



## Propagation and Cultivation of 'Ōhelo

Francis Zee, Amy Strauss, and Claire Arakawa

U.S. Department of Agriculture, Agricultural Research Service, Pacific Basin Agricultural Research Center,  
Tropical Plant Genetic Resources Management Unit, Hilo, Hawai'i

'Ōhelo (*Vaccinium reticulatum* Smith) is a small, native Hawaiian shrub in the cranberry family, commonly found in disturbed, open sites at 2000 to 12,000 feet (640 to 3700 m) elevation on Maui and Hawai'i. People frequently scour the landscape where it occurs, disrupting fragile habitats to harvest its delectable berries for use in jam, jelly, and pie filling. This impact on delicate environments might be reduced if 'ōhelo berry could be cultivated and marketed to meet demand for the fruit. This publication describes nursery and field procedures for growing out 'ōhelo seedlings and gives protocols for vegetative propagation of 'ōhelo both from cuttings and by tissue culture.

The study was conducted in a field and greenhouse at the University of Hawai'i's Volcano Agricultural Research Station. The site is at 4000 feet (1333 meters) elevation, with an average annual maximum temperature of 73°F, range 68–78° (23°C, range 20–26°), an average minimum temperature of 47°F, range 36–53° (8°C, range 2–12°), and average annual rainfall of 84 inches (210 cm).

In January 2005, berries were collected (Photo 2) and seeds were extracted using a blender set at medium speed and a high water-to-berry ratio. The seeds were air-dried on

paper towels for two days at ambient temperature. 'Ōhelo seeds are very small (100 seeds weigh 0.1 gram). Fresh seeds germinated 40–45 days after sowing into a 1:1:1 mixture of peat, vermiculite (Therm-o-rock® size #2), and perlite (Pahroc® Giant size #3) in a greenhouse at 60–80 percent shade (Photo 3). A germination test comparing eight media (Table 1) confirmed hāpu'u (Hawaiian tree fern) bark to be a good germination medium (Obata 1967, 1973), but this medium may be too wet, as there was higher seedling disease incidence after transplanting. The 1:1:1 mixture of peat, vermiculite, and perlite produced good germination of fresh seeds, and the seedlings grew well when transplanted to a 1:1:2 mixture at four months after germination. 'Ōhelo seeds stored at 39°F (4°C) lost viability after a year.



**1. 'Ōhelo is an attractive native Hawaiian shrub with potential as an ornamental.**

### Seedling care and maintenance

Four-month-old 'ōhelo seedlings were transplanted to 2-inch (5-cm) pots containing 1:1:2 peat, vermiculite, and perlite (Photo 4). Each pot was side-dressed with 14-14-14 slow-release Nutricote® fertilizer (1/8 tsp, 2 g). The plant foliage was sprayed every 2 weeks with a mild dilution of 10-30-20 foliar fertilizer (1/4 tsp per gallon, 0.4 g/liter). Seedlings were tip-pruned at transplanting to encour-



2. 'Ōhelo berries collected for seed.



3. 'Ōhelo seedlings germinated in 1:1:1 peat-vermiculite-perlite medium in a greenhouse with 60% shade.

**Table 1. Germination count of 'ōhelo seeds on various media after 45 days under 60% shade.**

Medium	Germination per 200 seeds
Hāpu'u dust	142
Vermiculite-perlite-peat 1:1:1	117
Cinder-soil 2:1	0
Peat-perlite 1:1	0
Soil (from Volcano Research Station)	75
Oasis® cube in Sunshine® mix	132
Sphagnum moss on cinder	62
Sifted fine black cinder	0



4. 'Ōhelo seedlings transplanted into individual pots.

age multiple branching. Well shaped 'ōhelo plants with a compact crown of reddish new growth are appealing as ornamentals (Photo 1).

At 10 months old (Nov 30, 2005), the seedlings were transplanted into 1-gallon (4-liter) pots containing the 1:1:2 medium and grown for 6 months more before being transplanted to the field. The plants were maintained on benches in a 20 x 20-foot portable canopy house with a clear plastic top and sides covered with ½-inch (1.25-cm) bird netting.

A leaf roller tentatively identified as the Mexican leaf roller, *Amorbia emigratella* Busck, damaged tender new shoots (Photo 5) and was readily controlled with bio-

logical-based insecticides such as Dipel® and XenTari®. Aphids were found infrequently on young 'ōhelo plants and caused little or no damage. Powdery mildew caused severe defoliation on young 'ōhelo plants if not controlled (Photo 6).

#### Field-planting 'ōhelo seedlings

Sixty seedlings were field-planted on June 12, 2006, in a 45 x 20-foot (15 x 7-meter) field plot (Photo 7). Three 5 x 2-foot (15 x 1-m) rows were prepared within the plot. The rows were amended as follows:

- Row 1 was amended with 3 cubic feet (0.08 m<sup>3</sup>) of steer manure, 18 lb (8.2 kg) treble superphosphate (0-





5. Mexican leaf roller, *Amorbia emigratella* Busck, on an 'ohelo seedling.



8. 'Ohelo berries harvested from the field planting.



