Low pH Enhances Rooting of Stem Cuttings of Rhododendron in Subirrigation[1]

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Abstract

Semi-hardwood terminal stem cuttings of *Rhododendron* L. 'P.J.M.', *R.* 'Catawbiense Album' and *R.* 'Purple Gem' were treated with a solution of 0.1% indolebutyric acid and 0.5% napthaleneacetic acid and inserted into perlite for rooting under subirrigation, without overhead mist, at solution pH of 4.5 or 7.5. All cultivars rooted in higher percentages and produced larger root balls at pH 4.5 than at pH 7.5. A second study with softwood stem cuttings of *R.* 'P.J.M.' and both softwood and semi-hardwood stem cuttings of *R.* 'Purple Gem' confirmed these findings and included an additional subirrigation treatment, a sphagnum peat slurry, pH 4.1, which produced nearly identical results to the pH 4.5 treatment.

Index words: adventitious root formation, intermittent mist, plant propagation.

Significance to the Nursery Industry

Advances in stem cutting propagation involving intermittent mist, fog, and plant growth regulators allow growers to propagate many species in large quantities with ease and flexibility. However, prolonged exposure to intermittent mist can decrease cutting quality. Subirrigation is a novel system of propagation that permits growers to root many softwood and semi-hardwood cuttings without mist. However, it has been reported that rhododendrons are difficult to propagate with subirrigation. Here we report on the rooting of stem cuttings from three rhododendron cultivars in a subirrigation system and the effect of subirrigation solution pH on rooting. We achieved 85% to 100% rooting on stem cuttings of rhododendron cultivars 'P.J.M.', 'Purple Gem', and 'Catawbiense Album' by decreasing solution pH from 7.5 to 4.5. A simple method of decreasing solution pH using milled sphagnum peat was also evaluated.

Introduction

In the early 1950s, development of intermittent mist systems allowed growers to root stem cuttings of many plants for the first time with commercially acceptable results. However, mist propagation has its problems, including leaf chlorosis, mineral nutrient leaching, salt build-up on cutting leaves, leaf necrosis, algal growth, waterlogged rooting medium, clogging of nozzles, and other maintenance requirements (7, 10). A novel method of rooting cuttings uses subirrigation as a water source instead of overhead intermittent mist (11, 14). Cuttings of several taxa [maple (*Acer* L.), barberry (*Berberis* L.), birch (*Betula* L.), chrysanthemum (*Dendranthema* (DC.) Desmoul.), coleus (*Solenostemon* Thonn.) and smoke bush (*Cotinus* Mill.)], rooted at least as well, if not better, with subirrigation than with intermittent mist (2, 14). Subirrigation systems utilize containers filled with a coarse medium, such as perlite, which supports the stem cuttings and conducts water to the cutting by capillary action from a water source maintained at a constant level below the cutting base (11, 14). However, poor success has been reported for rooting cuttings of rhododendron using subirrigation (2).

Rooting media provide support for the cutting and supply water and oxygen to the cutting base as root initiation, root growth and development proceed (7). Physical and chemical characteristics of the rooting medium, including solution pH, can strongly influence root initiation (9, 13). The pH of the perlite used in subirrigation systems may range from 6.0 to 8.0 (7). However, tissue culture research has shown that root initiation on thin cell layer explants of tobacco (*Nicotiana* L.) is greatest when the pH of the culture medium is 3.8 to 5.0 (1). A study of *in vitro* rooting of several Australian woody species found that explants developed roots with a medium pH of 4.0, but not when the medium pH was 5.5 (13). Harbage and Stimart (6) reported that root formation and the uptake, but not metabolism, of 1*H*-indole-3-butyric acid (IBA) in apple (*Malus* Mill.) were inversely related to the medium pH of tissue-cultured microcuttings and, furthermore, less auxin was required for rooting at lower pH (5).

Cuttings of rhododendron (*Rhododendron* L.) typically are rooted under intermittent mist in a medium of peat:perlite at ratios of 1:1 or 1:2 (by vol) (3, 7, 10). In our experience these media yield a pH under 5.0. Under intermittent mist cuttings of rhododendron take from 2 to 4 months to root, in which time they may suffer in quality from mist-associated problems noted previously. The objective of this research was to propagate stem cuttings of rhododendron under subirrigation and evaluate the effects of solution pH on rooting. We report here on the rooting of three cultivars of rhododendron, 'P.J.M.', 'Purple Gem', and 'Catawbiense Album' in subirrigation systems with solutions of different pH or a sphagnum peat slurry.

