Boron Requirement of Young ‘Sharwil’ Avocado Trees

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Abstract. Possible boron (B) deficiency symptoms were observed on avocado (Persea americana Mill. ‘Sharwil’) grown in Kona, Hawaii. To determine the B requirement of young, ‘Sharwil’ avocado trees, two greenhouse experiments were conducted. In a soil study, seven B treatments (0, 3.7, 11, 22, 44, 89, and 178 mg·kg–1 soil fines) were applied to 1-year-old grafted ‘Sharwil’ avocado trees grown for 13 weeks in a Tropofolist soil. Due to the low and variable fractions of soil fines in this rocky soil, extractable B concentration did not appear to be a good predictor of B requirements by avocados. Adequate foliar B concentrations in ‘Sharwil’ avocado trees based on dry weight and area of new leaves ranged from 37 (±3) to 65 (±4) and from 31 (±10) to 78 (±13) mg·kg–1 (dry-weight basis), respectively. (Means are followed by standard errors of the mean in parentheses.) In a hydroponics study, 6-month-old grafted ‘Sharwil’ avocado trees were supplied with four levels of B (0, 1, 10, and 100 μM). At 11 months after B treatment initiation, leaves with deformed margins and a “shot-hole” appearance were first observed at a solution level of 0 μM B. At 14 months after B treatment initiation, foliar B concentrations that were associated with 12% to 14% incidence of deformed leaves ranged from 9.8 to 13.5 mg·kg–1 (dry-weight basis). Although ‘Sharwil’ avocados are reportedly susceptible to B deficiency, foliar B concentrations required for adequate growth and those associated with B deficiency symptoms are similar to those for other cultivars.

In Hawaii, the principal avocado cultivar is ‘Sharwil’ (Bittenbender et al., 1989), a cross between Mexican and Guatemalan races. Avocado production in this state is centered in the district of Kona on the island of Hawaii, typically on Tropofolist (Histosol) organic soils. These soils are unique, because they are composed of a thin organic surface layer underlain by lava (U.S. Dept. of Agriculture–Soil Conservation Service, 1973). Misshapen fruits of ‘Sharwil’ with a lopsided appearance were observed during the 1989–90 season (Bittenbender, 1990). Most of these fruit deformities appeared similar to the sickle-shaped fruit with navel-like lesions caused by B deficiency (Broadley et al., 1991; Piccone and Whiley, 1987; Whiley et al., 1996). Boron deficiency was hypothesized to be the major cause of these misshapen fruits, because foliar B concentrations at >10 avocado farms in the Kona area during Fall 1989 averaged 20 mg·kg–1, which was much below recommended levels (Coetzer et al., 1993; Embleton and Jones, 1966; Piccone and Whiley, 1987; Whiley et al., 1996).

The primary function of B in higher plants is still uncertain, but there is a growing body of evidence that boron is required as a structural component of expanding cell walls (Brown and Hu, 1997; Fleischer et al., 1998; Matoh et al., 1996). In particular, B is required for pollen viability and pollen tube growth (Marschner, 1995). Robbertse et al. (1990) showed that optimal pollen tube growth of ‘Hass’ avocados occurred in flowers with B concentrations ranging from 50 to 75 mg·kg–1. Smith et al. (1997) demonstrated that germination of pollen from B-deficient ‘Hass’ avocado trees was 16-fold less than that from B-fertilized trees. Also, when ‘Hass’ trees with a marginal B status were sprayed with B at the start of anthesis, initial fruit set was increased 42% relative to that of controls, although final fruit yield was not affected (Smith et al., 1997).

Embleton and Jones (1966) reported that adequate foliar B concentration ranged between 50 and 100 mg·kg–1 for avocados, but they stated that additional data were needed to support this range. In Australia, Whiley et al. (1996) stated that the optimal foliar B concentration ranged from 40 to 60 mg·kg–1 for mature summer flush leaves of avocado prior to inflorescence development. In South Africa, Coetzer et al. (1993) reported that the optimal B concentration in leaves below the axillary buds ranged from 60 to 80 mg·kg–1. Genotypic differences have been reported in responses of avocados to B, the cultivar Sharwil being especially susceptible to B deficiency (Piccone and Whiley, 1987; Whiley et al., 1996). However, no information was available as to whether B requirements of ‘Sharwil’ avocados differed from those of other cultivars used to develop adequate ranges of foliar B concentrations.