6. 'Ohelo seedling with powdery mildew damage.



7. 'Ohelo field (planted on Oct 3, 2007) 16 months after field planting. Row 1, with larger plants, is on the left.

41-0), and 12 lb (5.4 kg) each of gypsum, ammonium sulfate, and potassium magnesium sulfate (0-0-22-22S K-mag<sup>®</sup>).

- Row 2 was amended as in Row 1, plus 3.8 cu ft (0.11 m<sup>3</sup>) of peat.
- Row 3 was amended as in Row 1, plus 1 cu yard (0.76 m<sup>3</sup>) of sifted black cinder.

Irrigation tubing ½ inch (1.27 cm) in diameter was installed 18 inches (45 cm) apart in each row and covered with weed mat. The plot was watered for 45 minutes every other day for a week prior to planting. Twenty holes were cut into the weed mat at 2-foot (0.7 m) spacing. One month after field planting, each plant was top-dressed with 1 oz (28 g) of Nutricote<sup>®</sup> 13-13-13 slow-release fertilizer. All plants received monthly foliar fertilizer at 1 tsp per gallon (1.8 g/liter), alternating between Miller<sup>®</sup> 4-41-27 and Total-GRO<sup>®</sup> Azalea-Acid 21-7-7.

In Rows 2 and 3, seedlings gradually declined 3 months after field planting; by the end of the first year, three and four plants had died in Rows 2 and 3, respectively. The canopies of the surviving plants in Rows 2 and 3 were sparse, with visible branch dieback (Photo 7). All plants in Row 1 were thriving a year after field planting, with thick, well formed canopies, and were producing berries (Photo 8); no serious insect, disease, or bird damage was observed throughout the field planting.

### Propagating 'ōhelo by cuttings

Healthy, upright, woody branches were harvested from selected 'ōhelo bushes for propagation. Each cutting consisted of a 2-inch-long internode below a whorl of intact leaves. The bottom of each cutting was dipped in Hormex® #1 rooting powder and stuck into a 2 x 2-cm Oasis® root cube that was well watered and drained prior to use. Three rows of rooting cubes were placed in a tray with 1-inch (2.5-cm) spacing between rows to maintain

moist but not wet conditions (Photo 9). The cuttings were placed in a greenhouse with 60 percent shade and protected from wind and heat. Overhead misting was set at two times a day for 5 minutes each. Sixty percent of the 'ōhelo cuttings rooted in 3 months (Photo 10). Rooted cuttings were transplanted into 4-inch (10-cm) pots containing the 1:1:2 medium (Photo 11). Plants propagated by cuttings were hardy and vigorous and flowered 8 months after transplanting.



9. 'Ōhelo propagation by cuttings.



11. Rooted 'ōhelo cuttings transplanted into individual pots.



10. Rooted 'ōhelo cutting in an Oasis® block.



### Tissue-culturing 'ōhelo

Selected 'ōhelo seedlings were initiated into tissue culture for preservation and multiplication.

#### Disinfection procedure

1. Select healthy young shoots with buds.
2. Remove leaves and trim to 1-cm pieces with one to three nodes.
3. Wash in detergent and tap water.
4. Soak cuttings in a solution of 15% Clorox® (v/v) in sterile distilled water with 2 drops of Tween 20® for 15 minutes; decant solution.
5. Soak cuttings in a solution of 10% Clorox (v/v) in sterile distilled water with 2 drops of Tween 20 for 15 minutes; decant solution.
6. Dissect shoots in a solution of 5% Clorox (v/v) in sterile distilled water with 2 drops of Tween 20 to prepare single axillary bud explants of 1–3 mm size.
7. Place explants into a solution of 5% Clorox (v/v) in sterile distilled water with 2 drops of Tween 20 for 5 minutes; decant solution.
8. Soak explants in a solution of 1% Clorox (v/v) in sterile distilled water with 2 drops of Tween 20 for 60 minutes; decant solution.
9. Rinse explants twice with sterile distilled water.
10. Transfer individual explants into individual tubes of initiation medium.

#### Composition of media for different stages of 'ōhelo tissue culture

The *base medium* is composed of modified WPM (Lloyd and McCown 1980) with 0.4 mg/liter of thiamine-HCl, 100 mg/liter myo-inositol, 30 g/liter sucrose, and 7 g/liter agar (Difco Bacto®). MS-based vitamins include (in mg/liter) 2.0 glycine, 0.5 nicotinic acid, 0.5 pyridoxine.HCl, and 0.1 thiamine-HCl (Murashige and Skoog 1962); pH of the medium is adjusted to 5.2 before autoclaving.