Methods and Materials

Semi-hardwood terminal stem cuttings 12.7 cm (5 in) long of 'P.J.M' and 'Catawbiense Album' were collected from field-grown stock blocks on August 2, 1996, and August 30, 1996, respectively. Similarly softwood terminal stem cuttings of 'Purple Gem' and 'P.J.M' were collected on April 20, 1997, from field-grown balled and burlapped stock plants forced in a warm greenhouse. Semi-hardwood terminal stems cuttings of 'Purple Gem' also were collected from stock blocks on August 10, 1997. Cuttings of 'P.J.M.' and 'Purple Gem' were trimmed to six leaves, while cuttings of 'Catawbiense

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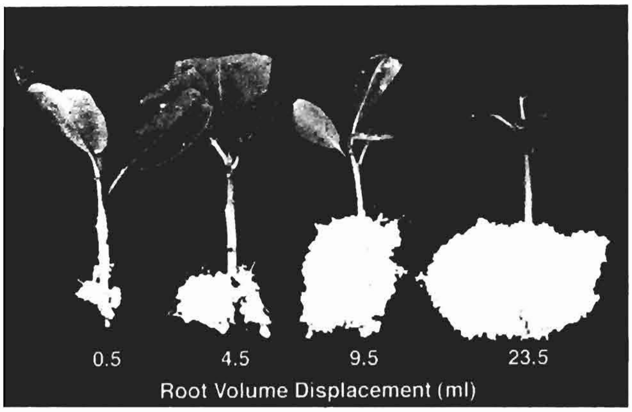

Fig. 1. Range of rooting on rooted stem cuttings of *Rhododendron* 'Catawbiense Album' after 63 days in a subirrigation system. Corresponding volume displacement values (ml) of roots (plus perlite not removed by washing) are indicated below each cutting. A root volume displacement of ≥ 4.5 ml (0.14 fl oz) was considered commercially acceptable.

Album' and were trimmed to four leaves. Leaves of 'Catawbiense Album' were further trimmed to approx. one-half the original leaf size (Fig. 1). Cuttings were wounded by making a 2.5 cm (1 in) slice on one side, and treated with a 1:10 aqueous dilution of Dip 'n Grow™ (1.0% IBA, 0.5% naphthalene acetic acid, Astoria-Pacific, Clackamas, OR) for 5 sec before insertion into the propagation medium.

The subirrigation system consisted of 2.4-liter (2.5 qt) No. 1 nursery containers [15 cm (5.9 in) wide × 18 cm (7.1 in)] filled with coarse perlite and placed in plastic flats [50 cm (19.7 in) × 35 cm (13.8 in) × 6 cm (2.4 in)] lined with 6 mil (1.4 mm, 0.06 in) black plastic and filled to a depth of 5.1 cm (2 in) with tap water (pH 7.5). Each nursery container contained ten stem cuttings inserted 6 cm (2.4 in) deep into perlite. The level of the subirrigation solution was maintained about 4 cm (1.6 in) below the cutting bases. The high pH treatment (pH = 7.5) consisted of perlite subirrigated with tap water. The low pH treatment consisted of perlite rinsed in a weak sulfuric acid solution prepared by titrating tap water with 0.01 N sulfuric acid to pH 4.5. This appeared to saturate the buffering capacity of the perlite. The acid-washed perlite was then subirrigated with tap water titrated to pH 4.5 with 0.01 N sulfuric acid. Replacement water of the appropriate pH was added to subirrigation flats weekly to compensate for evaporation. The pH was monitored weekly to ensure consistency. In this time the low pH treatment tended to rise to a pH of 4.6 to 4.7, and was readjusted weekly to pH 4.5 with 0.01 N sulfuric acid. A third treatment used, in the second year only, with cuttings of 'Purple Gem' and 'P.J.M.' was a subirrigation solution prepared from a slurry of milled sphagnum peat and tap water, 1:4 (by vol). The pH of this system was 4.1 ± 0.1 throughout the experiment. Tap water was added to the peat slurry weekly to compensate for evaporation.

