Growth of Container-grown Pin Oak and Japanese Maple as Influenced by Sulfur and Sulfated Micronutrients

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Abstract. Substrates of container-grown plants are commonly preplant amended with sulfated micronutrients to supply micronutrients. However, the cause for the increased growth may be due to micronutrient addition or other factors such as S addition or substrate acidification. Container-grown pin oak (*Quercus palustris* Muench) and Japanese maple (*Acer palmatum* Thunb.) seedlings were grown in a 100% pine bark substrate and amended (or not) with one of the following treatments: control (no amendment), Micromax, K₂SO₄, H₂SO₄, HCl, chelated micronutrients, elemental S, or CaSO₄. After 11 weeks, dry weights of plants in all treatments supplying S were higher than plants receiving no S. Dry weights of plants in all experiments receiving the chelate treatment were not higher than dry weights for control plants. These data indicate that S, not micronutrient application, is a primary cause of increased growth from the addition of sulfated micronutrients. However, it was demonstrated that there are conditions such as higher substrate solution pH (4.1 vs. 5.4), where Micromax may prove advantageous over sulfur alone since it would supply micronutrients as well as S.

Substrates of container-grown plants are commonly preplant amended with sulfated micronutrients with the intention of increasing the supply of micronutrients to the plant. Wright et al. (1999a) showed that seedling growth of nine container-grown tree species in pine bark was positively affected by the addition of Micromax (Scotts-Sierra, Marysville, Ohio), a sulfated micronutrient fertilizer package. However, Kelk (2002), investigating the influence of CuSO₄, FeSO₄, MnSO₄, and ZnSO₄ on the growth of *Quercus palustris* (Muench), found that growth was increased to the same degree as Micromax by amending pine bark with any three of the four sulfated micronutrients or by any one sulfated micronutrient in the absence of the other three. Growth of these treatments (supplying three of the four sulfated micronutrients or one of the four sulfated micronutrients) was 200% greater than plants without sulfated micronutrient addition. This raises the question of whether micronutrients are actually the cause for increased growth when sulfated micronutrients are added to pine bark substrates. Both Wright et al. (1999b) and Kelk (2002) found that preplant amending pine bark with sulfated micronutrients decreased substrate pH by about 0.3 units. This

decreased pH is most likely due to the release of H⁺ during the hydrolysis of the metallic micronutrient cations (Brady and Weil, 2004). Thus, plant response to sulfated micronutrient addition may be due to the increased supply of micronutrients, the decrease in substrate pH, or both of these factors. A decrease in substrate pH may increase micronutrient cation supply due to the conversion of insoluble micronutrient hydroxides to soluble metal cations inherent in the bark (Brady and Weil, 2004). Another possibility is that the growth increase may be due, at least in part, to the addition of sulfur (S), as sulfate, associated with the micronutrients. Sulfur is essential to the growth of higher plants especially since it is a major component of the amino acids cysteine and methionine (Leustek et al., 2000; Mengel and Kirkby, 2001). Several studies have shown a need for S additions to maximize plant growth. For example, peach (*Prunus persica* L. Batsch) grown in sand culture required 4 mg·L⁻¹ S (Finch et al., 1997); sugar beet (*Beta vulgaris* L.) grown in solution culture required 32 mg·L⁻¹ S (Kastori et al., 2000); chrysanthemum (*Dendranthema grandiflora*) required 8 mg·L⁻¹ S (Huang et al., 1997) in solution culture and 10 mg·L⁻¹ S (Macz et al., 2001) when container-grown in a peat-based substrate; and finally, container-grown stock (*Mathiola incana* 'Austral') and cabbage (*Brassica oleracea* 'Lion Heart') in a pine bark substrate required 25 and 27 mg·L⁻¹ S, respectively (Handreck, 1986). In general, Nelson (1996) states "that most greenhouse crops require at least 16 mg·L⁻¹ S or greater in irrigation water." Thus, the objective of this work was to determine which factors, or combination of factors, improves seedling growth of container-grown pin oak (*Quercus*

palustris) and Japanese maple (*Acer palmatum*) in a pine bark substrate.

