominant vegetation are described for each watershed in Table 1.

Table 1. Comparison of treatments and land use history for watersheds WS-3, WS-7 and WS-9.

Watershed	Treatments	Dates	Area (ha)	Dominant Vegetation
WS-3	Weir Installation	5/51	34.3	<i>Prunus serotina</i>
	Intensive selection cut	10/58-2/59		<i>Acer rubrum</i>
	Repeated cut	9/63-10/63		<i>Betula lenta</i>
	Patch cuttings w/herbicide	7/68-8/68		<i>Fagus grandifolia</i>
	Clearcut, left stream buffer	7/69-5/70		
	Cut buffer, clear channel	11/72		
	Begin ammonium sulfate treatment	1/89		
WS-7	Weir Installation	11/56	24.2	<i>Acer saccharum</i>
	Upper 12.1 ha clearcut	11/63-3/64		
	Herbicide upper cut	5/64-10/69		<i>Betula lenta</i>
	Lower 12.1 ha clearcut	10/66-3/67		<i>Liriodendron tulipifera</i>
	Herbicide lower cut	5/67-10/69		<i>Prunus serotina</i>
WS-9	Farmed	1800s to 1920s	11.6	<i>Larix leptolepis</i>
	Weir Installation	1957		<i>Quercus rubra*</i>
	Bulldozed forest regrowth, left stream buffer	11/83		<i>Prunus serotina*</i>
	Planted to Japanese larch	Spring 1984		<i>Smilax rotundifolia</i>
	Began ammonium sulfate treatment	4/87		<i>Rubus</i> spp.

*buffer zone along stream.

Chemical applications on the two treatment watersheds for this study, WS-3 and WS-9, consisted of aerial applications of granular ammonium sulfate fertilizer (21:0:0:24= N:P:K:S) three times per year to give an annual total application of 167 kg ha⁻¹. Treatments were designed to double the seasonal and annual inputs of N and S occurring in throughfall on these watersheds. Applications varied seasonally with 33 kg ha⁻¹ added generally in March and November and 101 kg ha⁻¹ added in July on each watershed. Applications began in April 1987 on WS-9 and in January 1989 on WS-3. Details of the treatment are given by Adams and others (1993).

The region is part of the Allegheny Plateau with steep slopes and shallow soils. Soils are predominantly loamy-skeletal, mixed mesic Typic Dystrochrepts of Calvin channery silt loam type derived from shale and sandstone from the Hampshire formation. Precipitation averages about 145 cm per year with even seasonal distribution. Mean monthly air temperatures average about -1 to 5° C in January to about 25° C in July. Adams and others (1994) give a detailed description of the hydrometeorology of the Fernow Experimental Forest region.

METHODS

Wood samples were taken from trees growing on two pairs of treated and control areas using different methods due to size of trees. On WS-3 and WS-7, 3-cm thick wood disks taken at breast height were harvested from five pole-sized black cherry trees felled at each site on July 29, 1992. On July 2-3, 1992, 4-mm diameter increment cores were collected with a standard increment borer at breast height at four locations around the circumference of five mature black cherry trees in the streamside buffer zone on WS-9 and the control area. Other species also were sampled, but black cherry was the only species sampled that occurred at Fernow and Clover Run sites.

Wood disks from WS-3 and WS-7 were sanded, then a band saw was used to cut the wood disks into quadrants and quadrants into radial strips. Radial strips were separated into age-class sections using a utility knife under low power magnification. Wood from each quadrant was divided into the following sections according to the years in which wood was formed:

Treatment Period	1989-1992
Pre-treatment I	1986-1988
Pre-treatment II	1981-1985

The sanded edges were removed prior to chemical analysis with a stainless-steel chisel to avoid chemical contamination by the sand paper. Samples from each quadrant were analyzed separately, but for the purposes of this paper data for all quadrants were combined.