The propagation system was maintained in a greenhouse under a single layer of 80% Chicopee shade cloth (Lumite, Gainesville, GA). During the 1996 experiments temperatures in the propagation area averaged 25 ± 5C (77F) day and 22 ± 3C (72F) night [range 19C (66F) to 32C (90F)], relative humidity averaged 88 ± 9% [range 61 to 97 %; measured using a CS500 temperature and relative humidity probe (Campbell Scientific, Logan, UT)]. Peak irradiance, recorded between noon and 1:00 PM, averaged 88 $\mu mol \cdot m^{-2} \cdot s^{-1}$ [range 27 to 116 $\mu mol \cdot m^{-2} \cdot s^{-1}$, measured using a Li-Cor LI-190SB quantum sensor (Li-Cor, Lincoln, NE)]. Environmental data were similar in 1997 (data not presented). Photoperiods during the late summer experiments ranged from 10.3 to 14.2 hours, and during the spring experiments from 10.6 to 15.3 hours in length.

Each treatment was evaluated using four replicate subirrigation systems (flats) containing one nursery container with 10 cuttings for each cultivar, for a total of 40 cuttings of each cultivar evaluated per treatment. The flats were placed on a greenhouse bench in a completely randomized design. On September 18, 1996, cuttings of 'P.J.M' were harvested and percentage rooting recorded. A cutting was considered rooted if it possessed one or more roots >1 mm (0.04 in) long. Rooted cuttings were washed gently in water to remove as much perlite as possible, and the cutting bases submerged in a 100 ml (3 fl oz) graduated cylinder to measure volume displacement. On October 31, 1996, cuttings of 'Catawbiense Album' were harvested, and percentage rooting and volume root displacement measured as above. Cuttings of 'Purple Gem' and 'P.J.M.' were harvested on June 22, 1997, and data recorded as above, except that root volume displacement was measured in 25 ml (0.75 fl.oz.) and 10 ml (0.3 fl.oz.) graduated cylinders as appropriate. Cuttings of 'Purple Gem' were also harvested on October 18, 1997.

Statistical analyses were performed for each cultivar at each harvest date to determine if solution pH influenced rooting results. Data were analyzed using ANOVA, and treatment means compared with Duncan's multiple range test (12). Percentage data were arcsine transformed before analysis.

Results and Discussion

Solution pH of the subirrigation system had a dramatic effect on root initiation or root development on cuttings of

Table 1. Rooting response and root volume displacement[z] of stem cuttings of *Rhododendron* 'Catawbiense Album' and *R.* 'P.J.M.' rooted in a subirrigation system at solution pH of 4.5 or pH 7.5.

Cultivar	Collection date	Rooting time (days)	Rooting percentage[y]		Root volume displacement (ml)[y]	
			pH 4.5	pH 7.5	pH 4.5	pH 7.5
'Catawbiense Album'	8/30/1996	63	88a[x]	73a	12.4a	2.5b
'P.J.M.'	8/2/1996	48	100a	53b	7.6a	0.8b

[z]Root volume displacement includes roots plus perlite not removed by washing.

[y]Means are the average of four replicate subirrigation systems containing 10 cuttings each of the indicated cultivars (40 cuttings of each cultivar per treatment).

[x]Means separation within row and rooting parameter by Duncan's multiple range test, $p \leq 0.05$.

Table 2. **Rooting response and root volume displacement[z] of cuttings of *Rhododendron* 'P.J.M.' and *R.* 'Purple Gem' rooted in a subirrigation system at solution pH of 4.5 or pH 7.5, or a peat slurry.**

Cultivar	Collection date	Rooting time (days)	Rooting percentage[y]			Root volume displacement (ml)[y]		
			pH 4.5	pH 7.5	Peat slurry	pH 4.5	pH 7.5	Peat slurry
'Purple Gem'	4/20/1997	63	98a[x]	80a	95a	3.5a	0.3b	3.2a
'P.J.M.'	4/20/1997	63	85a	38b	88a	2.8a	0.2b	3.9a
'Purple Gem'	8/10/1997	69	88a	50b	87a	3.7a	0.5b	3.8a

[z]Root volume displacement includes roots plus perlite not removed by washing.

[y]Means are the average of four replicate subirrigation systems containing 10 cuttings each of the indicated cultivars (40 cuttings of each cultivar per treatment).

[x]Means separation within row and rooting parameter by Duncan's multiple range test, $p \leq 0.05$.

every cultivar of rhododendron tested (Tables 1 and 2). Rooting of 'P.J.M.' at low pH was nearly twice that at high pH, and root volume displacement was 10 times greater on cuttings rooted at low pH (Table 1). Stem cuttings of 'Catawbiense Album' did not root significantly better at low pH, but did produce a root system five times larger in the low pH treatment (Table 1). A root volume displacement of about 2 ml (0.06 fl.oz.) was judged to be the minimum commercially acceptable size of a rooted cutting of 'P.J.M.', and a 4.5 ml (0.14 fl.oz.) displacement the minimum commercially acceptable size of a rooted cutting of 'Catawbiense Album' (Fig. 1). At a medium pH of 7.5, only 5 of 40 cuttings of 'Catawbiense Album', and 4 of 40 cuttings of 'P.J.M.', produced commercially acceptable root balls (Fig.