Materials and Methods

Experiment 1. In August 2003, stratified pin oak (*Quercus palustris* Muench) (Sheffield's Seed Company, Inc., Locke, N.Y.) seeds were sown (3 cm deep) in flats of unamended pine bark (PB). Two weeks after planting, seeds germinated. On 9 Sept. 2003, three (2.5 cm tall) seedlings were transplanted per 3.8-L (#1) plastic container into milled pine bark (PB) (*Pinus taeda* L.) substrate. Treatments were 1) unamended PB, 2) PB preplant amended with a commercial sulfated micronutrient fertilizer (Micromax), [O.M. Scott, Marysville, Ohio; 12% S, 0.1% B (Na₂B₄O₇), 0.5% Cu (CuSO₄), 12% Fe (FeSO₄), 2.5% Mn (MnSO₄), 0.05% Mo (Na₂MoO₄), and 1% Zn (ZnSO₄)] at the rate of 0.9 kg·m⁻³, 3) micronutrient chelate solution supplied by Peters Professional Compound 111 (Scotts-Sierra) (0.75% Mg (MgSO₄), 0.232% B (H₃BO₃), 0.114% Cu (Cu EDTA), 1.5% Fe (Fe EDTA), 0.75% Mn (Mn EDTA), 0.24% Mo (H₂₄Mo₇N₆O₂₄), and 0.075% Zn (Zn EDTA)) applied to PB (postplant) at a concentration supplying 1 mg·L⁻¹ Fe at each fertilization, 4) 0.011 N H₂SO₄ solution applied at each fertilization, 5) 0.012 N HCl solution applied at each fertilization, and 6) 100 mg·L⁻¹ S as K₂SO₄ applied at each fertilization. Irrigation frequency was based on plant need for water by lifting containers and assessing container weight. Seedlings were fertilized as needed to maintain an electrical conductivity (EC) of 1.0 to 1.5 dS·m⁻¹ in the substrate solution. Seedlings were fertilized with 250-mL of nutrient solution of 300 mg·L⁻¹ N (NH₄NO₃), 45 mg·L⁻¹ P (H₃PO₄), and 250 mg·L⁻¹ K (KCl). The K₂SO₄ treatment received 248 mg·L⁻¹ K as K₂SO₄ instead of KCl. Alkalinity, Ca, and Mg concentrations of irrigation water were 36, 10.2, and 4.2 mg·L⁻¹, respectively. Calcium and Mg supply using the pour-through (PT) method (Yeager et al., 1983) from PB and irrigation water (same PB lot used in this study) were 42 and 39 mg·L⁻¹, respectively (data not shown). Irrigation water micronutrient concentrations (mg·L⁻¹) were 0 Fe, 0 Mn, 0.04 Zn, and 0.002 Cu.

Pine bark solution was periodically extracted from containers (PT extractions) and analyzed for pH and EC to gauge the frequency of fertilizer application. Pine bark solutions were extracted (PT) on 1 Oct. 2003 and analyzed for pH, S, Fe, Cu, Mn, and Zn; ion concentrations were determined by inductively coupled plasma (ICP) analysis. Plants were grown on raised benches in the Virginia Tech (Blacksburg, Va.) Greenhouse Facility with an average daytime temperature of 24 °C and nighttime temperature of 21 °C. On 7 Nov. 2003 plant stems were severed at the soil surface. Shoots were dried for about 3 d at 65 °C and dry weights were recorded. The experimental design was completely randomized with four single container replications per treatment. All data were analyzed using SAS (version 8.02) PROC GLM.

Experiment 2. In February 2004, a second

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Fig. 1. Mean dry weight of container-grown pin oak seedlings (Expt. 1) as affected by various treatments. Means are results of three subsamples per container ($n = 4$). Data were collected from 3 seedlings per container. Means were separated using Duncan's multiple range test ($P \leq 0.05$). Treatments with the same letters are not significantly different.