For Clover Run sites, increment cores also were separated by hand with a scalpel under low magnification into three sections based on ring age:

Treatment Period	1987-1992
Pre-treatment I	1982-1986
Pre-treatment II	1977-1981

Wood from each of the four cores per tree was combined by sections for each tree to give enough mass for chemical analysis.

Sterile laboratory rubber gloves were used for all sample collection, handling and preparation. Samples were rinsed with deionized water several times during processing and were stored frozen until submitted for analysis. Wood samples were analyzed for P, K, Ca, Mg, Mn, Fe, Cu, B, Al, Zn, Na, Sr, N and S at the Agricultural Analytical Services Laboratory operated by the College of Agricultural Sciences at Penn State University, University Park, PA. Nitrogen analysis was conducted using the Dumas method (Campbell 1991) and sulfur analysis using ICP emission spectroscopy (Huang and Schulte 1985). Analysis for the other elements was also by ICP emission spectroscopy (Dahlquist and Knoll 1978).

Soil samples were collected in spring 1994 at rooting depth (0-15 cm) around sample trees on WS-9 and its control area. Samples were collected around black cherry (n=5) and red oak trees (n=5) in the treated and control areas. Soil chemical analyses were performed at the Agricultural Analytical Services Laboratory at Penn State using standard soil tests (North Dakota State Univ. 1988). Since soil chemistry did not vary between species at a site, soil data were

grouped for later comparisons between treatment and control areas. Statistical analysis involved simple t-tests to determine significant differences between treatment and control areas in soil data and tree-ring data by age classes.

RESULTS AND DISCUSSION

Treatment Effects at Each Site

Black cherry tree-ring chemistry changes in response to ammonium sulfate additions on WS-3 were quite pronounced (Table 2). Wood formed during the period 1989-1992, after initiation of treatment on WS-3, showed significant increases in P, K, Ca, B, Sr and S and significant decreases in Mn concentrations relative to WS-7. Pre-treatment wood on WS-3 formed during the 1986-88 (Pre-treatment I) period showed significant increases in P, K, Ca, Mg, B, Zn, Sr, and S and significant decreases in Al, Cu, Fe, and Mn concentrations. For the 1981-85 (Pre-treatment II) period, wood on WS-3 also showed significantly higher Ca, Mg, and Sr and significantly lower Mn, Cu, and Na relative to WS-7. Interestingly, more numerous and generally larger significant differences occurred in wood formed immediately prior to initiation of treatment (Pre-treatment I) than in wood formed after treatment. Nearly all of the differences between WS-3 and WS-7 were significant at the $p \leq 0.01$ level.

Table 2. Mean tree-ring element concentrations (mg/kg) for five black cherry trees on watersheds WS-3 (T=treated) and WS-7 (C=control) for periods of wood formation after and before initiation of soil ammonium sulfate treatments.

Element	Treatment 1989-92		Pre-treatment I 1986-88		Pre-treatment II 1981-85	
	C	T	C	T	C	T
P	122	146.0**	50.5	66.0**	20.0	29.5
K	1015	1213.0*	356	421.0**	225	234
Ca	431	502.0**	331.5	504.0**	171	320.0**
Mg	103	113	65	98.0**	27.5	57.0**
Mn	76.3	22.4**	66.4	22.5**	29.0	14.0**
Fe	10.9	10.9	8.1	5.25*	6.25	8.4
Cu	1.87	1.78	1.5	1.29*	1.76	0.98*
B	3.45	3.73*	2.72	2.98*	2.62	2.73
Al	8.6	1.7	2.9	1.30*	1.1	1.1
Zn	2.65	5.7	1.72	3.49*	2.49	2.06
Na	11.15	10	9.85	8.3	7.35	5.85*
Sr	3.91	5.22*	3.47	5.36*	2.78	4.08*
N	1273	1321	897	950	743	722
S	82.3	90.4*	60.0	64.4*	61.8	53.9

* = significant difference between C and T at $\alpha \leq 0.05$ level, ** at $\alpha \leq 0.01$ level

In contrast, mean black cherry tree-ring chemistry at WS-9 showed relatively minor response to ammonium sulfate applications (Table 3). Tree rings on WS-9 formed during the 1987-92 period, after treatment initiation, showed significant increases in only Ca and Fe concentrations relative to the control area. Wood formed for the five-year period immediately prior to treatment (1982-86) also showed Fe and Na increases and a decrease in S concentrations.

