

# Sodium Chloride Injury on Buds of *Acer platanoides*, *Tilia cordata*, and *Viburnum lantana*

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**Abstract.** Dormant lateral buds of Norway maple (*Acer platanoides* L.), littleleaf linden (*Tilia cordata* Mill.), and wayfaringtree viburnum (*Viburnum lantana* L.) were collected and exposed to nine NaCl concentrations: 0, 500, 1,000, 2,000, 4,000, 8,000, 16,000, 32,000, and 64,000 mg/L (0, 500, 1,000, 2,000, 4,000, 8,000, 16,000, 32,000, and 64,000 ppm) in December 2001 and January and March 2002. Electrolyte leakage and visual observations of inner and outer tissue discoloration were used to assess injury. Bud injury generally increased as NaCl concentration increased. Bud morphologies of each species were related to tissue discoloration patterns; naked buds were more susceptible to NaCl than those with bud scales. Buds also exhibited seasonal NaCl resistance; the greatest resistance occurred in December. Norway maple, wayfaringtree viburnum, and littleleaf linden buds experienced 50% electrolyte leakage at calculated NaCl values of 12,941, 16,901, and 42,594 mg/L (12,941, 16,901, and 42,594 ppm) NaCl, respectively, but no severe inner tissue discoloration occurred at any level of NaCl treatment. In January, 50% electrolyte leakage occurred at lower NaCl concentrations in Norway maple [7,165 mg/L (7,165 ppm)] and littleleaf linden buds [27,118 mg/L (27,118 ppm)]. Moderate to severe inner tissue injury was detected for all species at 1,000 mg/L (1,000 ppm) NaCl. Buds were most susceptible to NaCl injury in March, with moderate to severe inner tissue discoloration occurring in wayfaringtree viburnum and littleleaf linden buds at 500 mg/L (500 ppm) NaCl.

**Key Words.** Bud morphology; electrolyte leakage; salt tolerance; tissue discoloration; winter injury.

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Winter roads and walkways must be maintained ice-free to provide safe mobility for pedestrians and motorists. De-icing salts are used extensively to melt ice and snow and are then dispersed from road surfaces by plowing, runoff, and aerosol spray generated by traffic and wind, causing significant damage to plants (Hootman and Kelsey 1992). Sodium chloride (NaCl) is the most commonly used de-icing salt because of its effectiveness, availability, and comparatively low cost. However, the deleterious effects of NaCl to roadside trees and shrubs are well documented (Lumis et al. 1973; Hofstra et al. 1979; Dobson 1991). Plants are injured by NaCl either by root uptake or deposition onto plant surfaces causing osmotic stress and toxicity to tissues. Elevated soil salinity is generally localized to areas bordering salted roadways (Hootman et al. 1994), whereas salt spray travels greater distances. Hootman and Kelsey (1992) found that salt spray increased Na levels in needles of eastern white pine (*Pinus strobus* L.) up to 1,018 m (3,340 ft) from tollways and detected visible salt spray damage at 378 m (1,240 ft). Hofstra et al. (1979) similarly reported injurious Na and Cl accumulation in needles of white pine at distances of 100 m (330 ft) or more from roadways but found no elevated levels of Na and Cl in the soil at distances greater than 30 m (99 ft) from pavement.

Numerous studies have evaluated species for salt spray tolerance (Moxley and Davidson 1973; Lumis et al. 1975; Townsend and Kwolek 1987). However, salt spray concen-

trations in roadside observation studies were not quantified, and the salt concentrations used for species evaluations have varied. There is little information about the occurrence and severity of injury at different salt concentrations. Landscape plants are exposed to a wide range of salt concentrations, which vary by location, method and amount of salt application, and type of storm (Herrick 1988; Buttle and Labadia 1999). More knowledge on the relation between injury and varying salt concentrations is needed.

Salt injury has been correlated with the accumulation of Na<sup>+</sup> and Cl<sup>-</sup> ions in plant tissues (Hofstra and Lumis 1975; Sucoff et al. 1976). The symptoms of salt spray are delayed budbreak, reduced leaf size, marginal leaf scorch, witches' broom (tufted growth), and crown dieback in deciduous species (Lumis et al. 1975; Sinclair et al. 1987; Dobson 1991; Appleton et al. 1999). Salt spray enters deciduous species through nonlignified bud tissues, bud and leaf scars, and young shoots (Dobson 1991). Salt tolerance of many species appears to be related to their ability to preclude salt from entering and accumulating in sensitive tissues (Dirr 1976; Sinclair et al. 1987). Morphological factors such as bud size and the nature of scales affect salt uptake (Hofstra et al. 1979). Plants with resinous buds such as eastern cottonwood (*Populus deltoides* Bartr. ex Marshall) and common horsechestnut (*Aesculus hippocastanum* L.) or buds submerged in twigs of black locust (*Robinia pseudoacacia* L.)

and honeylocust (*Gleditsia triacanthos* L.) tend to preclude salt (Sinclair et al. 1987), whereas naked buds (lacking scales) of glossy buckthorn (*Rhamnus frangula* L.) are susceptible to salt spray (Lumis et al. 1973, 1975).

Norway maple (*Acer platanoides* L.), littleleaf linden (*Tilia cordata* Mill.), and wayfaringtree viburnum (*Viburnum lantana* L.) are common taxa in the U.S. urban landscape; therefore, so information about their salt tolerance would be useful. Norway maple [mature height of 12.2 to 18.3 m (40 to 60 ft)] and littleleaf linden [mature height of 15.2 to 21.3 m (50 to 70 ft)] are shade trees commonly used in street locations, parks, and residential and commercial landscapes. Wayfaringtree viburnum [mature height of 2.4 to 4.6 m (8 to 15 ft)] is a flowering ornamental shrub used for hedges, screens, and shrub borders. These species were chosen for their differences in bud morphology. Norway maple vegetative buds are small [3 to 6 mm (0.12 to 0.24 in) long] and have numerous, tightly arranged bud scales. Dissection of Norway maple buds reveals four outer, waxy bud scales and four or five fleshy, sticky, inner bud scales. Littleleaf linden buds are 6 to 9 mm (0.24 to 0.35 in) long and have four outer bud scales and three or four inner bud scales. Wayfaringtree viburnum vegetative buds are large [13 to 25 mm (0.52 to 1 in) long] and have a naked bud morphology (entirely lack bud scales).