Table 1. Substrate solution pH, S, Fe, Cu, Mn, and Zn concentrations for container-grown pin oak seedlings as affected by various treatments (Expt. 1). Means are results of three subsamples per container.

Treatment	pH	Element (mg·L ⁻¹)				
		S	Cu	Fe	Mn	Zn
Pin oak						
Control	4.13 b ^a	2.0 d	0.003 bc	0.07 c	0.74 cd	0.10 c
Micromax	4.15 ab	72.0 a	0.007 a	0.08 c	2.13 a	0.48 a
K ₂ SO ₄	4.3 a	40.7 c	0.002 c	0.10 b	0.41 d	0.13 bc
H ₂ SO ₄	4.1 b	52.8 b	0.002 c	0.08 bc	1.08 c	0.17 bc
HCl	4.1 b	2.4 d	0.003 bc	0.05 c	1.48 b	0.18 b
Chelate	4.15 ab	2.3 d	0.005 b	0.39 a	0.73 cd	0.15 bc

^aMeans separation in columns by Duncan's multiple range test, $P \leq 0.05$ ($n = 4$). Treatments within columns with the same letters are not significantly different.

experiment was conducted using pin oak (*Quercus palustris* Muench). Stratified seeds (Sheffield's Seed Co.) were sown (3 cm deep) in flats of unamended pine bark (PB). Two weeks after planting, seeds germinated. On 25 Feb. 2004, three (2.5 cm tall) seedlings were transplanted per 3.8-L (#1) plastic container into PB only (*Pinus taeda* L.) substrate. Treatments began 2 d later and were the same as Expt. 1 with the exception of two added treatments. These treatments were 1) PB preplant amended with 0.45 kg·m⁻³ of elemental S, and 2) PB preplant amended with 0.45 kg·m⁻³ of CaSO₄. Pine bark solutions were extracted (PT) on 24 Mar. 2004 and analyzed for pH, S, Fe, Cu, Mn, and Zn as discussed above. On 10 May 2004 plant stems were severed at the soil surface. Shoots were dried for about 3 d at 65 °C and dry weights were recorded. On 27 May 2004, dried shoot tissue was ground in a Cyclone Sample Mill (UD Corp., Boulder, Colo.) and analyzed for S, Cu, Fe, Mn, and Zn concentrations by ICP spectrometry (A & L Eastern Agricultural Laboratories, Richmond, Va.). The experimental design was completely randomized with six single-container replications per treatment. All data were analyzed using SAS (version 8.02) PROC GLM.

Experiment 3. In March 2004, a third experiment was conducted using Japanese maple (*Acer palmatum* Thunb.). Stratified seeds (Sheffield's Seed Co.) were sown (1 cm deep) in flats of unamended PB. On 24 Mar. 2004, three (2.5 cm tall) seedlings were transplanted per 3.8-L (#1) plastic container into PB only and treatments began. Treatments were the same as Expt. 2. Pine bark solutions were extracted (PT) on 17 May 2004 and analyzed for pH, S,

Fe, Cu, Mn, and Zn as discussed above. On 17 May 2004, plant stems were severed at the soil surface. Shoots were dried for about 3 d at 65 °C and dry weights were recorded. On 27 May 2004, dried shoot tissue was ground and analyzed for S, Fe, Cu, Mn, and Zn as discussed above. The experimental design was completely randomized with six single-container replications per treatment. All data were analyzed using SAS (version 8.02) PROC GLM.

Results and Discussion

Dry weights of pin oak (Expt. 1) were highest for the Micromax amended PB treatment and the H₂SO₄ treatment (Fig. 1). Dry weights for plants supplied with S and no micronutrients (K₂SO₄, H₂SO₄ treatments) were higher than those in the control treatment. Dry weights for plants supplied with Micromax were also higher than that for the chelate

treatment, which suggests the necessity of S supplied by Micromax. For treatments that acidified the substrate (H₂SO₄ and HCl), mean dry weight of the H₂SO₄ treatment was nearly twice as large as the HCl treatment for oak, thus demonstrating the benefit of S (Fig. 1).