Table 3. Mean tree-ring element concentrations (mg/kg) for five black cherry trees on watershed WS-9 (T=treated) and a nearby control area (C) for periods of wood formation after and before initiation of soil ammonium sulfate treatments.

Elements	Treatment 1987-92		Pre-treatment I 1982-86		Pre-treatment II 1977-81	
	C	T	C	T	C	T
P	90	80	60	50	20	20
K	670	700	460	420	230	290
Ca	390	430.0*	390	390	200	250
Mg	120	130	110	90	40	50
Mn	49.2	56.4	45.6	46.8	18.6	24.6
Fe	49.8	97.8*	27.6	84.2*	27.4	94.8
Cu	2.1	1.8	1.7	1.4	1.5	1.4
B	3.4	3.4	3.0	2.9	2.9	2.7
Al	3.8	4.4	2.9	3.3	2.9	2.4
Zn	154.0	30.2	112.2	17.6	68.0	22.0
Na	17.2	11.4	13.0	9.4*	11.0	7.4
Sr	3.6	3.6	3.6	3.5	2.7	2.9
N	840	910	640	610	520	570
S	460	460	440	430.0*	440	440

* = significant difference between control and treatment at $\alpha \leq 0.05$ level

All of these changes were significant at the $p \leq 0.05$ level. Lowering $p \leq 0.1$ would only have added significant changes in Zn and Na in Pre-treatment II samples and in Zn in Post-treatment samples, respectively. Pre-treatment wood formed during the 1977-1981 period showed no significant differences relative to the control area.

Causes of Treatment Differences between Sites

Large differences in tree-ring response to ammonium sulfate treatment occurred between watersheds WS-3 and WS-9. Increased base cation concentrations in wood on WS-3 are consistent with the mobilization of base cations in soil solution to maintain a charge balance with the anions added by treatment. Sulfate anions were added directly in the ammonium sulfate treatment and nitrification of added ammonium would produce NO_3 anions. Nitrogen fertilization from the added ammonium also may have increased rates of decomposition of soil organic matter which increased the overall supply of base cations. Sulfur increases in wood are most likely a direct result of the soil treatment. Reductions in trace metal concentrations (Al, Cu, Fe, Mn) may have been caused by increased base cation availability. Tree-ring trace metal concentration reductions on WS-3 also imply that soil acidification due to three years of treatment has not occurred yet, since soil acidification should generally lead to increased availability of trace metals.

In contrast to WS-3 results, six years of ammonium sulfate treatment on WS-9 produced very small to negligible tree-ring chemistry effects. Lower site fertility caused by years of mountain farming on Clover Run WS-9 (Adams and others 1993) may have depleted available soil base cations and produced low soil N pools. Consequently, N may be very limiting on WS-9 and more strongly retained. Data for treated watersheds show lower tree-ring Ca and N content on WS-9 than WS-3 in all years (Tables 2 and 3), supporting the suggestion of lower site fertility on WS-9 as a cause of reduced treatment response in tree rings. However, trees sampled on WS-9 were located in a streamside

buffer zone near the mouth of the watershed and may reflect soil conditions in this zone only. Helicopter additions of ammonium sulfate included the streamside buffer zone.

Increases in tree-ring Fe concentrations in wood formed during the 1987-92 and 1982-86 periods on WS-9 may signal initial stages of soil acidification caused by treatment. Treatment and control area differences in tree-ring Na and S at Clover Run cannot be related directly to treatment and may be due to natural site differences.