The *initiation medium* is the base medium plus 4 mg/liter zeatin (Read and Abdelnour-Esquivel 1991; Read and de Paz, personnel communication, 2005).

The *growth/multiplication medium* is the base medium plus 2–5 mg/liter 2iP.

The *rooting medium* is the base medium plus 2 mg/liter IBA.

The *maintenance medium* is the base medium without added growth regulators.



12. 'Ōhelo in vitro culture.

All cultures were maintained under cool-white fluorescent lamps for 16 hours per day at 73°F (23°C). After initiation, explants were transferred into fresh medium every 14 days until leaves developed (Photo 12). For multiplication, explants were transferred to the growth/multiplication medium every 14–21 days. For more plantlet production, stem cuttings (with two to three nodes) from elongated stems in culture were harvested and placed onto growth/multiplication medium to encourage axillary bud breaks. It takes approximately 6–8 weeks for cuttings to develop roots in the rooting medium. Once rooted, cuttings were planted out in a greenhouse.

#### Planting out tissue-cultured 'ōhelo in the greenhouse

Tissue-cultured 'ōhelo with well formed roots were removed from the vessels, washed clean of medium in tap water, and soaked in a bath of Superthrive® solution at 1/8 tsp per gallon (0.16 ml/liter) before planting in the 1:1:2 potting medium in 2-inch (5-cm) pots. The plants were watered thoroughly and placed under 60–80 percent shade with overhead misting set for 5 minutes every other day. Most transplants without roots died within the first month after transplanting. All plantlets were protected from wind and heat, and from being too dry or too wet.

Two weeks after planting, 1/8 tsp (2 g) of 13-13-13 Nutricote® was added to each pot, and once every month the plants were sprayed with a 1/2 tsp/gallon (0.9 g/liter) solution of 21-7-7 or 4-41-27 foliar fertilizer. In 3 months,

well rooted plants were transplanted into the 1:1:2 potting medium in 4-inch (10 cm) pots. When established, the tissue-cultured 'ōhelo plants were vigorous, but they were as susceptible to leaf rollers, powdery mildew, and aphids as the seedlings. 'Ōhelo plants propagated by cutting or tissue culture flowered about 8 months after transplanting.

### Conclusions

'Ōhelo seeds germinated readily under 60–80 percent shade in a well watered and well drained potting mixture of peat, vermiculite, and perlite. Seedlings younger than 3 months were sensitive to too much or too little watering in the nursery, but hardiness and vigor improved with age. Seedlings responded well to foliar fertilizers and slow-release fertilizers. Some seedlings flowered 10 months after germination, much sooner than the 5 years reported in the literature (Vander Kloet 1993, Wagner 1990). Sixteen-month-old seedlings were successfully field-planted in this study and produced berries the following year. Minimal soil amendments in the planting row resulted in better plant growth and survival compared to rows with added peat or cinder. This preliminary trial demonstrated successful vegetative propagation and field cultivation of 'ōhelo for the first time in Hawai'i. More studies should follow to determine the economic potential of 'ōhelo as an ornamental potted plant and demonstrate the potential of small-scale production of 'ōhelo berries, which can serve as a conservation measure to reduce impacts of harvesting the fruits from natural 'ōhelo habitats.

### Literature cited

- Murashige, T., and F. Skoog. 1962. A revised medium for rapid growth and bioassays with tobacco tissue cultures. *Physiology Plantarum*, 15:473–497.
- Obata, J.K. 1967. Seed germination in native Hawaiian plants. *Newsletter of the Hawaiian Botanical Society* 6(3):13–20.
- Obata, J.K. 1973. Propagating native Hawaiian plants (continued from February 1973). *Newsletter of the Hawaiian Botanical Society* 12(2):9–11.
- Vander Kloet, S.P. 1993. Biosystematic studies of *Vaccinium* section *Macropelma* (Ericaceae) in Hawaii. *Pacific Science* 47(1):76–85.
- Wagner, W.L., D.R. Herbst, and S.H. Sohmer. 1990. *Manual of the flowering plants of Hawai'i*. Bishop Museum Special Publication 83. University of Hawai'i Press and Bishop Museum Press, Honolulu. p. 593–595.
- Read, B.M., and A. Abdelnour-Esquivel. 1991. The use of zeatin to initiate in vitro cultures of *Vaccinium* species and cultivars. *HortScience* 26(10):1320–1322.
- Read, B.M., and Janine de Paz, personal communication. October 2005, Corvallis, Oregon.

### Acknowledgments

The authors thank Michael Tokura-Ellsworth, Russell Kai, Carol Riley, Kert Hamamoto, and Jason Okamoto, USDA/ARS PBARC, for their excellent technical assistance and suggestions. Thanks also go to Rhonda Loh of Hawai'i Volcanoes National Park and to CTAHR's Melvin Nishina for constructive review and suggestions and to Dale Evans for editing and production assistance.

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