Effect of Endomycorrhizal Inoculum on Root Initiation and Development of *Viburnum dentatum* L. Cuttings[1]

Stephen D. Verkade and David F. Hamilton[2]
Department of Horticulture
Purdue University
West Lafayette, IN 47906

Abstract

Viburnum dentatum L. cuttings were inoculated with *Glomus fasciculatum* (Thaxter) Gerdemann and Trappe spores, hyphae and associated soil and root segments at 5 spores/cm³ of medium. Fresh weight of roots, number of roots, and mycorrhizal condition were determined on the fourth and each subsequent week for 5 additional weeks. Inoculation during propagation resulted in uniform mycorrhizal infection (30-50%) and increased root development. A positive interaction between mycorrhizal development and root initiation also was observed.

Index words: *Glomus fasciculatum,* rooting, vegetative propagation

Introduction

Mycorrhizae has been shown to increase growth of many plant species during production (4, 6, 7, 8, 13). However, efficient and economical inoculation techniques must be developed before the use of mycorrhizae can be used commercially in plant production.

Presently, most plants are inoculated individually at the time of potting. A more efficient time to inoculate could be during propagation when large numbers of plants can be treated with mycorrhizal inoculum in one operation.

Mycorrhizal inoculation of woody plants during propagation is a convenient and logical time in the production cycle to efficiently inoculate large numbers of plants at the earliest possible time. There is no other time in the production cycle during which the plant density is as high, resulting in maximum efficiency of inoculation.

Propagation by cuttings is common for production of woody plants, but some species are difficult to root. The presence of mycorrhizal inoculum in a rooting medium may promote root initiation and/or development through chemical or biological interaction of the mycorrhizal fungi with the cutting. Lindermann and Call (5) found that ectomycorrhizal inoculum of *Thelephora terrestris* Ehrh. ex. Fr. in the rooting medium of cuttings of *Arctostaphylos uva-ursi* L. Spreng (bearberry) and *Vaccinium ovatum* Pursh. (huckleberry) increased rooting percentage and root volume. Holden (3) confirmed the beneficial effect of unidentified mycorrhizal fungi on the root development and plant growth of bearberry. Bannister and Norton (1) found that unidentified endomycorrhizae did not promote the growth of *Calluna vulgaris* (L.) Hull (heather) cuttings under high nutrient regimes, but under low nutrient regimes dry weight of roots and shoots were greater for cuttings with mycorrhizal roots. Incorporation of the mycorrhizal fungi *Conocybe tenera* (Shaeff. ex Fr.) Fayod into the rooting medium also increased root weight of *Populus sp.* L. (poplar) cuttings (11). Verkade and Hamilton (12) found that extensive mycorrhizal development occurred on roots of *Ligustrum obtusifolium* var. *regelianum* (Koehne) Rehd. after 6 weeks of rooting in a medium inoculated with *Glomus mosseae* (Thaxter) Gerdemann and Trappe. This coincided with substantial increases in root development, but no effect on root initiation was found. The objectives of this experiment were to determine the effectiveness of mycorrhizal inoculation during clonal vegetative cutting, and the effects of mycorrhizae on root initiation and growth of *Viburnum dentatum.*

Materials and Methods

Terminal softwood *Viburnum dentatum* cuttings, 20 cm (7-7/8 in) long with six leaves, were collected in July. Basal ends of cuttings were treated with 0.1% W/W Indolebutyric Acid and rooted in a medium of perlite:vermiculite (1:1, by vol), amended either with pasterized potting soil [perlite:sphagnum peat moss:topsoil (2:2:1, by vol)] at a rate of 7 volumes of the rooting medium to 1 volume of potting soil, or with mycorrhizal inoculum at the same rate. The mycorrhizal inoculum consisted of roots of *Lycopersicon esculentum* Mill., spores and hyphae of *Glomus fasciculatum,* and potting soil [perlite:sphagnum peat moss:topsoil (2:2:1, by vol)]. This provided approximately 54,600 spores per treated flat (5 spores/cm³) as estimated at the time of inoculation by microscopic examination, using sucrose centrifugation to isolate spores. Cuttings were stuck in flats and placed under intermittent mist (12 seconds/8 minutes) in a greenhouse at 26°C day/22°C night ± 3°C (78.8°F day/71.6°F night) with approximately 25% shade.

Each treatment was replicated 4 times, with three subsamples within each replicate for each of 5 sampling

[1]Received for publication February 20, 1986; in revised form February 19, 1987. Journal Paper No. 9876 of the Agricultural Experiment Station, Purdue University, West Lafayette, IN 47907, U.S.A. Prepared as part of an investigation by the Joint Highway Research Project, Engineering Experiment Station, Purdue University, in cooperation with the Indiana State Highway Commission and the U.S. Department of Transportation Federal Highway Administration.

[2]Graduate Student and Associate Professor of Horticulture, resp. Current addresses: University of Florida, 3205 College Avenue, Fort Lauderdale, FL 33314; and American Association of Nurserymen, 1250 I Street, N.W., Suite 500, Washington, DC 20005.

Table 1. Effect of *Glomus fasciculatum* on number of root initials formed by cuttings of *Viburnum dentatum* harvested after 4, 5, 6, 7, or 8 weeks of rooting.