Soil chemistry data were used to test the hypothesis that ammonium sulfate treatments have affected soil properties near sampled trees at the Clover Run sites. Mean chemistry for 10 soil samples around sampled trees in the streamside buffer zone on WS-9 and the control area (Table 4) showed no significant differences at either the $p \leq 0.05$ or $p \leq 0.1$ levels. No significant change in measured soil properties had occurred due to treatment around sample trees in the buffer zone on WS-9.

Table 4. Comparison of root-zone soil chemical properties around sample trees in the streamside buffer zone on watershed WS-9 and nearby control site.

Parameter	Control	WS-9
pH	4.57	4.43
P (kg/ha)	17.3	11.4
Acidity (meq/100g)	8.37	9.59
K	0.075	0.084
Mg	0.16	0.18
Ca	0.63	0.61
CEC	9.24	10.44

* indicates significant difference at the 0.05 level

Published streamflow chemical export data provide some checks on the tree-ring results for both sites. Differences in black cherry wood chemistry response to ammonium sulfate treatment between watersheds WS-3 and WS-9 were consistent with differences in NO_3 and Ca export in streamflow in response to treatment on these watersheds (Adams and others 1993). On WS-3, where tree-ring chemistry was markedly changed by treatment, large increases in NO_3 and Ca export were noted. On WS-9, where little tree-ring response was found, no significant increases in NO_3 or Ca were found. No significant increases in streamflow export of SO_4 were found on either watershed. Soil sulfate adsorption probably is responsible. Increased export on WS-3 indicates that treatment indeed did mobilize cations and tree-ring chemistry was able to record those changes. Lack of streamflow export changes on WS-9 also is consistent with no soil chemical changes and limited tree-ring chemistry changes in response to treatment on this watershed.

Some soils data do exist to help interpret dendrochemical results. Gilliam and others (1994) found that inorganic soil $\text{NO}_3\text{-N}$ concentrations at the 0-10 cm depth were significantly higher on WS-3 than on WS-7 after 3 years of treatment in support of the stream chemistry differences. Mean soil calcium concentrations on WS-3 were also much greater than on WS-7 but the differences were not significant due to high soil Ca variability on WS-3. Differences in soil K, Mg, P, $\text{NH}_4\text{-N}$, Cu, Fe, Mn, and Zn between WS-3 and WS-7 were not significant. The authors do cite unpublished data for the 0-5 cm soil depth which showed that soil on WS-3 was significantly more acidic than on WS-7. Although these soil data show NO_3 changes on WS-3 due to treatment, large changes in base cation availability as suggested by tree-ring chemistry data are not supported.

On the Clover Run WS-9 basin, Pickens and others (1995) present soils data which show that significant changes in soil Al, Mn, N, S, Ca, Mg, and pH occurred due to ammonium sulfate treatments. Only small tree-ring Ca increases could be attributed to treatment on WS-9. Tree ring samples were collected in the streamside buffer zone near the mouth of the watershed on WS-9, while soils data from Pickens and others (1995) were collected in the uplands portion of this basin. Considering that both tree-ring and stream chemistry data (Adams and others 1994) do not indicate acidification of WS-9 due to ammonium sulfate treatments, soils data presented by Pickens and others (1995) suggest a gradual acidification of soils on WS-9 from the uplands to the lowlands which has not yet affected the tree-rings, soils and water at the mouth of the basin.

Apparently, cation mobilization is largely due to NO_3 rather than SO_4 anions on WS-3. No significant changes in tree-wood N concentrations were found, even though NO_3 concentrations were increased in soil solution. Kashuba (1992) also found that scarlet oak tree-ring chemistry did not change in response to soil N fertilization. Tree-ring chemistry is more sensitive to base cation element availability changes, than N changes, since base cations can bind to pectic compounds in the cell wall of xylem. (Ferguson and Bollard 1976; Tomlinson and Tomlinson 1990). Nitrogen is not similarly adsorbed.