Except for Fe, micronutrient solution concentrations for oak (Expt. 1) were generally higher for Micromax than for all other treatments (Table 1). Substrate solution S concentrations for treatments (Expt. 1) with the highest dry weights (Micromax, K₂SO₄, H₂SO₄) were at least 17 times higher than treatments that did not supply S (control, HCl, chelate) (Table 1) and were above the recommended concentrations for a variety of crops (Handreck, 1986; Nelson, 1996). The substrate solution pH of the two acid treatments (H₂SO₄ and HCl) was the same as that of the control treatment (Table 1), but only the dry weight of the H₂SO₄ treatment was higher than the control treatment (Expt. 1) (Fig. 1). These data also demonstrate the benefit of S addition. This experiment was conducted two other times and gave essentially the same results (data not shown).

Dry weights of plants in the Micromax amended PB treatment were the highest for both pin oak in Expt. 2 and Japanese maple in Expt. 3 (Fig. 2); however, dry weights for plants supplied with sulfur and no micronutrients (K₂SO₄, H₂SO₄, elemental S, CaSO₄ treatments) were higher than dry weights for plants in the control treatments for both species. Dry weights for plants supplied with Micromax were also higher than those for the chelate treatment, again indicating the benefit of S supplied by Micromax. For treatments that acidified the substrate (H₂SO₄ and HCl), mean dry weights of the H₂SO₄ treatments were three

Fig. 2. Mean dry weight of container-grown pin oak seedlings (Expt. 2) and Japanese maple seedlings (Expt. 3) as affected by various treatments. Means are results of three subsamples per container ($n = 6$). Data were collected from 3 seedlings per container. Means were separated using Duncan's multiple range test ($P \leq 0.05$). Treatments with the same letters are not significantly different.

Table 2. Substrate solution pH, S, Fe, Cu, Mn, and Zn concentrations for container-grown pin oak seedlings (Expt. 2) and Japanese maple seedlings (Expt. 3) as affected by various treatments. Means are results of three subsamples per container.

Treatment	pH	Element (mg·L ⁻¹)				
		S	Cu	Fe	Mn	Zn
Pin oak						
Control	5.4 a ^z	2.0 d	0.002 c	0.05 b	0.16 c	0.05 cd
Micromax	5.5 a	51.0 b	0.007 a	0.04 bc	0.71 a	0.32 a
K ₂ SO ₄	5.5 a	39.1 c	0.002 b	0.03 bc	0.13 c	0.05 cd
H ₂ SO ₄	5.2 bc	49.0 b	0.002 bc	0.04 bc	0.56 b	0.09 b
HCl	5.1 c	2.6 d	0.002 c	0.03 bc	0.53 b	0.08 bc
Chelate	5.1 c	3.3 d	0.002 c	0.48 a	0.42 b	0.10 b
Elemental S	5.4 a	6.0 d	0.002 c	0.02 c	0.12 c	0.05 d
CaSO ₄	5.4 ab	71.5 a	0.002 c	0.03 bc	0.27 c	0.07 b-d
Japanese maple						
Control	5.35 a	1.39 e	0.002 b	0.048 d	0.09 b	0.09 b
Micromax	5.03 c	43.8 ab	0.006 a	0.084 c	0.23 a	0.23 a
K ₂ SO ₄	5.37 a	40.21 bc	0.003 b	0.16 b	0.08 b	0.08 b
H ₂ SO ₄	5.22 b	31.36 cd	0.003 b	0.076 c	0.11 b	0.11 b
HCl	5.05 c	1.74 e	0.002 b	0.033 d	0.21 a	0.21 a
Chelate	5.38 a	1.3 e	0.002 b	0.316 a	0.1 b	0.1 b
Elemental S	5.18 b	9.63 e	0.002 b	0.081 c	0.13 b	0.13 b
CaSO ₄	5.48 a	28.47 d	0.002 b	0.095 c	0.08 b	0.08 b

^zMeans separation in columns by Duncan's multiple range test, $P \leq 0.05$ ($n = 6$). Treatments within columns with the same letters are not significantly different.