Objectives of the soil study conducted in the greenhouse were to determine: a) the effect of B fertilization on the vegetative growth of young ‘Sharwil’ avocado trees grown in a Tropofolist soil; and b) the range of B concentrations in leaves and soil fines that are associated with optimum vegetative growth. Objectives of the hydroponics study were to determine: a) symptoms of B deficiency and toxicity on young ‘Sharwil’ avocados; and b) foliar B levels associated with such symptoms.

Materials and Methods

Soil study. The two greenhouse experiments were conducted at a greenhouse in Waiakea, Hawaii (19°39′N, 155°05′W). Avocado seeds of ‘Malama’ were germinated in Feb. 1991 in perlite under 50% shade and intermittent mist. They were transplanted to black 20 × 40-cm polyethylene pots containing 13.1 kg (oven dry weight) of rocks and soil fines from the Kaimu series (isohyperthermic, euc, Typic Tropofolist). Components of this medium were (on a dry-weight basis): rocks >25 mm in diameter, 20%; rocks between 25 and 6 mm, 38%; rocks between 6 and 2 mm in diameter, 24%; and soil fines <2 mm in diameter, 17%. The two larger rock fractions were mixed and placed at the bottom of the pot. The smallest rock fraction and the soil fines were mixed and placed over the larger rock fractions. The proportion of rocks and soil fines in this medium and their distribution in the pot were based on measurements and observations from a Kaimu series soil profile which was located in a ‘Sharwil’ orchard. The soil and rock fractions were obtained from Waiaea, Hawaii (19°22′N, 155°52′W).

In the nonamended soil fines, total N was determined by a micro-Kjeldahl method (Isaac and Johnson, 1976), organic carbon was measured by the method of Hanes (1984), available P was analyzed by the modified Truong method (Ayers and Hagihara, 1952), and exchangeable cations were determined by the ammonium acetate (pH 7) method (Thomas, 1982). Hot water–extractable B levels in the nonamended soil fines were measured using the method of Mahler et al. (1984) and Wolf (1974). Analyses were conducted by the Ag-
ricultural Diagnostic Service Center of the Univ. of Hawai‘i–Manoa. Analysis of the nonamended soil fines indicated (on an oven-dried soil basis): pH, 5.8; total N, 11.4 g·kg⁻¹; organic carbon, 125 g·kg⁻¹; dilute sulfuric acid-extractable P (Truog-P), 150 mg·kg⁻¹. Exchangeable cations were in mg·kg⁻¹ soil fines: K, 310; Ca, 3700; and Mg, 450.

Two and 11.5 months after seed germination, 50 g of Osmocote 13.5–13.5–13.5 (Sierra Chemical Co., Milpitas, Calif.) were broad-cast over the soil surface of each pot. This formulation of a slow-release fertilizer does not contain B. Six months after seed germination, ‘Sharwil’ scionwood was grafted onto the ‘Maluma’ seedling rootstock.

Six months after grafting, trees were randomized and blocks established on the basis of initial plant size and position on the greenhouse bench. Seven B treatments (0, 8, 24, 48, 95, 191, and 381 mg·kg⁻¹ per pot) were applied by pipetting 50 mL of appropriate concentrations of H₃BO₃ onto the soil surface (Table 1). There were a total of 30 pots, with four replicates of treatments 0 to 4 and five replicates of treatments 5 and 6. Trees were irrigated with 500 mL·d⁻¹ (or 1.6 cm·d⁻¹) of tap water that contained nondetectable concentrations of B (<0.01 mg·L⁻¹) as measured by the azomethine-H method (Wolf, 1974). Pesticides used for mite control included malathion [diethyl (dimethoxythiophosphoryl)thio] succinate; Malathion 25W, FMC Corp., Philadelphia] applied immediately prior to B treatment initiation plus 4 weeks later, and dienochlor [ethyl-N-(chloroacetyl)-N-(2,6-diethylphenylglycinate]; Pentac Aquaffl Miticide, Sandoz Crop Protection; Des Plaines, Ill.) sprayed 8 weeks after application of B treatments.