The objective of this study was to quantify the concentrations of NaCl that produce injury on dormant buds of Norway maple, littleleaf linden, and wayfaringtree viburnum. Bud morphologies of each species were also examined for use as a potential selection criterion for salt spray tolerance.

## MATERIALS AND METHODS

### Plant Materials

Stem nodal samples containing lateral, vegetative buds were collected from Norway maple, littleleaf linden, and wayfaringtree viburnum. Samples were collected at McKay Nursery in Waterloo, Wisconsin, U.S. (43°11' N latitude, 88°59' W longitude). The collection sites were several hundred meters from a secondary road and thus were likely minimally affected by de-icing salts. One lateral branch node was taken per plant at each collection. A total of 162 plants were sampled at each collection period for electrolyte leakage and visual observation tests. In addition, eight branch samples from each species were placed in vases with tapwater, at ambient temperature, 21°C (70°F), for 16 hr per day for 3 weeks at all three collection periods, as a means of determining the stage of dormancy. The fluorescent light used provided a photosynthetic photon flux [PPF (400 to 700 nm)] of 24.1  $\mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$  (1.78 klx), as measured at the tops of the branch samples with a cosine-corrected LI-COR LI-189 quantum/radiometer/photometer (LI-COR, Lincoln, NE). Dormancy was estimated from the number of days to bud-

break, which was recorded when immature leaves became visible.

A factorial combination of three species (Norway maple, littleleaf linden, and wayfaringtree viburnum) and nine salt concentrations [0, 500, 1,000, 2,000, 4,000, 8,000, 16,000, 32,000, and 64,000 mg/L (0, 500, 1,000, 2,000, 4,000, 8,000, 16,000, 32,000, and 64,000 ppm)] of A.C.S.-certified crystalline NaCl was used. The experiment was repeated three times during 2001–2002 (4 December, 30 January, and 12 March). Temperature was recorded at a weather station in Madison, WI (43°8' N latitude, 89°20' W longitude) (Figure 1).

One centimeter (0.4 in) nodal samples were cut for Norway maple and littleleaf linden buds, and 2 cm (0.8 in) samples were cut for larger, wayfaringtree viburnum buds. Nodal samples were rinsed in deionized water for 30 sec, placed into 60 mL (1.8 oz) vials containing one of the nine NaCl solutions, and shaken on an Innova® 2100 platform shaker (New Brunswick Scientific Co., Inc., Edison, NJ) at 140 rpm for 24 hr at 4°C (39°F). Buds were left on stem nodes to reduce NaCl uptake via xylem and phloem tissues. Samples were removed from solutions and rinsed in deionized water for 1 min to remove exterior salinity. Bud injury was determined using electrolyte leakage and visual observation methods, similar to methods reported by Shirazi and Fuchigami (1993).

### Electrolyte Leakage

One bud was cut from each node and placed into individual 25 mL (0.75 oz) vials containing 8 mL (0.24 oz) deionized water (six buds per species/NaCl treatment were used). Vials were shaken for 20 hr at ambient temperature to facilitate electrolyte leakage from injured tissues. Initial electrical conductivity measurements were recorded for each vial using an Acromet AR20 electrical conductivity meter (Fisher Scientific, Chicago, IL). Vials were then immersed in a hot water

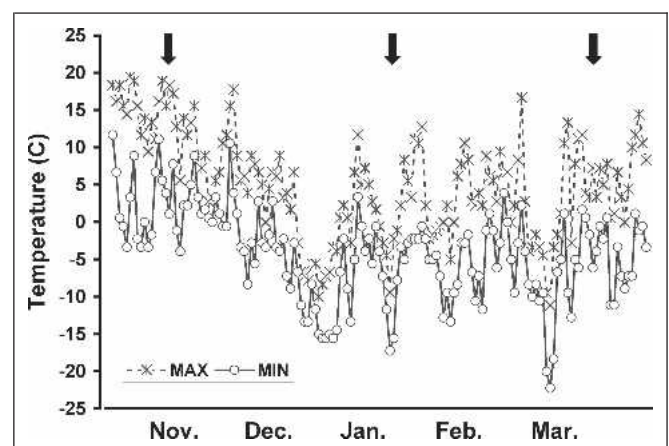


Figure 1. Daily maximum and minimum air temperatures from November 2001 to March 2002 in Madison, Wisconsin (43°8' N latitude, 89°20' W longitude). Arrows indicate collection dates.

bath (Fisher Isotemp, Indiana, PA) at 80°C (176°F) for 1 hr to induce cell rupture. The vials were again placed on the Innova 2100 platform shaker for 20 hr at 21°C (70°F), and final conductivity was measured for each vial. The percentage of electrolyte leakage for each bud was calculated as: (initial conductivity/final conductivity) × 100.