Radial Variations of Treatment Effects

Contrary to expectations, wood formed prior to, rather than following, treatment appeared to be affected most on WS-3. A larger number of significant differences were observed for the 1986-88 period, just prior to treatment, than for wood from the 1989-92 period occurring after treatment. Several explanations can be given for this occurrence: 1) radial translocation of elements could have occurred which would redistribute elements from post-treatment to pre-treatment wood, 2) previous watershed treatments could have affected tree rings on WS-3 or WS-7 in such a way that wood formed before ammonium sulfate treatment was inherently different between watersheds, and 3) ammonium sulfate treatment effects on WS-3 may be changing gradually over time.

Radial Translocation/Sapflow. Radial translocation of elements in xylem often has been suggested as an explanation of a treatment response in tree rings formed in years prior to treatment (McClenahan and others 1988). Parenchyma cells in wood rays represent living tissue, and radial translocation of carbohydrate in rays has been documented. Thus, radial translocation of chemicals could have occurred from rings affected by treatment to rings formed before treatment. However, this issue is controversial since tree rings formed in years just prior to treatment can also be affected by treatment, if those rings were participating in vertical sapflow at the time of treatment. In tree species with diffuse porous wood, such as black cherry, sapflow occurs in a band of sapwood of indeterminate thickness. By definition, sapflow cannot occur in heartwood. The number of annual rings involved in sapflow during treatments in this experiment is not known, but observations of sapwood thickness can at least be used to indicate the number of rings potentially available for sapflow.

Sapwood included most if not all of the wood analyzed on the two watersheds. The heartwood/sapwood boundary observed when the wood samples were cut in 1992 was located between 4-9 annual rings from the bark (mean 6.1 years) on WS-7 and 7-11 annual rings from the bark (mean 8.5 years) on WS-3. Since wood only up to 11 years of age at the time of sampling was considered in this study and the sapwood/heartwood boundary probably migrated toward the bark after treatment, sapflow probably could have occurred at some time after treatment was initiated in most, if not all, of the 11 annual rings analyzed.

Response to ammonium sulfate treatment in wood formed immediately prior to the treatment period (1986-88) could be easily explained by sapflow at treatment time in these three rings. Treatment effects on wood formed during the 1981-85 period implies that one or more annual rings formed 4-8 years prior to treatment also contributed to sapflow when treatments were initiated. Sapflow in at least some of these rings cannot be ruled out.

Effects of Past Treatments. Experimental treatments on Fernow watersheds WS-3 and WS-7 prior to the time of ammonium sulfate treatment may have contributed to differences in the chemistry of black cherry wood, but this is not considered likely. These watersheds were subjected to different forest cutting and herbicide treatments as part of

earlier experiments (Table 1); however, the trees on each basin had been allowed to regrow naturally for at least 19 years prior to ammonium sulfate treatments. Wood formed only up to 8 years prior to ammonium sulfate treatment was analyzed in this study. This wood probably was not affected significantly by prior experimental treatment, but this can not be established with certainty.

Changing Treatment Effects. Since ammonium sulfate treatments were applied each year from 1989 through 1992 on WS-3, gradually changing soil response to treatments also could have influenced radial variations. Continued ammonium sulfate treatments are expected eventually to deplete base cations, cause soil acidification, and increase availability of trace metals. Timing of such base cation depletion/acidification cannot be predicted. That wood formed after treatment showed fewer and smaller significant differences than wood formed before treatment suggests that base cation depletion had begun after only three years of treatment, although no direct evidence for soil acidification was found. As treatment of WS-3 continues, tree-ring chemistry may eventually show soil acidification effects.

CONCLUSIONS

The chemical element content of black cherry tree rings is an indicator to the effects of ammonium sulfate fertilization on the soil which is at least as sensitive as measurements of streamflow or soil chemistry. Sapflow and/or radial translocation in black cherry wood may interfere with the detection of the precise timing of soil changes using dendrochemistry.

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