

# Effect of aluminum on the growth and nutrition of tulip-poplar seedlings

Heidi B. Lux and Jonathan R. Cumming

**Abstract:** Atmospheric deposition of nitrogen and sulfate is acidifying some ecosystems, potentially increasing the availability of aluminum in the soil solution. The effects of Al on tulip-poplar (*Liriodendron tulipifera* L.) were investigated with seedlings grown in sand culture. Seedlings were exposed to Al at four concentrations ranging from 0 to 800  $\mu\text{M}$ . Reductions of 77% in shoot biomass and 71% in root biomass were observed at 200  $\mu\text{M}$  Al. Aluminum reduced the concentrations of multivalent cations in leaves and roots. The Ca concentration of leaves was reduced at 400  $\mu\text{M}$  Al, and the Mg concentration of roots was significantly reduced at 200  $\mu\text{M}$  Al. Relationships between tissue Ca, Mg, and Al indicated that Al alters the translocation of Ca and Mg to the shoots and the accumulation of Mg in the roots. If mature trees respond in a similar fashion as seedlings noted here, then tulip-poplar has a low threshold for Al toxicity, which may lead to declines in the health of tulip-poplar populations in ecosystems exposed to elevated levels of acidic deposition.

**Résumé :** Les dépôts atmosphériques d'azote et de sulfate causent l'acidification de certains écosystèmes, ce qui pourrait augmenter la disponibilité de l'aluminium dans la solution du sol. Les effets de Al sur le tulipier de Virginie (*Liriodendron tulipifera* L.) ont été étudiés avec des semis cultivés dans le sable. Les semis ont été exposés à quatre concentrations de Al allant de 0 à 800  $\mu\text{M}$ . Des réductions de 77% de la biomasse de la tige et de 71% de la biomasse racinaire ont été observées en présence de Al 200  $\mu\text{M}$ . L'aluminium a réduit la concentration des cations multivalents dans les feuilles et les racines. La concentration de Ca foliaire a été réduite avec Al 400  $\mu\text{M}$  et la concentration de Mg dans les racines a été significativement réduite avec Al 200  $\mu\text{M}$ . Les relations entre Ca, Mg et Al dans les tissus montrent que Al altère la translocation de Ca et Mg vers la tige et l'accumulation de Mg dans les racines. Si les arbres matures réagissent de façon similaire à ce que nous avons observé chez les semis, cela signifie que le tulipier de Virginie a un seuil de toxicité à Al bas, ce qui pourrait entraîner un déclin de la santé des populations de tulipier de Virginie dans les écosystèmes exposés à des niveaux élevés de dépôts acides.

[Traduit par la Rédaction]

## Introduction

Aluminum has marked impacts on plants, causing disruptions in nutrient acquisition and assimilation, root development, and growth (Foy et al. 1978; Delhaize and Ryan 1995; Kochian 1995). There is a wide range of Al sensitivity in crop species, and in environments where Al levels are high, this sensitivity can limit growth and yield (Foy et al. 1978). Forest tree species also exhibit variable sensitivity to Al (Kelly et al. 1990; Raynal et al. 1990; Sucoff et al. 1990), and in acidic forest soils, Al may affect tree growth (Shortle and Smith 1988). Furthermore, as soil systems become acidified because of elevated nitrogen and other acid inputs from the atmosphere, Al is increasingly mobilized in the soil solution (Joslin and Wolfe 1992). Exposure to Al in acidified soils may lead to declines in tree health (Ulrich 1984; Shortle and Smith 1988) and, ultimately, changes in species composition in some forest ecosystems.

While the impacts of Al on several forest tree species have been reported (Kelly et al. 1990; Raynal et al. 1990; Sucoff et al. 1990), the responses of many other ecologically

and commercially important species have not been assessed. Tulip-poplar (*Liriodendron tulipifera* L.) is a major tree species in southern Appalachian deciduous forests. Given that these same forests receive considerable inputs of acidic deposition (Adams et al. 1993; Peterjohn et al. 1996), our goal was to assess the sensitivity of tulip-poplar to Al, focusing on changes in growth and nutrition brought about by exposure to Al in sand culture.