### Visual Discoloration

Following NaCl treatments, buds not used in the electrical conductivity test were placed in 15 cm (6 in) Petri dishes containing filter paper, moistened with deionized water, sealed with parafilm, and incubated in dark coolers at 21°C (70°F) for 10 to 12 days. One bud per node was then removed, cut longitudinally, and examined under a dissecting microscope for tissue discoloration (six buds per species/NaCl treatment). Buds were rated on outer tissue discoloration, inner (primordial) tissue discoloration, and fungal growth. Outer tissue of scaled buds was defined as the outer, lignified bud scales and immediately interior nonlignified bud tissue. Outer tissue for naked buds included the tomentose surface covering of primordial leaves and the outer edge of leaf primordia. Inner tissue was defined as any bud tissue located interior to outer bud tissue. Observations on discoloration were rated on a scale of 1 to 5 [1 = green/yellow tissue, 2 = detection of light-colored brown tissue (<10% of bud surface area), 3 = light-colored brown tissue (>10% of bud surface area), 4 = medium-brown tissue (100% of bud surface area), 5 = dark-brown to black tissue (100% of bud surface area)]. Fungal growth was rated on a scale of 1 to 3 [1 = no fungal growth, 2 = detection of fungal growth (5% to 10% of bud surface area), 3 = notable fungal growth (>10% of bud surface area)]. Three or four nodal sections from each species were cut longitudinally and examined for tissue discoloration. Although discoloration and fungal growth sometimes occurred on the nodal ends, little discoloration occurred near the bud attachment point, suggesting that minimal NaCl uptake had occurred through xylem and phloem tissues.

The experimental design within each sampling date was a completely randomized design with a factorial arrangement of treatments. Individual buds were the experimental units. Data were subjected to analysis of variance (ANOVA) procedures and regression analysis (SAS Institute Inc., Cary,

NC) (Table 1). Percentage data were also analyzed after arcsin transformation; results were similar to the untransformed data, so mean results presented are for untransformed data. Mean separations were performed by pairwise *t*-test comparisons at  $P \leq 0.05$ . Significant treatment effects for electrolyte leakage data identified by ANOVA procedures were subject to a log transformation to achieve a linear regression model for estimated NaCl concentrations corresponding to 50% electrolyte leakage (Figure 2).

## RESULTS AND DISCUSSION

### Electrolyte Leakage

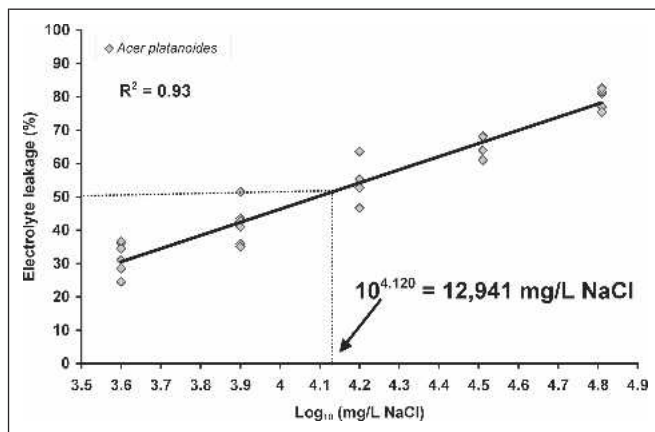
Measurements of electrolyte leakage from dead and injured cells provided a quantitative estimate of total bud NaCl injury. Electrolyte leakage from buds generally increased when NaCl concentrations increased and was dependent on the time of year (dormancy state) and the species (Figure 3). Sodium ions from NaCl displace  $\text{Ca}^{2+}$  ions in the plasma membrane of cells, possibly accounting for the increased electrolyte leakage (Taiz and Zeiger 1998).

### December Collection

Daily maximum temperatures preceding the December collection ranged from -1°C to 16°C (30°F to 61°F) (Figure 1). The buds were fully dormant (under endodormancy, Lang et al. 1987); no budbreak occurred in any of the cut branch samples. Average electrolyte leakage ranged from 28% in buds treated with deionized water to 68% in buds treated with 64,000 mg/L (64,000 ppm) NaCl (Figure 3A). Buds of Norway maple, littleleaf linden, and wayfaringtree viburnum had a significant quadratic response to NaCl ( $P \leq 0.0001$ ). A log transformation of significant electrolyte leakage data was conducted to achieve a linear regression model that estimated NaCl concentrations corresponding to 50% electrolyte leakage (Figure 2). Tissues having electrolyte leakage measurements greater than 50% are generally considered dead (Shirazi and Fuchigami 1993). However, because the relation between electrolyte leakage and bud survival could differ among species and tissue type (inner bud tissue or outer bud tissue), NaCl concentrations at 50% electrolyte leakage were regarded as severely injurious and potentially lethal but may not represent a lethal dose (LD).

**Table 1. Analysis of variance (ANOVA) for electrolyte leakage and inner and outer tissue discoloration for buds collected in December, January, and March.**

	Electrolyte leakage			Inner tissue discoloration			Outer tissue discoloration		
	4 Dec. 2001	30 Jan. 2002	12 March 2002	4 Dec. 2001	30 Jan. 2002	12 March 2002	4 Dec. 2001	30 Jan. 2002	12 March 2002
Species	<0.0001	<0.0001	<0.0001	0.051	<0.0001	<0.0001	0.101	<0.0001	<0.0001
NaCl	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Species × NaCl	0.001	0.002	0.881	0.001	0.445	<0.0001	0.002	0.009	<0.0001

