**pH Dominates Leucadendron ‘Safari Sunset’ Growth**

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Abstract. The objectives of the present research were to study the effects of pH, NH₄NO₃ ratio, and P concentration in the nutrient solution on development of Leucadendron R. Br. ‘Safari Sunset’ [L. salignum Bergius × L. laureolum (Lam.) Fourn.]. The experiment was conducted in aero-hydroponic systems and involved six treatments in a nonfactorial design: two pH levels (5.5 and 7.5), two P levels (7 and 20 mg·L–1), and two NH₄:NO₃ ratios (60:40 and 25:75). The pH of the root environment was the most important factor controlling growth. Root cells were longer in plants grown at pH 5.5 than at pH 7.5, but width was not affected. Altering the NH₄:NO₃ ratio did not affect development regardless of pH. Increasing the P concentration from 7 to 20 mg·L–¹ significantly decreased root fresh weight at the low pH and slightly reduced shoot growth. Nitrogen, P, K, Zn, and Mn concentrations were higher, while that of Fe was lower in plants grown at low pH. Reducing the NH₄:NO₃ ratio did not affect N concentration but increased P and K concentrations in the shoots. Increasing the P concentration significantly raised the P content of shoot and root tissues but reduced the content of Fe, Zn, and Mn.

The Proteaceae family originated in Australia and South Africa, where most species grow on leached, acidic soils, which are poor in available minerals. Growth reduction and leaf necrosis or chlorosis are generally attributed to phosphorus toxicity (Buining and Cresswell, 1993; Nichols et al., 1979). However, little information is available on the nutritional requirement of plants of this family (Parks et al., 1996).

In the last decade, an effort has been made in Israel to cultivate Proteaceae species (protea) for cut flowers (Ben-Jaacov, 1986; Ben-Jaacov et al., 1989). Despite the suitable climate, Israeli protea growers have encountered problems because of unfavorable soil characteristics, such as high pH and high free lime content. To avoid soil limitations ‘Safari Sunset’, the main commercially cultivated protea produced in Israel, is grown in tuff, a volcanic pyroclastic material characterized by high porosity (0.6 L·L⁻¹) and high saturated hydraulic conductivity (Wallach et al., 1992), or is grafted on lime-resistant rootstocks (Ben-Jaacov et al., 1992).

Growing plants on an artificial substrate and using modern irrigation and fertilization equipment can provide appropriate conditions for plant growth, including control of the pH in the rhizosphere. The pH in the root environment can affect plant growth through two major mechanisms: 1) a direct effect on cell elongation and root hair growth (Taiz, 1984; Tang et al., 1992; White, 1990); and 2) indirect effects through nutrient availability and ion uptake by plants (Marschner, 1995). Rhizosphere pH is affected by the cation and anion uptake ratio (Marschner, 1995) in the nutrient solution, and by microbial activity (mainly nitrification and denitrification). Altering the nitrogen source, e.g., the NH₄:NO₃ ratio, can also influence the pH in the rhizosphere.

In a previous study, Silber et al. (1998) found that nutritional treatments affected the growth of L. ‘Safari Sunset’ planted in pots filled with tuff. However, since N levels (at fixed NH₄:NO₃ ratio) or the NH₄:NO₃ ratios (at fixed N level) affected the rhizosphere pH as well, distinguishing between the main treatment effects (NPK levels and NH₄:NO₃ ratio) and that of pH changes was not feasible. Differentiating between these two effects is only possible by using a system such as the aero-hydroponic system.

The objectives of this study were to determine the effects of nutrient solution pH, NH₄:NO₃ ratio, and P concentration on nutrient uptake and growth of L. ‘Safari Sunset’, grown in an aero-hydroponic system.

