Propagation and Acclimatization of 'Norton' Grapevine©

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Difficulty is often experienced by commercial propagators in propagating the 'Norton' grapevine (*Vitis aestivalis*, also known as 'Cynthiana') by cuttings, the normally employed method of grapevine multiplication. Therefore, we endeavored to propagate 'Norton' by micropropagation. Cultures were readily established in vitro by placing axillary buds taken from 3-year-old greenhouse-grown potted vines on Murashige and Skoog medium supplemented with 4 µM benzyladenine (BA) and 0.5 mg thiamine per liter. When in vitro-derived axillary buds were cultured on various levels of BA and naphthaleneacetic acid (NAA), excellent multiplication was obtained when 4 to 8 µM BA was included in the medium. Naphthaleneacetic acid had little effect on shoot number or number of nodes per shoot. Plantlets were simultaneously acclimated and rooted ex vitro in rehydrated compressed peat pellets (Jiffy-9). Rooting was nearly 100% and over 90% of rooted microcuttings were successfully established in the greenhouse and later transferred to the field.

INTRODUCTION

The American hybrid *Vitis* sp. 'Norton' is a premium wine grapevine for use in the central Midwest (Reisch et al., 1993). It has several desirable characteristics, but difficulty associated with propagation has limited its use in vineyards (Tarara and Hellman, 1991). Grapevines are traditionally propagated from cuttings of dormant 1-year-old canes (Hartmann et al., 2002). Propagation of 'Norton' through this method has proven difficult because cuttings root poorly (Avery, 1999). In vitro propagation offers another method of increasing plant material for this cultivar. Micropropagation of 'Norton' has previously been reported (Norton and Skirvin, 2001). The goal of this project was to improve in vitro plantlet quality and acclimate plantlets without an in vitro rooting step. Potential benefits of this research include increased plant material at a lower cost for growers and increased availability of this grape for wineries.

MATERIALS AND METHODS

Greenhouse-grown 3-year-old potted plants with actively growing shoots (30–50 cm in length) were used as explant source material. Axillary buds (0.5 cm × 0.5 cm) were excised, surface disinfested for 15 min in 10% commercial bleach solution and washed three times for 5 min in sterile water. Explants were placed in 25-mm culture tubes containing 10 ml Murishige and Skoog (MS) medium supplemented with 4 µM 6-benzyladenine (BA) and 0.5 $mg \cdot L^{-1}$ thiamine and gelled with 7.5 $g \cdot L^{-1}$ Bacto-Agar. Established cultures were transferred monthly to fresh medium before experiments were begun. Explants were propagated by placing two-node segments, with leaves removed, horizontally on the medium. Cultures were maintained at 23±1 ºC for 16 h per day under cool white fluorescent light (28 $\mu mol \cdot s^{-1} \cdot m^{-2}$).

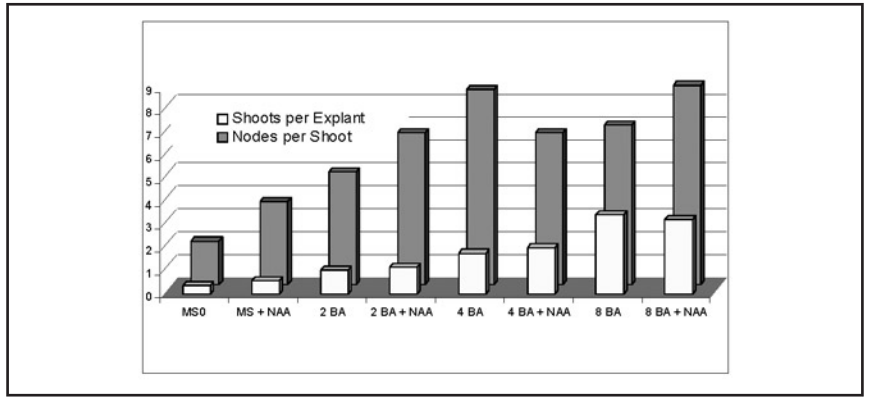

Figure 1. Effect of cytokinin and auxin on the number of shoots per explant and nodes per shoot.

In Vitro Propagation and Multiplication. Murishige and Skoog medium supplemented with BA (0, 2, 4, or 8 μM), naphthaleneacetic acid (NAA) (0 or 0.5 μM), 0.5 mg·L^{-1} thiamine and gelled with 7.5 g·L^{-1} Bacto-Agar (pH 5.6±0.1) was used in all multiplication experiments. Explants were incubated as described above.

Ex Vitro Acclimation. Twenty-five microcuttings from 4- to- 6 week-old cultures with or without a five second dip in1000 ppm (0.1%) indolebutyric acid (IBA) were transferred to hydrated peat pellets in sundae cups with lids for rooting and acclimation. After 4 days, plantlets and peat pellets were planted in potting medium in 1-L plastic pots in the greenhouse under partial shade. Sundae cup lids were used to maintain plantlet humidity. Removing the lids gradually reduced humidity and after four weeks the plantlets were fully acclimated.

RESULTS AND DISCUSSION

Multiplication. The effects of cytokinin and auxin, on number of shoots per explant and number of axillary buds per shoot are presented in Fig. 1. Auxin did not have an effect on either the number of shoots per explant or the number of nodes per shoot. The interaction between cytokinin and auxin was not significant.

Acclimation. Fifty plantlets were transferred ex vitro for acclimation. After 4 weeks, 92% (23 of 25), and 88% (22 of 25) of the plantlets, respectively, survived and were growing vigorously. Plantlets rooted quickly and produced 3–7 primary roots (data not shown).

CONCLUSIONS

- Establishment of in vitro cultures was successful on MS medium containing 4 μM BA and 0.5 mg thiamine per liter.
- Excellent multiplication was obtained when in vitro-derived axillary buds were cultured on MS medium containing 4 to 8 μM BA.

- Microcuttings were simultaneously rooted and acclimated ex vitro in rehydrated Jiffy-9 peat pellets in plastic containers conferring high humidity.
- Over 90% of rooted microcuttings were successfully established in the field.

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LITERATURE CITED

Avery, J.D. 1999. Propagation of Norton-Cynthiana. Proc. 14th Annual Midwest Regional Grape and Wine Conf. pp.1-6.

Hartmann, H.T., D.E. Kester, and **F.T. Davies, Jr.** 2002. Plant propagation: principles and practices. Prentice Hall, Englewood Cliffs, New Jersey.

Norton, M.A. and **R.M. Skirvin.** 2001. Micropropagation of 'Norton' winegrape. HortTechnology. 11(2):206-208.

Reisch, B.I., R.N. Goodman, M.H. Martens, and **N.F. Weeden.** 1993. The relationship between Norton and Cynthiana, red wine cultivars derived from *Vitis aestivalis*. Am. J. Enol. Vitic. 44(4):441-444.

Roberts, P.L. 1999. This hill, my hand, Cynthiana's wine. Resonant Pub., Baltimore, Maryland.

Rombough L.J. The grape grower. Chelsea Green Pub. Co., White River Junction, Vermont.

Tarara, J.M. and **E.W. Hellman.** 1991. 'Norton' and 'Cynthiana'— premium native winegrapes. Fruit Var. J. 45(2):66-69.

Williams P.L. and **A.J. Antcliff.** 1984. *Vitis berlandieri* and *Vitis cinerea* from hardwood cuttings. Am. J. Enol.Vitic. 35(2):75-76.