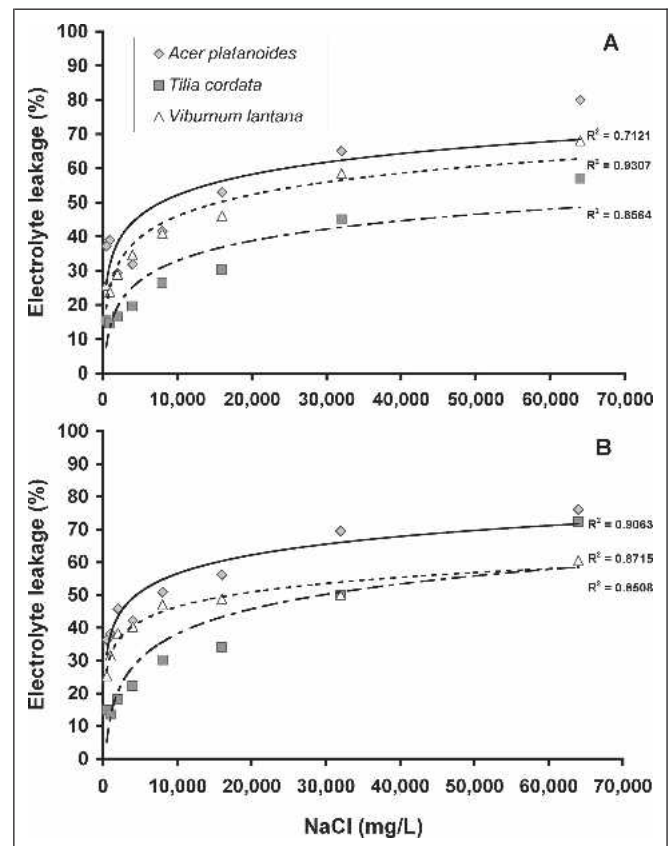


**Figure 2.** Determination of NaCl concentration corresponding to 50% electrolyte leakage for *Acer platanoides* buds collected in December 2001. Equation for the NaCl response is  $Y = 39.5x - 111.7$ , where  $Y$  = electrolyte leakage (%) and  $x = \text{Log}_{10}$  of NaCl concentration.

The interaction between species and NaCl concentration was significant in December ( $P \leq 0.001$ ) (Table 1 and Figure 3A). Buds of Norway maple were the most susceptible to NaCl-induced electrolyte leakage, experiencing 50% electrolyte leakage at 12,941 mg/L (12,941 ppm), followed by wayfaringtree viburnum buds at 16,901 mg/L (16,901 ppm) and littleleaf linden buds at 42,594 mg/L (42,594 ppm) NaCl (Table 2). Buttle and Labadia (1999) reported 6,506 mg/L (6,506 ppm) Na and 9,916 mg/L (9,916 ppm) Cl [equivalent to 10,725 and 25,206 mg/L (10,725 and 25,206 ppm) NaCl, respectively] in snow sampled from highway medians. Therefore, concentrations of NaCl that induced 50% electrolyte leakage (severe injury) in buds of wayfaringtree viburnum and Norway maple were within the range of those found in roadside snow. Although no reports of NaCl concentrations more than 25,206 mg/L (25,206 ppm) were found, higher NaCl concentrations are probable in snow, slush, and melt-water found directly on road surfaces, especially in locations requiring high salt applications (i.e., intersections, bridges, and highway ramps).

### January Collection

Daily maximum temperatures preceding the January collection ranged from  $-5^{\circ}\text{C}$  to  $13^{\circ}\text{C}$  ( $23^{\circ}\text{F}$  to  $55^{\circ}\text{F}$ ) (Figure 1). The buds were fully dormant; no budbreak occurred in any of the cut branch samples. Average electrolyte leakage ranged from 23% in buds treated with deionized water to 71% in buds treated with 64,000 mg/L (64,000 ppm) NaCl. The interaction between species and NaCl was significant for January-collected buds ( $P \leq 0.01$ ) (Table 1 and Figure 3B). Buds of Norway maple, littleleaf linden, and wayfaringtree viburnum again had a significant quadratic response to NaCl ( $P \leq 0.0001$ ). Species rankings for bud electrolyte leakage in re-



**Figure 3.** Effect of NaCl and species on electrolyte leakage of buds collected in December 2001 (A) and January 2002 (B). Standard error of the treatment mean difference was 2.81 for December 2001 and 5.18 for January 2002.

sponse to NaCl were similar to those of December-collected buds, although 50% electrolyte leakage of littleleaf linden and Norway maple buds occurred at lower NaCl concentrations. Norway maple buds suffered 50% electrolyte leakage at 7,165 mg/L (7,165 ppm), followed by wayfaringtree viburnum at 18,804 mg/L (18,804 ppm) and littleleaf linden at 27,118 mg/L (27,118 ppm) NaCl (Table 2). The reason for the decreased NaCl tolerance in littleleaf linden and Norway maple buds is uncertain. It is possible that the endogenous seasonal rhythm for cold hardiness (Weiser 1970; Harrison et al. 1978) affected the salt tolerance of buds. Numerous woody species exhibit greatest resistance to cold injury between mid-December and early January (Kang et al. 1997; McNamara et al. 2002). Slight midwinter changes in metabolism, tissue water content, or bud morphology (Ashworth and Wisniewski 1991; Rinne et al. 1994) may have occurred, although undetected, thus altering the buds' NaCl tolerance.

### March Collection

Maximum daily temperatures preceding the March collection ranged from  $-18^{\circ}\text{C}$  to  $13^{\circ}\text{C}$  ( $0^{\circ}\text{F}$  to  $55^{\circ}\text{F}$ ) (Figure 1). Budbreak

**Table 2. Concentrations of NaCl corresponding to 50% electrolyte leakage in dormant vegetative buds.**

Month <sup>z</sup>	Species	NaCl (mg/L)	±SE
December	<i>Acer platanoides</i>	12,941 <sup>x,y</sup>	±406 <sup>w</sup>
	<i>Tilia cordata</i>	42,594 <sup>v</sup>	±9,488
	<i>Viburnum lantana</i>	16,901	±2,064
January	<i>Acer platanoides</i>	7,165	±2,973
	<i>Tilia cordata</i>	27,118	±4,133
	<i>Viburnum lantana</i>	18,804	±7,314

<sup>z</sup>March data are not shown (nonsignificant).

<sup>x</sup>Log<sub>10</sub> transformation of quadratic data were conducted in order to fit a linear model.

<sup>y</sup>Represents the mean of values obtained from six linear models.