Materials and Methods

The experiment was conducted in a screen house (10% shade) in Bet Dagan, Israel (35°E, 31°N, 50 m altitude), irrigated by natural sunlight at a temperature range between 12 and 35 °C. Two-month-old L. ‘Safari Sunset’ plants were transplanted into an aero-hydroponic system (Fegin et al., 1984). Each plot consisted of 12 plants placed in two separate polystyrene boxes mounted on a 140-L, covered container. Roots were continuously exposed to the nutrient solution, which was spread on the roots by means of a plastic tube system. The solution was collected in the bottom of the container and continuously recirculated. The experiment included six treatments (Table 1) with two pH levels (5.5 and 7.5), two P levels (0.23 and 0.65 mmol·L⁻¹) and two NH₄NO₃ ratios (60:40 and 25:75).

The treatments were chosen to reflect different conditions that take place in the rhizosphere of L. ‘Safari Sunset’ because of changing NPK levels or NH₄:NO₃ ratio (Silber et al., 1998). Potassium concentration in the nutrient solution of all the treatments was 1.3 mmol·L⁻¹. The solutions were prepared with commercial fertilizers [KNO₃, (NH₄)₂SO₄, NH₄NO₃, KCℓ, KH₂PO₄], and typical tap water, as used in production greenhouses in Israel. The tap water contained (mmol·L⁻¹): NO₃-N–0.7, P–0.01, Ca–1.25, Mg–0.8, Na–4.3, and Cl–3.9. Microelement concentrations were (µmol·L⁻¹): Fe–12.3, Mn–6.2, Zn–2.6, Cu–0.4, Mo–0.2 and B–23, all EDTA-based, plus 36 µmol·L⁻¹ Fe as EDDHA-Fe. The pH was monitored daily and adjusted to the desired pH level by adding 0.1 mmol·L⁻¹ NaOH or H₂SO₄. The electrical conductivity was ≈2 dS·m⁻¹ and was not changed significantly by addition of H₂SO₄ or NaOH. The solutions were renewed weekly, and water lost was replaced daily. The experiment included five replicates arrayed in a completely randomized nonfactorial design.

At the end of the experiment the plants were harvested and root and shoot fresh and dry weights (after drying at 60 °C) were determined. Dry plant material was ground to pass a 20-mesh sieve. Total N, P, and K in tissue were determined after digesting with H₂SO₄·H₂O₂ using an autoanalyzer (Technicon Corp., Tarrytown, N.Y.) (for N and P) or a flame photometer (for K). Calcium, Mg, Fe, Zn, and Mn were determined by atomic absorption after digesting the dry tissue with HNO₃·HClO₃.

Scanning electron microscopy (SEM) was used to examine roots grown in nutrient solution (N1P1) at the two pH treatments. The samples were fixed in 3% glutaraldehyde in 0.1 M cacodylate buffer, pH 7.0, and dehydrated in a graded acetone series up to 100%. Directly after this step, the samples were critical-point-dried. They were mounted on SEM stubs and sputter-coated with gold to a thickness of 0.1 mm. The samples were viewed and photographed with a JSM-T330A instrument (JEOL, Tokyo).

Data were subjected to analysis of variance using the GLM procedure of SAS (SAS Inst., Cary, N.C.).

Table 1. Nitrogen and P concentrations (mmol·L⁻¹) added to the irrigation water.

| Treatment | Nutrient solution | NH₄-N | NO₃-N | P |
|-----------|-------------------|-------|-------|---|
| 5.5       | N1P1              | 2.5   | 1.1   | 0.23|
|           | N1P2              | 2.5   | 1.1   | 0.65|
|           | N2P1              | 1.1   | 2.5   | 0.23|
| 7.5       | N1P1              | 2.5   | 1.1   | 0.23|
|           | N1P2              | 2.5   | 1.1   | 0.65|
|           | N2P1              | 1.1   | 2.5   | 0.23|
Table 2. Effects of pH and nutrient content of solutions on fresh and dry weights (g/plant) of shoot and root of ‘Safari Sunset’ plants.