## Materials and methods

First-year seedlings of tulip-poplar were collected from a deciduous forest opening near Parsons, W.V., in August 1997. Seedlings were planted in flats of washed sand, watered as needed, and allowed to acclimate to the sand environment in a greenhouse for 3 weeks. Subsequently, seedlings were individually transplanted to D16 tree tubes (Stuewe and Sons, Corvallis, Oreg.) containing 200  $\text{cm}^3$  acid-washed sand. These were placed in a growth chamber with a 14-h light period at 24°C followed by a 10-h dark period at 19°C. Photosynthetically active radiation was 250  $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ , and relative humidity was maintained at 60%. Plants were watered twice daily with 20 mL of nutrient solution using an automated pump system (Cumming and Weinstein 1990). Before Al addition and pH adjustment, solutions contained 1.2 mM  $\text{NO}_3^-$ ; 0.4 mM  $\text{NH}_4^+$ ; 0.5 mM K; 0.2 mM Ca; 0.05 mM P; 0.1 mM Mg and  $\text{SO}_4^{2-}$ ; 50.5  $\mu\text{M}$  Cl; 25  $\mu\text{M}$  B; 2  $\mu\text{M}$  Mn and Zn; and 0.5  $\mu\text{M}$  Cu, Na, Co, and Mo. Aluminum treatments of 0, 200, 400, and 800  $\mu\text{M}$  Al as  $\text{Al}_2(\text{SO}_4)_3$  were delivered in the nutrient solution to each plant

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**H.B. Lux and J.R. Cumming,**<sup>1</sup> Department of Biology, West Virginia University, Morgantown, WV 26506, U.S.A.

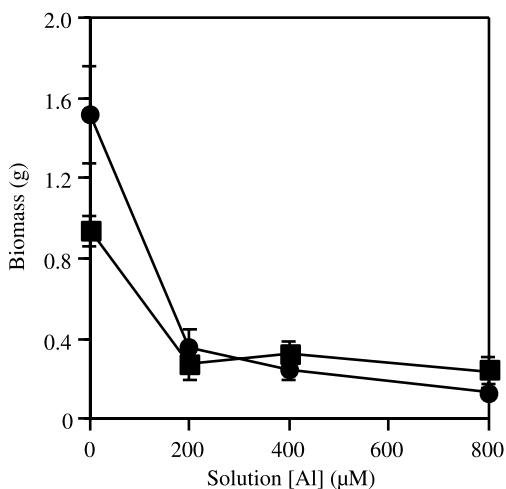
<sup>1</sup>Corresponding author. e-mail: jcumming@wvu.edu

**Table 1.** Concentration of Al and macronutrient elements in leaves and roots of *Liriodendron tulipifera* seedlings exposed to Al in sand culture.

Solution Al ( $\mu\text{M}$ )	Al ( $\mu\text{g}\cdot\text{g}^{-1}$ )	Ca ( $\text{mg}\cdot\text{g}^{-1}$ )	Mg ( $\text{mg}\cdot\text{g}^{-1}$ )	K ( $\text{mg}\cdot\text{g}^{-1}$ )	P ( $\text{mg}\cdot\text{g}^{-1}$ )
<b>Concentration in leaves</b>					
0	161 $\pm$ 27c	7.41 $\pm$ 0.54a	1.03 $\pm$ 0.08a	5.68 $\pm$ 0.56b	1.01 $\pm$ 0.14a
200	431 $\pm$ 102bc	5.47 $\pm$ 0.69ab	0.99 $\pm$ 0.11a	8.71 $\pm$ 0.73a	1.22 $\pm$ 0.18a
400	635 $\pm$ 106ab	4.23 $\pm$ 0.60b	0.80 $\pm$ 0.09a	8.73 $\pm$ 0.63a	1.07 $\pm$ 0.16a
800	824 $\pm$ 118a	5.00 $\pm$ 0.85ab	0.70 $\pm$ 0.13a	4.88 $\pm$ 0.89b	0.92 $\pm$ 0.22a
<i>P</i> *	<0.001	0.016	0.137	0.004	0.711
<b>Concentration in roots</b>					
0	551 $\pm$ 142b	3.09 $\pm$ 0.17a	1.57 $\pm$ 0.08a	12.4 $\pm$ 0.6a	1.39 $\pm$ 0.13a
200	1914 $\pm$ 132a	3.04 $\pm$ 0.22a	1.18 $\pm$ 0.10b	13.7 $\pm$ 0.8a	1.58 $\pm$ 0.17a
400	2080 $\pm$ 422a	2.47 $\pm$ 0.19ab	0.91 $\pm$ 0.09bc	13.8 $\pm$ 0.7a	1.43 $\pm$ 0.15a
800	1606 $\pm$ 133a	2.13 $\pm$ 0.17b	0.68 $\pm$ 0.08c	10.4 $\pm$ 0.6b	0.97 $\pm$ 0.13a
<i>P</i> *	0.002	0.006	<0.001	0.006	0.054