<sup>w</sup>Calculated as lower confidence interval subtracted from upper confidence interval, divided by two.

<sup>v</sup>Two observations were not used because electrolyte leakage did not reach 50%.

occurred in the branch samples of all species within 2 weeks of collection, indicating the buds were under ecodormancy (Lang et al. 1987). No significant interaction between species and NaCl concentration occurred (Table 1). Average electrolyte leakage ranged from 33% in buds treated with deionized water to 79% in buds treated with 64,000 mg/L (64,000 ppm) NaCl (data not shown). Main effects of NaCl and species were both significant ( $P \leq 0.0001$ ). Differences in bud electrolyte leakage between species were insignificant at concentrations of less than 8,000 mg/L (8,000 ppm) NaCl, but species differences occurred at concentrations of 8,000 mg/L (8,000 ppm) NaCl and greater (data not shown). Wayfaring-tree viburnum buds had the highest average electrolyte leakage (53.2%), followed by buds of Norway maple (47.3%) and littleleaf linden (31.1%) when averaged over all NaCl concentrations. This is in contrast to the other two collection periods, when Norway maple buds exhibited the highest electrolyte leakage, followed by wayfaringtree viburnum and littleleaf linden buds, respectively. It appeared that increased physiological and morphological activity related to spring deacclimation decreased the buds' NaCl resistance, especially in wayfaringtree viburnum buds. Cold temperatures in March 2002 may have also contributed to injury in buds of all species, possibly accounting for the increased electrolyte leakage in control buds (Figure 1).

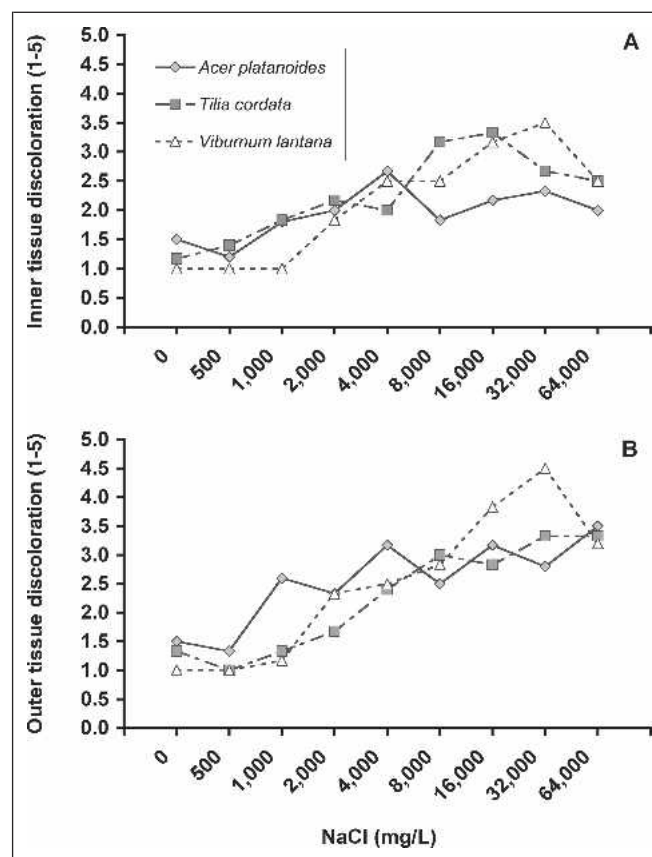
**Visual Discoloration**

Visual observations were conducted to determine whether electrolyte leakage was occurring from outer or inner bud tissue. Injury to inner bud tissue was considered more important than injury to scales and outer bud tissue because primordial tissue was affected. Injured tissue turned brown to black after the 10 to 12 day incubation period. Bud tissues with a rating of 3 (light-colored browning >10% of area) were considered to be moderately injured, and bud tissues with a

rating of 4 (medium-brown tissue in 100% of area) or 5 (dark-brown to black tissue in 100% of area) were considered to be severely injured. In general, outer tissue discoloration was more severe than inner tissue discoloration. Fungal growth occurred on buds of all three species at the 64,000 mg/L (64,000 ppm) NaCl treatment but was inconsistent at lower NaCl concentrations (data not shown). Buds with severe tissue discoloration and/or fungal growth often had a water-soaked appearance.

**December Collection**

Interactions between species and NaCl concentration for inner ( $P \leq 0.001$ ) and outer ( $P \leq 0.01$ ) tissue discoloration were significant in December (Table 1 and Figure 4). Buds from all species exhibited some tolerance to NaCl because no



**Figure 4. Effect of NaCl and species on inner (A) and outer (B) tissue discoloration of buds collected in December 2001. Standard error of the treatment mean difference was 0.37 for inner tissue discoloration and 0.45 for outer tissue discoloration. Observations were rated on a scale of 1 to 5 (1 = green/yellow tissue, 2 = detection of light-colored brown tissue (<10%), 3 = light-colored brown tissue (>10%), 4 = medium-brown tissue (100%), 5 = dark-brown to black tissue (100%)).**

severe inner tissue discoloration occurred (Figure 4A). Inner tissue injury for Norway maple buds remained low for all NaCl treatments; they had significantly less inner tissue discoloration than littleleaf linden and wayfaringtree viburnum buds at 16,000 mg/L (16,000 ppm) NaCl, and wayfaringtree viburnum buds at 32,000 mg/L (32,000 ppm) NaCl. Severe outer tissue discoloration was detected only in wayfaringtree viburnum buds at 32,000 mg/L (32,000 ppm) NaCl (Figure 4B). Severe outer tissue discoloration in wayfaringtree viburnum buds is probable at greater NaCl concentrations, although it was not detected at 64,000 mg/L (64,000 ppm) NaCl.