Two weeks prior to the application of B treatments, 12 leaves per tree were sampled from the youngest fully expanded flush of leaves. Then, 9 weeks after B application, about seven leaves per tree were sampled from the youngest fully expanded flush of leaves. Leaves were rinsed three times in deionized water, blotted to remove excess moisture, dried at 75 °C to constant weight, and analyzed for B by the azomethine-H method (Wolf, 1974).

The experiment was terminated 13 weeks after B treatment initiation. Leaves were separated into new leaves (those that developed after B treatment initiation) and old leaves (those that developed before application of B treatments). Roots were washed free of media and separated into fine roots and the tap root. Leaf areas and fine root lengths were determined using a digital image analysis system (Decagon Devices, Pullman, Wash.). These plant parts were weighed both before and after drying at 75 °C.

Treatment effects were evaluated by analysis of variance (ANOVA) (SAS, 1982). Linear B (B), block, and quadratic B (B²) effects were calculated. A probability level of 0.05 or less was considered to be statistically significant. Dry weight and area of new leaves were regressed against foliar B concentrations, using linear and several nonlinear regression models (SAS Institute, 1982). For nonlinear regression models, the coefficient of determination ($r^2$) was calculated to be 1 – (residual sum of squares/corrected total sum of squares). Criteria in determining the regression model with the best fit for each relationship were the highest $r^2$ and adequacy of fit as judged from plots of residual vs. predicted values. Standard errors for the optimal range of foliar B concentrations associated with 90% of maximum leaf areas or leaf dry weights were calculated using the delta method (Bishop et al., 1975).

To determine B sorption of this soil, B as boric acid (H₃BO₃) was added to duplicate 100-g soil samples that were air-dried and sieved (<2 mm). Rates of B applied in mg·kg⁻¹ soil fines were: 0.0, 0.5, 1.0, 1.5, 2.0, 5.0, 10, 20, 50, 100, and 200. The soil fines were brought up to 15% water content and incubated in plastic bags for 18 d and analyzed for hot water-extractable B (Mahler et al., 1984; Wolf, 1974).

**Hydroponics study.** Avocado seeds from ‘Itzamna’ were germinated in June 1991 in a 1 perlite : 1 vermiculite mix (by volume). Seedlings were transplanted to black, 20 × 40 cm polyethylene pots containing rockwool and fertilized with a complete nutrient solution (Tomato 4–18–38; Chem-Gro, Colorado Springs, Colo.). Macronutrient concentrations were, in mm: NO₃-N, 1.84; P, 0.33; K, 1.04; Ca, 0.73; Mg, 0.3; and S, 0.3. Micronutrient concentrations were, in μm: Fe as FeEDTA, 10; Mn, 5; Zn, 1; Cu, 1; Mo, 0.1; and B, 24. Submersible pumps were placed in 100-L plastic containers with nutrient solution to fertigate the seedlings at a rate of 2.5 L·d⁻¹. As the seedlings increased in size, fertigation rates were gradually increased to ≈2.5 L·d⁻¹.

‘Sharwil’ scionwood was grafted onto the rootstock 6 months after the start of germination. Five months after grafting, the ‘Sharwil’ trees were pruned. At 6 months after grafting, B treatments were initiated. Nitrogen solutions were made from reagent grade chemicals with the same concentrations as those used earlier, with B levels of 0, 1, 10, and 100 μm. Trees were fertigated at the rate of ≈2.5 L·d⁻¹. A randomized complete-block design was used and each treatment was replicated six times.

After 2 months of B treatments, leaves and stems that were produced after the start of the experiment were sampled. Leaves were dried at 70 °C and analyzed for B concentration as described previously.

To control mites, dienochlor was sprayed at 5 and 7 months after initiation of B treatments. To control scales, malathion was applied 12 months after start of treatments.

At 14 months after treatment initiation, leaves and stems were separated into new growth (since the last sampling) and old growth (present before the last sampling above the graft union). Leaf areas, and fresh and dry weights of leaves and stems were determined. Leaf samples were analyzed for B as described in the previous study.