Table 3. Concentrations of elements found in dry weight tissue samples of container-grown pin oak seedlings in Expt. 2 and Japanese maple seedlings in Expt. 3 as affected by various treatments. Means are results of three subsamples per container.

	S	Cu	Fe	Mn	Zn
Treatment	(%)	(mg·kg ⁻¹)			
Pin oak					
Control	0.09 c ^z	4.0 b	63.8 b	966 a	45.0 c
Micromax	0.12 b	5.3 a	78.8 ab	614 c	71.5 a
K ₂ SO ₄	0.15 a	3.0 bc	86.0 a	711 bc	55.8 b
Chelate	0.08 c	2.5 c	63.8 b	916 ab	37.8 d
Japanese maple					
Control	0.09 c	5.0 a	84.5 b	469 a	45.0 c
Micromax	0.19 b	4.3 a	67.8 c	300 b	77.5 a
K ₂ SO ₄	0.29 a	3.5 a	74.0 bc	233.5 c	51.3 bc
Chelate	0.08 c	5.0 a	107.5 a	304.5 b	40.0 c

^zMeans separation in columns by Duncan's multiple range test, $P \leq 0.05$ ($n = 6$). Treatments within columns with the same letters are not significantly different.

times and one and one-half times greater than the HCl treatments for oak (Expt. 2) and maple (Expt. 3), respectively, thus demonstrating the benefit of S (Fig. 2). In contrast to Expt. 1, the Micromax treatment increased growth (Expts. 2 and 3) more than that of other treatments that supplied S alone. This may be due to the higher pH (above 5.0) in the latter experiments, which decreased micronutrient availability by increasing adsorption and precipitation of nutrient cations (Brady and Weil, 2004). The substrate solution pH for all treatments in Expt. 1 was approximately 1 unit lower than the substrate solution pH of all treatments (Expts. 2 and 3) (Tables 1 and 2). As evidence of this, (although not statistically analyzed) substrate solution micronutrient concentrations of the control treatment in Expt. 1 were higher than those in Expts. 2 and 3. This finding is consistent with research by Wright et al. (1999a) who reported that increasing pine bark substrate pH by liming reduced growth of nine container-grown landscape tree species as well as PB solution Fe, Mn, Cu, and Zn concentrations. Micronutrient solution concentrations for both species were generally higher for Micromax than for all other treat-

ments (Table 2). Therefore, substrate solution micronutrient concentrations (all except for Micromax) (Expts. 2 and 3) may be deficient, and the addition of Micromax (micronutrients and S) would increase growth more than when only S is added.

Treatments resulting in the highest dry weights for oak and maple (Micromax, K₂SO₄, H₂SO₄, elemental S, CaSO₄) also had the highest S substrate solution and tissue concentrations (Tables 2 and 3). As with Expt. 1 substrate solution S concentrations were within recommended ranges (Nelson, 1996) and shoot concentrations were comparable to S leaf tissue concentrations found for Japanese maple (0.19% to 0.30%) and pin oak (0.16% to 0.19%) (Mills and Jones, 1996). Tissue Cu and Zn concentrations for oak (Expt. 2) and maple (Expt. 3) in the Micromax treatment were greater or equal to concentrations in the control treatment (Table 3). Also, plant tissue micronutrient concentrations in these two experiments were within sufficiency ranges for the Micromax treatment (Mills and Jones, 1996). Tissue S concentrations for the control treatments (Expt. 3) were below recommended concentrations (Mills and Jones, 1996), but

within sufficiency ranges for the Micromax and S treatments (Table 3). This clearly points to S addition as a major reason why Micromax increases growth of container-grown pin oak and Japanese maple.

In conclusion, the addition of sulfated micronutrients to the PB substrate of pin oak and Japanese maples increased growth primarily by supplying S. Sulfur fertilization is therefore critical for optimal growth of these species when grown in a PB substrate. Also, there are conditions such as high substrate pH, or insufficient levels of micronutrients in a PB substrate that may warrant the addition of micronutrients.

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