Patterns of tissue discoloration reflected the unique bud morphology of each species. The numerous, tightly arranged waxy bud scales of Norway maple may have inhibited NaCl penetration to inner bud tissues, whereas the tomentose surface of naked wayfaringtree viburnum buds offered little protection. Lumis et al. (1973) similarly reported that naked buds of glossy buckthorn (*Rhamnus frangula* L.) were more injured by salt spray than common buckthorn (*Rhamnus catharticus* L.), a scaled species. In addition to the presence of bud scales, the number and arrangement of bud scales may be a factor in the salt tolerance of buds. Littleleaf linden buds have slightly fewer bud scales than Norway maple buds, possibly accounting for the occurrence of moderate inner tissue injury. A bud's ability to preclude salt from penetrating and accumulating in sensitive tissues relates to the bud's salt tolerance (Dirr 1976; Sinclair et al. 1987).

### January Collection

Detection of severe inner and outer tissue discoloration indicated that buds from all species became more susceptible to NaCl injury in January. The interaction between species and NaCl was not significant for inner tissue discoloration (Table 1). All species exhibited a similar pattern of inner tissue injury in response to NaCl; the pattern plateaued at 1,000 mg/L (1,000 ppm) NaCl (data not shown). The differences in the severity of inner tissue discoloration among species were evident ( $P \leq 0.0001$ ) (Tables 1 and 3), indicating a relation

**Table 3. Effect of species on inner tissue discoloration of buds collected in January 2002 combined over nine NaCl concentrations.**

Species	Inner tissue discoloration rating <sup>z,x,y</sup>
<i>Acer platanoides</i>	2.9 a
<i>Tilia cordata</i>	3.6 b
<i>Viburnum lantana</i>	4.2 c

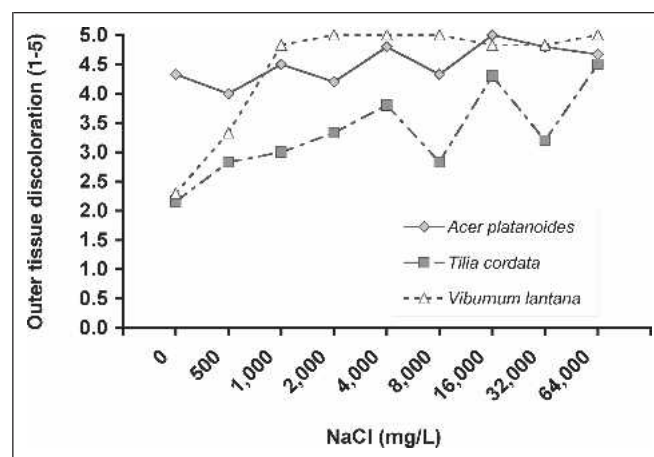
<sup>z</sup>Represents the mean of 54 buds.

<sup>x</sup>Rating scale: 1 to 5 [1 = green/yellow tissue, 2 = detection of light-colored brown tissue (<10%), 3 = light-colored brown tissue (>10%), 4 = medium-brown tissue (100%), 5 = dark-brown to black tissue (100%)].

<sup>y</sup>Mean separation within column by pairwise *t*-tests,  $P \leq 0.05$ .

between bud morphology and inner tissue injury. Wayfaringtree viburnum, which has naked buds, displayed severe inner tissue discoloration, whereas littleleaf linden had moderate discoloration and Norway maple had low inner tissue discoloration. Both littleleaf linden and Norway maple have multiple bud scales; however, the inner bud scales of Norway maple are sticky and contain milky sap, perhaps precluding salt from entering into primordial tissue.

The interaction between species and NaCl was significant for outer tissue discoloration ( $P \leq 0.01$ ) for January-collected buds (Table 1 and Figure 5). Species rankings for outer tissue injury differed from those of inner tissue injury. Although Norway maple buds had low to moderate inner tissue discoloration, severe outer tissue discoloration occurred at all NaCl treatments. Outer bud tissues of wayfaringtree viburnum were also sensitive to NaCl: moderate injury occurred at 500 mg/L (500 ppm) NaCl and severe injury occurred at a concentration as low as 1,000 mg/L (1,000 ppm) NaCl. Littleleaf linden buds had the lowest outer tissue discoloration of the three species, exhibiting moderate injury at 2,000 mg/L (2,000 ppm) NaCl and severe injury at 16,000 mg/L (16,000 ppm) NaCl. Bud scale surfaces of littleleaf linden have a glossy sheen, indicating the presence of protective cuticular wax. Although there is little research about the relation between cuticular wax and salt spray tolerance of buds, research on needles from numerous species of pine (*Pinus* spp. L.) and spruce (*Picea* spp. A. Dietr.) has reported surface wax to be an effective barrier to salt spray (Townsend and Kwolek 1987; Dobson 1991).



**Figure 5. Effect of NaCl and species on outer tissue discoloration of buds collected in January 2002. Standard error of the treatment mean difference was 0.50. Observations were rated on a scale of 1 to 5 (1 = green/yellow tissue, 2 = detection of light-colored brown tissue (<10%), 3 = light-colored brown tissue (>10%), 4 = medium-brown tissue (100%), 5 = dark-brown to black tissue (100%)). Inner tissue discoloration was nonsignificant.**

### March Collection

Buds from all species were most susceptible to NaCl injury in March because inner and outer tissue discoloration occurred at lower NaCl concentrations than in January (Figures 5 and 6). Species differences in inner tissue discoloration were significant in response to NaCl ( $P \leq 0.0001$ ; Table 1 and Figure 6A). Severe inner tissue discoloration occurred at 500 mg/L (500 ppm) NaCl in buds of wayfaringtree viburnum. Littleleaf linden buds showed moderate injury at the deionized water treatment and severe injury at 1,000 mg/L (1,000 ppm) NaCl. Possible freezing injury before collection in January and March in sampled buds may account for observations of severe injury at the deionized water treatment (Figure 1). Equivalent injury in Norway maple buds occurred at higher NaCl concentrations. Inner tissues of Norway maple buds