**Note:** Values are means  $\pm$  SE ( $n = 3$ ). Leaves or roots from two plants per treatment were pooled, ground, and digested together prior to elemental analyses. Means followed by different letters are significantly different ( $P < 0.05$ ) based on the Tukey–Kramer HSD test.

\*Level of significance of the Al effect.

**Fig. 1.** The effect of Al in solution on biomass of shoots (circles) and roots (squares) of tulip-poplar seedlings grown in sand culture. Values are means  $\pm$  SE.

( $n = 7$ ). All solutions were adjusted to a pH of 4.0 with HCl or NaOH as necessary. Aluminum concentrations in excess solution draining from the tree tubes were periodically assessed by the eriochrome cyanine R method (American Public Health Association 1985). Values for the 200 and 400  $\mu\text{M}$  treatments were within 10% of target concentrations; the 800  $\mu\text{M}$  treatment was typically low because of the formation of aluminum hydroxide and (or) phosphate precipitates (Snoeyink and Jenkins 1980).

Plants were harvested at the end of a 9-week exposure period. Stems were clipped at the sand surface and were washed in a 0.1% Tween-80 solution, rinsed with deionized water, dried at 60°C for 24 h, and weighed. Roots were rinsed free of sand and dried at 60°C for 24 h and weighed. Leaves were separated from stems, and all leaf and root tissue was ground to pass a 1-mm screen. Because of the small plant size at the higher Al concentrations, tissue from two plants per treatment was pooled for elemental analysis ( $n = 3$  for these analyses). Tissue was analyzed for total N using a Carlo Erba 1500 NCS analyzer. Subsamples of tissue were digested using a sulfuric acid –  $\text{H}_2\text{O}_2$  digest (Parkinson and Allen 1975) and analyzed on a Perkin Elmer P400 inductively coupled

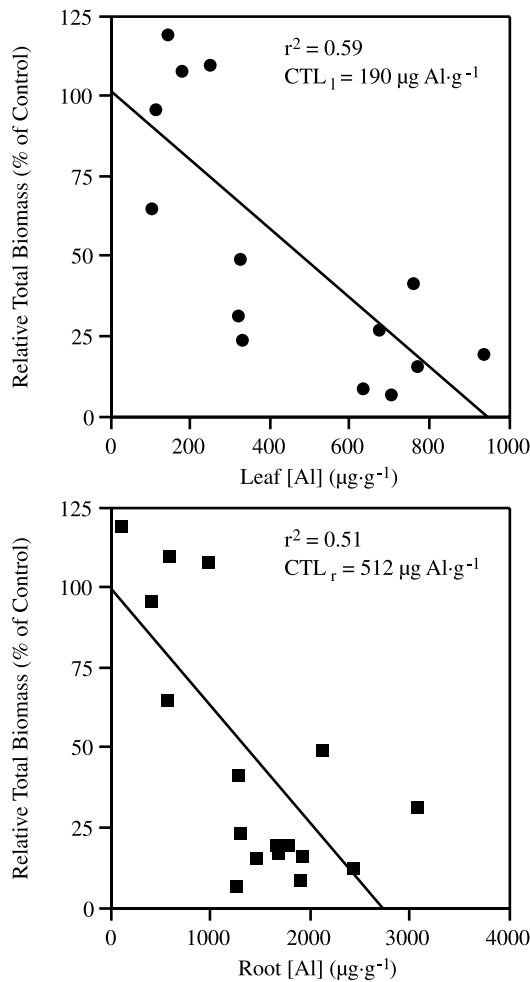
plasma emission spectrophotometer for Al, P, Ca, and Mg. Tissue K was analyzed on a Perkin Elmer 5000 atomic absorption spectrophotometer using the same digests.