Analyses of variance were conducted using SAS programs (SAS, 1982), and linear B (B), block, and quadratic B (B²) effects were calculated. A probability level of 0.05 or less was considered to be statistically significant.

### Results and Discussion

**Soil study.** The concentration of hot water-extractable B in the nonamended soil fines was 0.95 mg·kg⁻¹; this was in the adequate range of 0.59 to 2.58 mg·kg⁻¹ (on air-dried soil basis) found in soils of healthy avocado orchards in California (Haas, 1943). However, soil fines in our study comprised only 17% (dry-weight basis) of the growth medium.

A linear increase in concentration of hot water-extractable B was observed in the soil fines as the rate of B application increased (Fig. 1). This linear regression model was used to calculate initial extractable B concentrations in soil fines due to B treatments (Table 1). In general, when the concentration of extractable B in soil exceeds 5 mg·kg⁻¹, B toxicity symptoms in plants are likely to occur (Reisenauer et al., 1973). In our study, applied B levels ≥11 mg·kg⁻¹ soil fines should have
resulted in potentially toxic extractable soil B levels (Table 1).

Foliar B concentrations did not differ significantly among 'Sharwil' avocado trees prior to the start of the B treatments. The mean foliar B concentration was 69 (±4.8) mg·kg⁻¹. Nine weeks after the start of the treatments, foliar B concentrations increased with increasing B application up to 89 mg·kg⁻¹ fines and then decreased at the highest B application rate (Table 1). Similar results were found for total leaf B contents (data not shown). Foliar B concentrations of trees in the control treatment averaged 39 mg·kg⁻¹ (Table 1), which is a level below those recommended for optimal yields (Coetzee et al., 1992; Emberton and Jones, 1966; Whiley et al., 1996).

Dry weights of new leaves decreased significantly with higher B application rates, particularly at levels ≥44 mg·kg⁻¹ soil fines (Table 1). Leaf area and tap root dry weight responded similarly to increased B application rates (Table 1). The decrease in taproot growth of 'Sharwil' trees at higher B applications rates was similar to that found in two out of three sampling dates for total root dry weight of 'Hass' trees grown at B levels ≥102 mg per pot (Coetzee et al., 1994). This level of B, which is toxic for young 'Hass' trees on a per plant basis, is similar to that found for young 'Sharwil' trees, because application of 95 mg B per pot resulted in a concentration of 44 mg·kg⁻¹ fines.

Dry weights of fine roots averaged 12.4 (±1.2) g per plant and were not affected by B treatments (data not shown). Length of fine roots decreased significantly with increasing B application rates, particularly at levels ≥44 mg·kg⁻¹ fines (Table 1). Apparently, lengths of fine roots were affected more by B treatments than were dry weights. Our results agree with the field observation by Whiley et al. (1996) that avocado trees at an advanced stage of B deficiency had lost most of their feeder roots.

No visual B deficiency symptoms were observed during this study. Foliar B toxicity symptoms were observed 2 months after B application, and were characterized by leaf tip, marginal, and interveinal chlorosis, and necrotic spots similar to those symptoms observed on 'Fuerte' avocado seedlings (Haas, 1929). In addition, "crinkling" of leaves, premature leaf abscission, depression of vegetative flushing, and stunted fine root growth (Table 1) were observed in young 'Sharwil' trees.

Boron toxicity symptoms were observed at foliar B concentrations >190 mg·kg⁻¹ and soil B levels ≥44 mg·kg⁻¹ fines (Table 1). The foliar B concentrations associated with B toxicity in 'Sharwil' trees are within the range of 100 to 250 mg·kg⁻¹ reported by Emberton and Jones (1966) to be in excess for avocado leaves. However, initial soil-extractable B levels ≥18 mg·kg⁻¹ soil fines that were associated with B toxicity in 'Sharwil' avocados (Table 1) greatly exceeded the 5 mg·kg⁻¹ soil reported by Reisenauer et al. (1973) to be toxic for a number of plant species. One possible explanation is that soil fines comprised only 17% of the total dry weight of this Tropofolist soil. As a result, a soil-extractable B concentration of 18 mg·kg⁻¹ soil fines could be considered to be "diluted" by inert rocks, resulting in an actual soil-extractable B concentration of only 3 mg·kg⁻¹ medium. Since percentage of soil fines in these Tropofolist soils varies greatly among farms in Kona, Hawaii, and even within a single orchard (C. Smith, personal communication), extractable B concentrations of soil fines alone do not appear to be adequate for predicting B requirements of 'Sharwil' avocados grown in these unique soils.