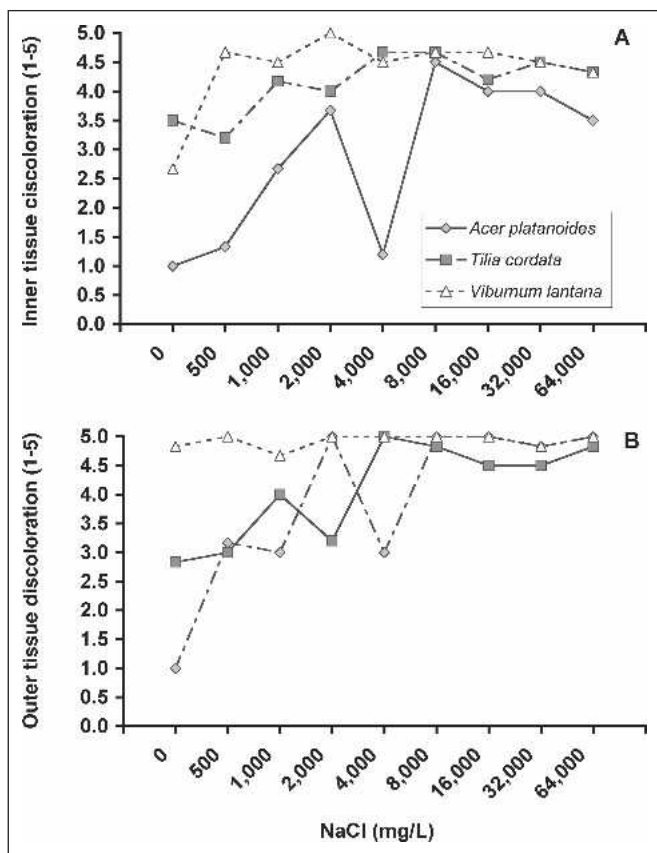


Figure 6. Effect of NaCl and species on inner (A) and outer (B) tissue discoloration of buds collected in March 2002. Standard error of the treatment mean difference was 0.52 for inner tissue discoloration and 0.42 for outer tissue discoloration. Observations were rated on a scale of 1 to 5 (1 = green/yellow tissue, 2 = detection of light-colored brown tissue (<10%), 3 = light-colored brown tissue (>10%), 4 = medium-brown tissue (100%), 5 = dark-brown to black tissue (100%)).

exhibited moderate injury at 2,000 mg/L (2,000 ppm) NaCl and severe injury at 8,000 mg/L (8,000 ppm) NaCl. An unexplainable sudden drop in inner tissue discoloration in Norway maple buds occurred at 4,000 mg/L (4,000 ppm) NaCl. However, salt levels of 4,000 mg/L (4,000 ppm) NaCl are likely to be injurious, as moderate discoloration occurred at a lower NaCl concentration.

The interaction between species and NaCl was also significant for outer tissue discoloration ( $P \leq 0.0001$ ) (Table 1 and Figure 6B). Wayfaringtree viburnum buds suffered severe outer tissue discoloration in response to all NaCl treatments. Equivalent injury occurred at higher NaCl concentrations in scaled buds. Norway maple and littleleaf linden buds showed moderate outer tissue injury at 500 mg/L (500 ppm) NaCl. Severe outer tissue injury occurred at 1,000 mg/L (1,000 ppm) NaCl in littleleaf linden buds and at 2,000 mg/L (2,000 ppm) NaCl in Norway maple buds.

In addition to time of year (dormancy state) and NaCl concentration, NaCl injury depends on the type of evaluation method (i.e., visual tissue discoloration or electrolyte leakage). Combining electrolyte leakage and visual observation tests provides a more detailed representation of NaCl injury than measuring only one variable. Electrolyte leakage tests provided estimations of total bud injury but did not account for the location of injury within buds (Calkins and Swanson 1990). Inner bud tissues contain leaf primordia and meristematic tissue that generate new stems, foliage, lateral buds, and reproductive structures (Taiz and Zeiger 1998). Thus, inner tissue injury could severely inhibit new growth in spring. Simini and Leone (1986) reported that budbreak in Norway maples was not affected by a salt spray concentration of 40,000 mg/L (40,000 ppm) Cl or 65,900 mg/L (65,900 ppm) NaCl. Although Norway maple buds suffered substantial electrolyte leakage at NaCl concentrations considerably lower than 65,900 mg/L (65,900 ppm) NaCl, visual observations showed that the majority of injury occurred in bud scales and outer tissue. Higher inner tissue discoloration in buds of littleleaf linden and wayfaringtree viburnum may be an indication that their buds were less effective at precluding NaCl penetration. Inner tissue injury in these species may impede new growth in spring.

### CONCLUSIONS

Results of this experiment imply that buds with numerous bud scales have increased primordial tissue protection against salt spray with lower internal tissue discoloration. However, these results should be interpreted with caution. Bud morphology should be considered when testing species for salt tolerance, but a plant's response to salt can be influenced by many factors, including genetic differences, type of exposure (soil salt or salt spray), exposure intensity, biotic or abiotic, and climatological/seasonal factors. The bud's salt tolerance

may or may not reflect the whole plant's tolerance of a given species.

Increased use of deicing salt is expected to continue in the United States due to continuing public safety demands, and as urbanization and roadways continue to spread. As a result, more plants will suffer exposure to salt spray. The following are recommendations for reducing salt spray damage:

- Reduce or avoid salt applications in early spring, if possible, as damage to plants is most severe in spring. Abrasives such as sand and crushed rock can substitute for a portion of the salt.
- Rinse above-ground plant parts after salt spray exposure in early spring.
- Use caution when planting species with naked buds and other salt-sensitive species adjacent to high-speed thoroughfares and in street planters, medians, parking lot landscapes, and other areas receiving exposure to salt spray.