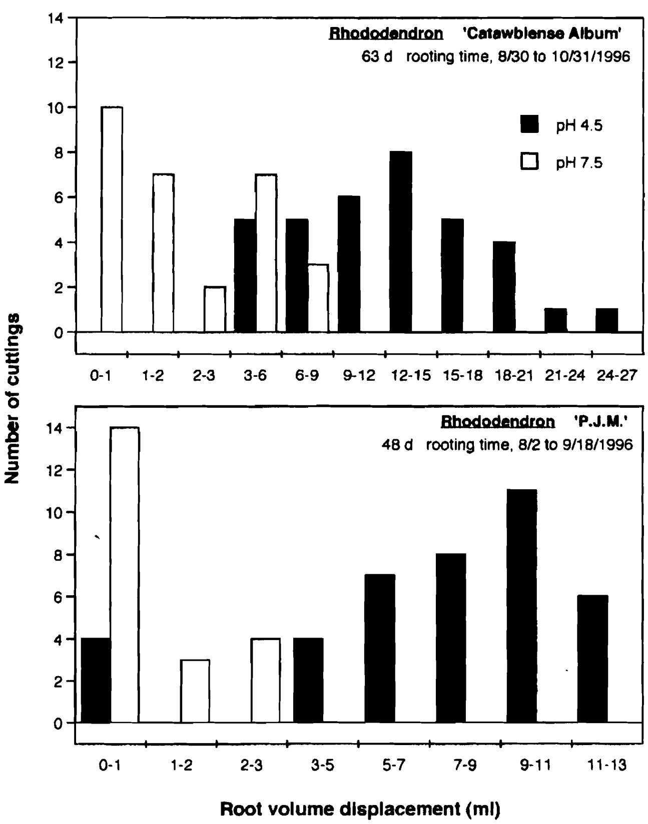

Fig. 2. **Volume displacement of roots (plus perlite) of stem cuttings of *Rhododendron* 'Catawbiense Album' and *R.* 'P.J.M.' rooted in a subirrigation propagation system with solution pH of 4.5 or 7.5. Root volume displacements of ≥ 4.5 ml (0.14 fl oz) and ≥ 2 ml (0.06 fl oz) were considered commercially acceptable for 'Catawbiense Album' and 'P.J.M.', respectively.**