Biomass and nutrition data were analyzed using one-way analyses of variance. Biomass data exhibited heterogeneous variances and were log transformed prior to analysis. For clarity, non-transformed growth data are presented in the figures. Means separations were undertaken using the Tukey–Kramer HSD protocol. Regression analyses were undertaken for nutrition variables to assess patterns of accumulation and relationships between nutrients. All analyses were implemented using the statistical package JMP (SAS Institute Inc., Cary, N.C.).

## Results and discussion

Tulip-poplar seedlings were sensitive to Al, with 77 and 71% reductions in shoot and root mass, respectively, occurring between 0 and 200  $\mu\text{M}$  Al (Fig. 1). Plants grown under the 400 and 800  $\mu\text{M}$  treatments were not statistically smaller than those grown at 200  $\mu\text{M}$  Al, although plants grown at 800  $\mu\text{M}$  Al exhibited leaf lesions. The Al concentration of foliage of tulip-poplar seedlings increased linearly with solution Al concentration (Table 1). In contrast, roots accumulated up to 2.08  $\text{mg Al}\cdot\text{g}^{-1}$  when Al was present in the nutrient solution, in a pattern independent of external Al concentration (Table 1). Reductions in whole plant growth were correlated with foliar and root tissue Al concentrations (Fig. 2). Relationships between growth and tissue metal concentrations have previously been used to calculate the critical toxicity level (CTL) for a given metal (Thornton et al. 1986). The CTL is the tissue concentration of a metal at which whole plant biomass is reduced by 20% in comparison with control plants (Thornton et al. 1986). This index is a reflection of a species' sensitivity to a metal as it is accumulated in tissues. For tulip-poplar, the calculated CTL was 190  $\mu\text{g Al}\cdot\text{g}^{-1}$  in leaf tissue and 512  $\mu\text{g Al}\cdot\text{g}^{-1}$  in root tissue (Fig. 2). Honey locust (*Gleditsia triacanthos* L.), the most sensitive tree species studied to date (Sucoff et al. 1990), exhibited leaf and root CTLs of 40  $\mu\text{g}\cdot\text{g}^{-1}$  and 2960  $\mu\text{g}\cdot\text{g}^{-1}$ , respectively. The leaf CTL of tulip-poplar is comparable with

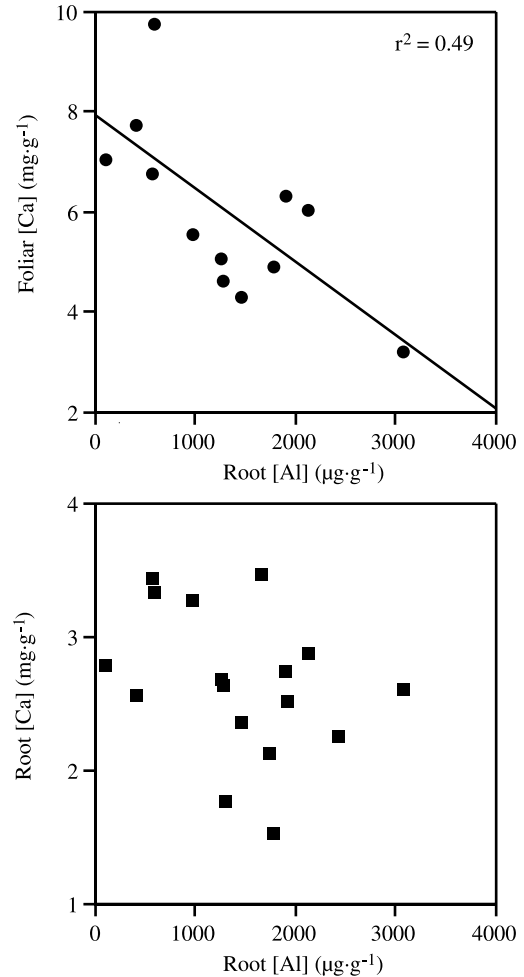
**Fig. 2.** Relationship between whole plant seedling biomass and leaf (top) or root (bottom) Al concentrations. Critical toxicity thresholds (CTLs) are calculated as the tissue Al concentration at which plant growth is reduced by 20% in comparison with controls (Thornton et al. 1986).



that of northern red oak (*Quercus rubra* L.) (118 µg·g<sup>-1</sup>; Kruger and Sucoff 1989) and lower than sugar maple (*Acer saccharum* Marsh.) (320 µg·g<sup>-1</sup>; Thornton et al. 1986). The root CTL for tulip-poplar is substantially lower than CTLs of both red oak (7178 µg·g<sup>-1</sup>) and sugar maple (>5000 µg·g<sup>-1</sup>). Taken together with dose-response data (Fig. 1), these patterns indicate that tulip-poplar is extremely sensitive to the accumulation of Al in root tissues.