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**Résumé.** Des bourgeons latéraux dormants d'érable de Norvège (*Acer platanoides* L.), de tilleul à petites feuilles (*Tilia cordata* Mill.), et de viorne commune (*Viburnum lantana* L.) ont été récoltés et soumis à neuf concentrations de NaCl: 0, 500, 1,000, 2,000, 4,000, 8,000, 16,000, 32,000, et 64,000 mg/L (0, 500, 1,000, 2,000, 4,000, 8,000, 16,000, 32,000, et 64,000 ppm) en décembre 2001 ainsi qu'en janvier et mars 2002. La perte en électrolytes ainsi que des observations visuelles de la décoloration des tissus interne et externe ont été utilisés pour déterminer le degré de dommages. Les dommages au bourgeon s'accroissaient généralement avec l'augmentation de la concentration en NaCl. La morphologie des bourgeons de chacune des espèces était associée à des patrons déterminés de décoloration des tissus; les bourgeons nus étaient plus susceptibles au NaCl que ceux avec des écailles. Les bourgeons présentaient aussi des résistances variables selon les périodes de l'année au NaCl; la résistance la plus élevée se produisait en décembre. L'érable de Norvège, le tilleul à petites feuilles et la viorne commune ont présenté une perte de 50% d'électrolytes à des valeurs calculées de NaCl de 12,941, 16,901, et 42,594 mg/L (12,941, 16,901, et 42,594 ppm) de NaCl respectivement; cependant, aucune décoloration sévère du tissu interne ne s'est produite avec aucun de ces traitements de NaCl. En janvier, une perte de 50% d'électrolytes s'est produite à des concentrations plus faibles de NaCl chez l'érable

de Norvège (7,165 mg/L ou ppm) et le tilleul à petites feuilles (27,118 mg/L ou ppm). Des dommages modérés à sévères du tissu interne ont été détectés chez toutes les espèces à 1,000 mg/L (1,000 ppm) de NaCl. Les bourgeons étaient plus susceptibles à des dommages par le NaCl en mars avec une décoloration modérée à sévère du tissu interne qui se présentait chez la viorne commune et le tilleul à petites feuilles à 500 mg/L (500 ppm) de NaCl.

**Zusammenfassung.** Im Dezember 2001 und Januar, März 2002 sammelten wir schlafende Knospen von (*Acer platanoides* L.), (*Tilia cordata* Mill.) und (*Viburnum lantana* L.) und setzen ihnen 9 verschiedene NaCl-Konzentrationen zu. Die elektrolytische Leckage und die Beobachtungen der inneren und äußeren Gewebeverfärbungen wurden verwendet, um Verletzungen zu untersuchen. Die Knospenverletzungen stiegen mit der Erhöhung der NaCl-Konzentration an. Die Knospenmorphologie jeder Baumart wurde mit den Gewebeverfärbungen in Verbindung gebracht, nackte Knospen waren reagierten stärker auf NaCl als bedeckte Knospen. Die Knospen zeigten auch saisonalen Widerstand, am meisten im Dezember. Der Austritt von Elektrolyten betrug 50% bei kalkulierten NaCl-Werten von 12.941, 16.901, und 42.594 mg/L. dennoch gab es keine ernste Verfärbung der inneren Gewebe. Im Januar 2002 tauchte ein 50% Elektrolytaustritt bei niedrigeren NaCl-Werten bei den Ahornen und Linden auf. Mittlere bis schwere innere Gewebeverletzungen traten bei allen Arten bei 1,000 mg/L NaCl auf. Im März waren die Knospen der Linden und Viburnum sehr verletzungsanfällig mit mittleren bis schweren Verfärbungen der inneren Gewebe bei einer Konzentration von 500 mg/L NaCl.

**Resumen.** Se colectaron yemas laterales de maple Norway (*Acer platanoides* L.), tilo (*Tilia cordata* Mill.), y viburno (*Viburnum lantana* L.) y se expusieron a nueve concentraciones de NaCl: 0, 500, 1,000, 2,000, 4,000, 8,000, 16,000, 32,000, y 64,000 mg/L (0, 500, 1,000, 2,000, 4,000, 8,000, 16,000, 32,000, y 64,000 ppm), en Diciembre de 2001, Enero y Marzo de 2002. Para medir el daño se utilizaron observaciones visuales y medición de electrolitos tanto exterior como interna de la decoloración de los tejidos. El daño a las yemas generalmente aumentó a medida que la concentración de NaCl incrementó. Las morfologías de las yemas de cada especie estuvieron relacionadas a los patrones de decoloración de los tejidos; las yemas desnudas fueron más susceptibles a NaCl que aquellas con escamas. Las yemas también mostraron resistencia estacional a la NaCl; la mayor resistencia ocurrió en Diciembre. Las yemas de maple, viburno y tilo experimentaron pérdida de electrolitos del 50%, a valores calculados de NaCl de 12,941, 16,901, y 42,594 mg/L (12,941, 16,901, y 42,594 ppm) de NaCl, respectivamente; sin embargo, no ocurrió decoloración severa a cualquier tratamiento de NaCl. En Enero, ocurrió el 50% de pérdida de electrolitos a concentraciones bajas de NaCl en yemas de maple [7,165 mg/L (7,165 ppm)] y tilo [27,118 mg/L (27,118 ppm)]. Se detectó moderado a severo daño en los tejidos para todas las especies a 1,000 mg/L (1,000 ppm) de NaCl. Las yemas fueron más susceptibles a daño por NaCl en Marzo con daño moderado a severo, con decoloración de los tejidos internos en viburno y tilo a 500 mg/L (500 ppm) de NaCl.