| pH  | Treatment | Fresh wt | Dry wt |
|-----|-----------|----------|--------|
|     | Shoot     | Root     | Shoot  | Root   |
| 5.5 | N1P1      | 67.2a    | 21.7a  | 11.0   | 1.7a   |
|     | N1P2      | 51.5 ab  | 13.0bc | 10.4   | 1.5a   |
|     | N2P1      | 58.6 a   | 20.4 ab| 9.1    | 1.4a   |
| Mean|           | 59.1     | 18.3   | 10.2   | 1.5    |
| 7.5 | N1P1      | 34.3 bc  | 7.9 c  | 8.0    | 0.8 b  |
|     | N1P2      | 23.8 c   | 5.7 c  | 6.4    | 0.6 b  |
|     | N2P1      | 34.5 bc  | 8.4 c  | 8.0    | 0.8 b  |
| Mean|           | 30.9     | 7.5    | 7.5    | 0.7    |
| Mean| N1P1      | 50.7     | 14.8   | 9.0    | 1.2    |
|     | N1P2      | 37.7     | 9.3    | 8.4    | 1.0    |
|     | N2P1      | 46.6     | 14.3   | 8.6    | 1.1    |
|     | LSD-pH    | 11.6     | 4.2    | 1.9    | 0.3    |
|     | F–pH      | 16.***   | 8.1*** | 5.3    | 15***  |
| F–Tr| 10.1      | 7.3     | 3.4    | 0.6    |
| Mean| 6.6***     | 8.1***   | NS     | 5.7*** |

1Mean separation within columns by LSD test, P ≤ 0.05.
2LSD-pH and F–pH tests between the two pHs.
3LSD-Tr and F–Tr-F and LSD tests between treatments.
4*, **, ***Nonsignificant or significant at P ≤ 0.05, 0.01, or 0.001, respectively.

Table 3. Effects of pH and nutrient content of solutions on element concentrations in shoot and root of Leucadendron ‘Safari Sunset’ plants.

| pH  | Treatment | Shoot | Root |
|-----|-----------|-------|------|
|     | N         | P     | K    | Fe   | Zn   | Mn   | N    | P    | K    |
| 5.5 | N1P1      | 2.14a | 0.19c | 0.23ab| 1.9abc| 1.8a | 4.7b | 2.44 | 0.21b| 0.22ab|
|     | N1P2      | 1.78b | 0.32a | 0.20b | 1.1d  | 1.2b | 3.6c | 2.43 | 0.27a| 0.18bc|
|     | N2P1      | 2.05a | 0.24a | 0.25a | 1.15cd| 1.8   | 6.6c | 2.09 | 0.29a| 0.26a |
| Mean|           | 1.99   | 0.25  | 0.23  | 1.5   | 1.6  | 5.0 | 2.31 | 0.26  | 0.22 |
| 7.5 | N1P1      | 1.47c | 0.09d | 0.14cd| 1.6bc | 0.9b | 1.9d | 2.59 | 0.11c| 0.12cd|
|     | N1P2      | 1.20d | 0.11d | 0.12d | 1.9ab | 1.1b | 1.5d | 2.10 | 0.09c| 0.09d |
|     | N2P1      | 1.29d | 0.08d | 0.17c | 2.0a  | 1.8a | 1.2c | 3.08 | 0.77c| 0.16bc|
| Mean|           | 1.32   | 0.95  | 0.14  | 1.8   | 1.3  | 1.5 | 2.57 | 0.10  | 0.12 |
| Mean| N1P1      | 1.80  | 0.14 | 0.19  | 1.7   | 1.4  | 3.3 | 2.52 | 0.16  | 0.17 |
|     | N1P2      | 1.49  | 0.22 | 0.16  | 1.5   | 1.2  | 2.6 | 2.26 | 0.18  | 0.13 |
|     | N2P1      | 1.67  | 0.16 | 0.21  | 1.8   | 1.8  | 3.9 | 2.27 | 0.2   | 0.21 |
| Mean| LSD–pH    | 0.02  | 0.11 | 0.02  | 0.24  | 0.24 | 0.47| 0.54 | 0.03  | 0.04 |
| Mean| F–pH      | 126***| 211***| 65***| 8.3***| 8.5***| 211***| NS   | 98***| 21***|
| Mean| LSD–Tr    | 0.04  | 0.19 | 0.03  | 0.38  | 0.38 | 0.54| 0.93 | 0.06  | 0.06 |