Aluminum in the rhizosphere altered the concentrations of nutrients in roots and leaves. Root Ca concentrations were reduced by 20–31% when solution Al levels were equal to or greater than 400 µM (Table 1). These changes in root Ca concentrations were paralleled by larger reductions in leaf Ca concentrations, with Al reducing leaf Ca concentration by 43% at the 400 µM level (Table 1). Root Mg concentrations were reduced by 25–57% at and above 200 µM Al (Table 1). Foliar Mg concentration declined with exposure to Al, but differences among means were only marginally significant ( $P = 0.14$ ). The concentrations of K in roots and shoots exhibited complex patterns as a result of exposure to

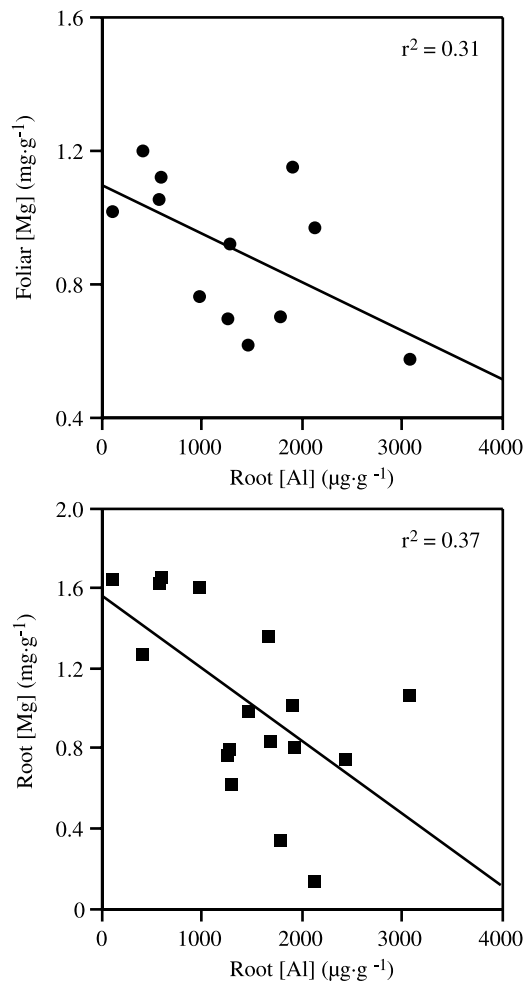
**Fig. 3.** Relationship between the concentrations of Ca in foliage (top) or roots (bottom) and root Al concentrations of tulip-poplar seedlings exposed to Al in sand culture.



Al (Table 1). Increases in tissue concentration possibly reflect reverse growth dilution effects, where Al limited growth without an impact of Al on K uptake and translocation (Marschner 1995). Reductions in tissue K concentrations at 800 µM Al may reflect reductions in the uptake and translocation of K. Aluminum did not affect tissue N concentrations (roots, 1.85%; leaves, 2.28%) or tissue P concentrations (Table 1).

The impacts of Al on the accumulation of Ca and Mg in roots and the subsequent translocation of these nutrients to shoots may play a critical role in plant response to Al in the rhizosphere (Godbold et al. 1988; Shortle and Smith 1988; Kochian 1995). The concentrations of Ca and Mg in roots represent both apoplastic and symplastic pools (Marschner 1995). Foliar concentrations similarly represent apoplastic and symplastic pools; however, delivery to the shoot is primarily dependent upon apoplastic transfer from the soil solution to the stele in the root (Ferguson and Clarkson 1976) and subsequent movement in the transpiration stream to the leaf (Marschner 1995). In the present study, foliar Ca concentrations, but not root concentrations, were negatively correlated with root Al concentrations (Fig. 3). In contrast, both