This Tropofolist soil is obviously low in B, because foliar B concentration in the absence of

### Table 1. Effects of increasing B application rates on estimated initial extractable soil B, foliar B concentrations at 9 weeks after start of treatments, dry weight of new leaves and taproots, leaf area, and length of fine roots of 'Sharwil' avocados grown for 13 weeks in a soil study.

<table>
<thead>
<tr>
<th>Added B (mg·kg⁻¹ soil fines)</th>
<th>Initial extractable B (mg·kg⁻¹ soil fines)</th>
<th>Foliar B (mg·kg⁻¹)</th>
<th>Dry wt of new leaves (g)</th>
<th>Dry wt of taproot (g)</th>
<th>Leaf area (m²)</th>
<th>Length of fine roots (m)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0</td>
<td>2.2</td>
<td>39 (14)</td>
<td>18.6 (3.1)</td>
<td>25.8 (4.4)</td>
<td>0.30 (0.05)</td>
<td>27.0 (4.8)</td>
</tr>
<tr>
<td>3.7</td>
<td>3.5</td>
<td>49 (9)</td>
<td>28.0 (2.3)</td>
<td>30.8 (2.6)</td>
<td>0.43 (0.06)</td>
<td>28.6 (4.9)</td>
</tr>
<tr>
<td>11</td>
<td>6.0</td>
<td>82 (19)</td>
<td>21.2 (5.4)</td>
<td>29.1 (4.8)</td>
<td>0.41 (0.09)</td>
<td>27.0 (2.0)</td>
</tr>
<tr>
<td>22</td>
<td>9.9</td>
<td>77 (14)</td>
<td>26.1 (4.0)</td>
<td>31.6 (8.1)</td>
<td>0.45 (0.08)</td>
<td>31.1 (7.4)</td>
</tr>
<tr>
<td>44</td>
<td>18</td>
<td>254 (65)</td>
<td>7.7 (2.0)</td>
<td>18.4 (4.7)</td>
<td>0.25 (0.08)</td>
<td>20.1 (6.7)</td>
</tr>
<tr>
<td>89</td>
<td>33</td>
<td>311 (59)</td>
<td>8.6 (3.1)</td>
<td>17.8 (3.5)</td>
<td>0.19 (0.07)</td>
<td>20.4 (8.1)</td>
</tr>
<tr>
<td>178</td>
<td>64</td>
<td>189 (44)</td>
<td>8.8 (3.3)</td>
<td>20.9 (2.2)</td>
<td>0.16 (0.07)</td>
<td>15.0 (2.5)</td>
</tr>
</tbody>
</table>

ANOVA: Pr > F

| B | 0.0006 | 0.002 |
| Block | 0.0200 | 0.870 |
| B² | 0.0001 | 0.049 | 0.09 | 0.390 | 0.77 |

initial extractable B concentrations were estimated by linear regression equation (Fig. 1).

Means are followed by standard errors of the mean (in parentheses).

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**Fig. 2.** Relationship between increasing foliar B concentrations sampled at 9 weeks after start of B treatments and (A) dry weight or (B) area of new leaves of 'Sharwil' avocados grown for 13 weeks in the soil study. Nonlinear regression equations: (A) Dry weight = B/(2.23 – 0.058*B + 0.000907*B²); and (B) Area = B/(55 – 0.024*B + 0.023*B²).
of B fertilization was marginally low (Table 1). Also, high B application rates clearly can depress growth of new leaves and of both tap and fine roots (Table 1).

To estimate the adequate range of B concentrations in ‘Sharwil’ avocado leaves, growth of new leaves over a 13-week period were regressed against foliar B concentrations sampled 9 weeks after the start of B treatments, using nonlinear models (Fig. 2). These nonlinear models were selected based on statistical “goodness of fit,” and not for physiological reasons.