Week	Number of Root Initials	
	Inoculated	Non-inoculated Control
4	5.25 c[z]	2.25 c
5	33.75 a	8.25 c
6	33.50 a	25.00 ab
7	49.75 a	34.25 a
8	42.75 a	27.50 a

[z]Means followed by the same letter or letters are not significantly different according to the Student-Newman-Keuls' Test, at the 5% level.

Table 2. Effect of *Glomus fasciculatum* on root fresh weight of *Viburnum dentatum* cuttings after 4, 5, 6, 7, and 8 weeks of rooting.

Week	Root Fresh Weight (g)	
	Inoculated	Non-inoculated Control
4	0.0000 d[z]	0.0000 d
5	0.0083 d	0.0033 d
6	0.0000 d	0.0000 d
7	0.5091 b	0.2793 c
8	0.7736 a	0.2605 c

[z]Means followed by the same letter or letters are not significantly different according to the Student-Newman-Keuls' Test, at the 5% level.

dates, giving a total of 60 cuttings/treatment. Roots were sampled weekly for 5 weeks beginning with the fourth week after inoculation. At each sampling time, measurements included root fresh weights, number of root initials emerging from stem cuttings. Mycorrhizal development was determined by root staining and microscopic observation (2, 9). Data were analyzed by analysis of variance, using Student-Newman-Keul's test of significance (5% level) to separate means.

Results and Discussion

Inoculation increased the number of root initials emerging from stem cuttings 5 weeks after sticking, with smaller increases during subsequent weeks (Table 1). Roots of inoculated cuttings had between 30 to 50% of the cortical cells infected, while no mycorrhizal development was detected on roots of non-inoculated cuttings. Increased root initiation in inoculated plants occurred only after other roots had begun to form. Therefore, it is likely that this effect occurs only after infection and may be mediated through an effect of the fungus on plant metabolism, rather than an effect of a fungal exudate prior to infection (5, 10).

Mycorrhizal formation significantly increased root fresh weight after 7 weeks of rooting (Table 2). Several weeks of root development were necessary for differences in root growth to occur on cuttings.

Significance to the Nursery Industry

Mycorrhizal inoculum is relatively expensive to produce. However, inoculation of woody plants during propagation is convenient and it may be logical to efficiently inoculate large numbers of plants at this time. There is no other time in the production cycle during which the plant density is as high, resulting in efficient use of inoculum.

Literature Cited

1. Bannister, P. and W.M. Norton. 1974. The response of mycorrhizal and non-mycorrhizal rooted cuttings of heather (*Calluna vulgaris* (L.) (Hull) to variations in nutrient and water regimes. New Phytol. 73:81-89.

2. Gray, L.E. and J.W. Gerdemann. 1973. Uptake of sulfer-35 by vesicular-arbuscular mycorrhizae. Plant Soil 39:687-689.

3. Holden, V.L. 1978. The use of mycorrhizae in the propagation of *Arctostaphylos uva-ursi.* Proc. Intern. Plant Prop. Soc. 28:132-133.

4. Johnson, C.R., J.N. Joiner, and C.E. Crews. 1980. Effects of N, K and Mg on growth and leaf nutrient composition of three container grown woody ornamentals inoculated with mycorrhizae. J. Amer. Soc. Hort. Sci. 105:286-288.

5. Linderman, R.G. and C.A. Call. 1977. Enhanced rooting of woody plant cuttings by mycorrhizal fungi. J. Amer. Soc. Hort. Sci. 102:206-208.

6. Maronek, D.M., J.W. Hendrix, and J. Kiernan. 1980. Differential growth responses to the mycorrhizal fungus *Glomus fasciculatus* of southern magnolia and bar harbor junipers grown in containers in composted hardwood bark-shale. J. Amer. Soc. Hort. Sci. 105:206-208.

7. Menge, J.A., E.L.V. Johnson, and R.G. Platt. 1978. Mycorrhizal dependency of several citrus cultivars under three nutrient regimes. New Phytol. 81:553-559.

8. Menge, J.A., J. LaRue, C.K. Labananskas, and E.L.V. Johnson. 1980. The effect of two mycorrhizal fungi upon growth and nutrition of avocado seedlings grown with six fertilizer treatments. J. Amer. Soc. Hort. Sci. 105:400-404.

9. Phillips, J.M. and D.S. Hayman. 1970. Improved procedures for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. Trans. Br. Mycol. Soc. 55:158-161.

10. Slankis, V. 1973. Hormonal relationships in mycorrhizal development. In: Ectomycorrhizae, Their ecology and physiology. G.C. Marks and T.T. Kozlowski, Eds. Academic Press, New York. pp: 232-298.

11. Veldemann, R. 1981. Influence of mycorrhizae on the rooting of poplar cuttings. Mendedelingen Van De Faculteit Landbouwwetenschappen Rijksuniversiteit Gent. 45(2):169-173 (Abstr.).

12. Verkade, S.D. and D.F. Hamilton. 1980. Mycorrhizae and their uses in the nursery. Proc. Intern. Plant Prop. Soc. 30:353-363.

13. Verkade, S.D. and D.F. Hamilton. 1983. Effects of soil fertility on growth, nutrient concentration and mycorrhizal development of *Liriodendron tulipifera* seedlings inoculated with the vesicular-arbuscular fungus, *Glomus fasciculatus.* Sci. Hortic. 21:243-252.

14. Wick, R.L. and L.D. Moore. 1983. Japanese holly: A host for endomycorrhizal fungi. J. Environ. Hort. 1:12-14.