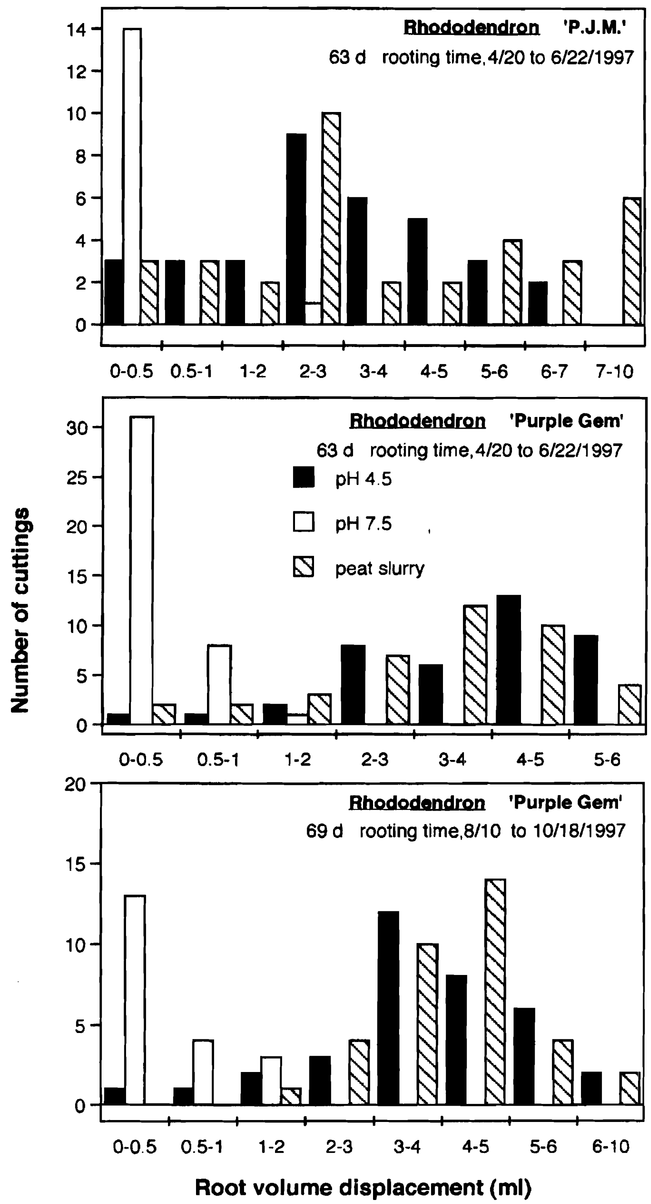

Fig. 3. **Volume displacement of roots (plus perlite) of stem cuttings of *Rhododendron* 'P.J.M.' and *R.* 'Purple Gem' rooted in a subirrigation propagation system with solution pH of 4.5 or 7.5, or a peat slurry. Root volume displacements of ≥ 2 ml (0.06 fl oz) were considered commercially acceptable for both cultivars.**

2). At the lower solution pH, cuttings of 'Catawbiense album' yielded 33 of 40 cuttings with root volumes > 4.5 ml (0.14 fl.oz.), while 'P.J.M.' yielded 36 of 40 cuttings with root volumes > 3 ml (0.09 fl.oz.).

In the second year (Table 2), rooting percentage of cuttings of 'P.J.M.' was again doubled at pH 4.5, and root volume displacement again increased more than 10-fold. Rooting percentage of cuttings of 'Purple Gem' in the spring did not differ significantly with treatment, while those propagated in late summer rooted better at pH 4.5. Root volume displacement of rooted cuttings of 'Purple Gem' increased dramatically with the low pH treatment in both experiments (Table 2). In every experiment the peat slurry produced rooting percentages and root volume displacements equivalent to those at pH 4.5 (Table 2). In this second experiment a root volume displacement of 2 ml (0.06 fl.oz.) was judged to be commercially acceptable for cuttings of both 'P.J.M.' and 'Purple Gem'. Given this threshold, at pH 7.5 only 1 of 40 cuttings of 'P.J.M.' and no cuttings of 'Purple Gem' produced commercially acceptable rooting (Fig. 3). At pH 4.5 or with the peat slurry, 'P.J.M.' produced 25 and 27 commercially acceptable root balls, respectively, out of 40 cuttings. In these same treatments, 'Purple Gem' produced 37 and 33 commercially acceptable root balls, respectively, in the spring, and 31 and 34 acceptable root balls, respectively, in late summer (Fig. 3).

In additional experiments with *R.* 'P.J.M' and *Ilex* x *meserveae* S.Y. Hu., we amended the subirrigation medium from perlite alone to perlite:peat ratios of 5:1 and 10:1 (by vol). Both of these mixes produced inferior results, with considerable stem rot, presumably due to excess water retained by the rooting medium (data not presented). The success of the subirrigation system appears to rely on use of perlite as a rooting medium. Perlite is a unique rooting medium as it holds only 25% of its volume as water, nearly all of which is available to the cutting (4). Grange and Loach (4) demonstrated that in perlite held at a volumetric water content of 20% cuttings absorbed water at 80% of the rate at which the same cuttings would absorb pure water. By comparison, in a medium of peat:perlite (1:1 by vol), also held at 20% volumetric water content, cuttings absorbed water at only 25% of the rate from pure water. Because perlite holds so little water by volume, it provides excellent aeration to the base of the cutting. Adequate aeration around the base of the cutting is critical to successful root initiation and growth (9, 10).

Use of sulfuric acid to acidify the subirrigation solution may not be a practical means of controlling solution pH for many propagators. Solution pH is stable for only a few days before it must be readjusted. A more practical means of maintaining a stable low pH may be to add milled sphagnum peat to the subirrigation solution.

Lee *et al.* (8) found that brief treatment with 2 N sulfuric acid improved rooting of stem cuttings of several ornamental woody species. Though it may be possible low solution pH facilitated IBA uptake in our experiments, as proposed by Harbage and Stimart (6), this must have occurred after auxin application, when cuttings were exposed to low pH from the subirrigation solution.

With further refinement, cutting propagation using subirrigation could be a viable and economical alternative to intermittent mist for commercial cutting propagation.

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