The B concentration adequate for 90% of maximum dry weight of new leaves ranged from 37 (±4) to 65 (±5) mg·kg⁻¹ (Fig. 2A), while that for 90% of maximum area of new leaves ranged from 31 (±13) to 78 (±10) mg·kg⁻¹ (Fig. 2B). These ranges for ‘Sharwil’ avocado seedlings are lower than those previously reported for ‘Fuerte’ avocados of 50 to 100 mg·kg⁻¹ (Embleton and Jones, 1966) or for South African avocados of 60 to 80 mg·kg⁻¹ (Coetzer et al., 1993), but similar to that reported for Australian avocados of 40 to 60 mg·kg⁻¹ (Whiley et al., 1996). These ranges for ‘Sharwil’ avocados are based on vegetative growth, and yield response curves can vary depending on whether the tissues sampled are vegetative or reproductive (Marschner, 1995). In a later paper, we will compare these results with those obtained based on ‘Sharwil’ fruit yields.

Hydroponics study. Within 30 d after initiation of B treatments, interveinal chlorosis and marginal necrosis were observed in leaves grown at 100 μM B. These symptoms are characteristic of B toxicity in avocados (Haas, 1929). Two months after initiation of treatments, there were no significant effects of B on leaf dry weight or leaf area. Average leaf dry weight per tree was 19.9 (±1.1) g and average leaf area was 26.9 (±1.5) m². Boron concentration in leaves increased significantly with increasing B levels in solution, and the foliar B concentration associated with B toxicity symptoms was 232 (±23) mg·kg⁻¹ (Table 2).

Avocado is reportedly sensitive to irrigation waters having >92 μM B (Gupta et al., 1985). Also, Haas (1929) found toxicity symptoms in ‘Fuerte’ avocado seedlings grown in sand culture with 92 to 185 μM B. Thus, ‘Sharwil’ avocado trees exhibit B toxicity symptoms at solution B concentrations similar to those reported for other cultivars.

Eleven months after initiation of B treatments, leaves with deformed margins and a “shot-hole” appearance were first observed at 0 μM B (Fig. 3). “Shot-holes” in leaves were associated previously with B deficiency in field-grown avocado trees (Broadley et al., 1991; Whiley et al., 1996). However, this report is the first to demonstrate that an induced B deficiency results in appearance of “shot-holes” in ‘Sharwil’ avocado leaves.

At 14 months after initiation of treatments, increasing solution B levels significantly increased foliar B concentrations in new leaves, and significantly decreased percentage of deformed leaves (Table 2). The foliar B concentrations associated with 12% to 14% deformed leaves ranged from 10.3 to 13.5 mg·kg⁻¹ (Table 2). The older leaves (produced prior to the start of the experiment) did not exhibit such deformities. Haas (1943) found that B deficiency in avocado seedlings caused terminal dieback, burned and distorted leaves, and corky, split midribs of leaves; however, he did not report appearance of “shot-holes” in leaves. In addition, Haas (1943) reported that foliar B concentrations associated with B deficiency symptoms ranged from 9 to 18 mg·kg⁻¹; these levels are similar to those that we found to be associated with a 12% to 14% incidence of “shot-holes” in ‘Sharwil’ leaves.

At the 14-month harvest, dry weights of new leaves significantly decreased with increasing B levels, particularly at 100 μM B (Table 2). New leaf area averaged 0.59 (±0.03) m² per plant and were not affected significantly by B levels.

Conclusions

Based on vegetative growth in the greenhouse, adequate foliar B concentrations of young ‘Sharwil’ avocado trees ranged from 40 to 70 mg·kg⁻¹. Boron deficiency symptoms were characterized by appearance of “shot-holes” and deformed margins in leaves. Foliar B concentrations associated with 12% to 14% deformed leaves ranged from 10.3 to 13.5 mg·kg⁻¹.
13.5 mg·kg\(^{-1}\). Boron toxicity symptoms of ‘Sharwil’ avocados were found at levels of 100µM B in nutrient solution. Foliar B concentrations associated with B toxicity symptoms ranged from 190 to 232 mg·kg\(^{-1}\). Levels of extractable soil B in these unique, rocky, Tropofolist soils were not good predictors of B requirements of ‘Sharwil’ avocado trees because of the low and variable fractions of soil fines.

**Literature Cited**