**Fig. 4.** Relationship between the concentrations of Mg in foliage (top) or roots (bottom) and root Al concentrations of tulip-poplar seedlings exposed to Al in sand culture.



foliar and root Mg concentrations were negatively correlated with root Al concentrations (Fig. 4). These patterns suggest that Al may alter the uptake and translocation of Ca and Mg via different mechanisms. Although speculative, the patterns we observed suggest that Al (adhering to the Donnan free space; Godbold et al. 1988; Cronan 1991) is very effective at displacing Mg from the apoplast and reducing its movement to the transpiration stream. In contrast to Mg, root Ca concentration was not correlated with root Al concentration in tulip-poplar. This suggests that the ionic interference between Al and Ca is not as significant as that between Al and Mg in controlling cation transfer to the shoot, possibly because of the higher concentration of Ca than Mg in the nutrient solution used. If Donnan interactions are not operating to reduce Ca in the root, then Al must be impacting Ca transfer to the shoot by an alternative mechanism. This could possibly be by a reduction in the area of root capable of delivering Ca to the xylem (through reduced root growth and increased root suberization) or through the loss of transpiration-supported solution flow due to lower leaf area on Al-treated plants (Rengel and Robinson 1989). As tulip-poplar has a high Ca requirement in contrast to other deciduous tree spe-

cies (Blinn and Buckner 1989), this restriction of Ca translocation to the foliage may play a major role in Al toxicity in this species.

The patterns of root and foliar nutrition observed for tulip-poplar are similar to those reported for other hardwood species, including sugar maple (Thornton et al. 1986), European birch (Goransson and Eldhuset 1987), and red oak (Kruger and Sucoff 1989). However, a major difference is the observed changes in tulip-poplar occurred at lower external Al concentrations than in these other species. We grew tulip-poplar on a low concentration nutrient solution because the sensitivity of plants to Al is influenced by ionic strength of the nutrient solution (Blamey et al. 1991); our use of a low concentration solution may have aided in uncovering the sensitivity of tulip-poplar to Al. While the growth of seedlings exposed to solutions without Al did not appear to be limited by the nutrient solution, the concentrations of nutrients in foliage did reflect lower nutrient availability. The concentrations of P and K in foliage of seedlings from the present study were approximately one half and that of Ca and Mg were one fifth the values reported by Blinn and Buckner (1989) for tulip-poplar growing in the field.

Tulip-poplar is an ecologically and economically important tree species in the southern Appalachian Mountains (Hicks 1998). These same forests receive high levels of acidic deposition (Adams et al. 1993), but there is limited data on the concentrations of Al in the soil solution that may affect tulip-poplar in the field. We have been measuring (unpublished data) soil lysimeter samples from two watersheds in the Fernow Experimental Forest in Parsons, W.V., one of which has been receiving applications of nitrogen at two times ambient deposition levels since 1989 (Adams et al. 1993). Soil-solution Al concentration has averaged 26  $\mu\text{M}$  in the untreated watershed, whereas Al in soil water in the watershed receiving elevated acidic deposition has averaged 107  $\mu\text{M}$ . Given the steep slope of the Al dose-response curve generated for tulip-poplar seedlings (Fig. 1), it is clear that regeneration of this species may be impacted by long-term acidic deposition into forested ecosystems.

## Conclusions

Tulip-poplar shows considerable sensitivity to Al, with substantial reductions in growth and altered patterns of ion accumulation at Al concentrations as low as 200  $\mu\text{M}$ . This sensitivity appears to be the result of the tendency of this species to accumulate Al in tissues, especially roots, as evidenced by low CTLs. The accumulation of Al in the roots of tulip-poplar altered the uptake and translocation of Ca and Mg, which may be the primary factors leading to growth limitation. The CTLs for tulip-poplar were reached at lower solution Al concentrations than those used in studies of other forest tree species, illustrating that tulip-poplar is one of the more Al-sensitive species in the eastern deciduous forest. Although the responses of tree seedlings and mature trees to Al may not be identical, the observed Al sensitivity of tulip-poplar seedlings suggests that Al at concentrations observed in the field may have a significant negative impact on this species by reducing tulip-poplar regeneration or by directly affecting the health of mature trees in forests receiving elevated inputs of acidic deposition.

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