1Mean separation within columns by LSD test, P ≤ 0.05.
2LSD-pH and F–pH tests between the two pHs.
3LSD-Tr and F–Tr-F and LSD tests between treatments.
4*, **, ***Nonsignificant or significant at P ≤ 0.05, 0.01, or 0.001, respectively.
At acid pH, lowering the NH₄NO₃ ratio affected only the P and Mn concentrations (Table 3). Reducing the NH₄NO₃ ratio usually increases cation and reduces anion concentrations because of anion—cation balance (Marschner, 1995). Thus, the higher Mn concentration in the shoots is consistent with the above mechanism, but the higher P concentration (shoot and root) is not (Marschner, 1995; Mengel and Kirkby, 1987). The effects of NH₄NO₃ ratio on elemental concentrations at the basic pH were inconsistent (increasing Fe and Zn while decreasing that of Mn), and were probably secondary effects of injury of the root system at basic pH.

The effects of varying the P concentration in the nutrient solution on elemental concentrations in the plants were more significant for plants grown at pH 5.5, probably because their roots developed normally. Increasing the P concentration in the nutrient solution significantly raised the P content in shoot and root, but reduced the concentrations of the Fe, Zn, and Mn (Table 3). Micronutrient contents in shoots were not in the deficient or toxic range, according to Marschner (1995) or Jones et al. (1991), and, therefore, we assumed that the micronutrient supply did not limit plant growth. Phosphorus concentration in L. ‘Safari Sunset’ plants increased because of increased P concentration in the nutrient solution, and not because of the “Zn deficiency-enhanced P uptake” mechanism, as proposed by Cakmak and Marschner (1986) for cotton (Gossypium hirsutum L.). The extension of the term “P-induced zinc deficiency” (Cakmak and Marschner, 1986, 1987; Loneragan and Webb, 1993; Marschner and Cakmak, 1986) to Fe and Mn is probably incorrect because of the relatively high metal content in shoots, even in plants with high P content (Table 3). Growing the plants for a longer time might have increased the differences between high- and low-P plants and have permitted signs of metal deficiency or “P toxicity” to become visible.

The exact mechanism causing lower metal contents in the shoot of the high-P-fed plants is not clear. No conditions for adsorption or precipitation of metal-P prevail in a hydroponic system at low pH. No “dilution effect” (Loneragan and Webb, 1993; Marschner, 1995) can be invoked, since shoot and root dry weights were not significantly affected by elevation of the P concentration (Table 2). The lower content of metals may have resulted from an inhibition of metal absorption by the root system or a translocation of metals to the shoots, or it might have resulted from internal immobilization because of the formation of metal-phosphate compounds, such as the formation of Zn-phytate in several crops reported by van Steveninck et al. (1993).

Conclusions

The pH of the root environment was the most important factor affecting L. ‘Safari Sunset’ growth in the present study. Whether pH affects plant development directly through physiological mechanisms that influence root hair formation, or indirectly through mechanisms of nutrient availability, is not clear. Apparently, element solubility in an aeroponic system is excellent and does not restrict the uptake of elements. In fact, precipitation of insoluble compounds of metal-P or metal oxides may occur on the external surface of the roots grown in high pH, which reduces metal solubility. However, the fact that Fe, Zn, and Mn concentrations, even in shoots of plants grown at pH 7.5, were within the sufficiency range supports the hypothesis that pH mainly affected physiological factors.

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