Propagation protocol for

Jacquemontia reclinata House, a federally endangered species of South Florida

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ABSTRACT

Beach jacquemontia (*Jacquemontia reclinata* House [Convolvulaceae]) is an endangered endemic plant from southeast Florida that can be propagated by seeds or cuttings. Freshly harvested seeds from cultivated and wild plants can germinate easily in a greenhouse under South Florida ambient conditions. A higher germination success after short-term seed storage suggests an after-ripening effect; however, orthodox methods are acceptable for long-term storage. Soaking treatments do not affect overall germination. Cuttings can produce new plants when treated with rooting hormone and planted in perlite. Arbuscular mycorrhizae fungi inoculation of cultivated plants is recommended for outplanting into habitats lacking natural fungal inoculum. These propagation techniques have yielded plants used for reintroduction and recovery activities of the species.

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KEY WORDS

coastal dune, coastal strand, Convolvulaceae, *ex situ* collection, soaked seeds, storage behavior

NOMENCLATURE

USDA NRCS (2005)

Figure 1. Jacquemontia reclinata House (Convolvulaceae) blooming in Bill Baggs Cape Florida State Park, Florida. Photo by Samuel J Wright

Jacquemontia Choisy is a tropical and subtropical American genus that belongs to the morning-glory family, Convolvulaceae. There are 80 to 100 species in this genus, and *J. reclinata* House, also known as beach clustervine or beach jacquemontia, is closely related to 3 other threatened or endangered jacquemontias that occur in southern Florida: *J. curtisii* Hallier, *J. havanensis* (Jacq.) Urban, and *J. pentanthos* (Jacq.) G. Don. *Jacquemontia reclinata* can be distinguished from the other jacquemontias by the presence of tiny hairs along the margins of its outer sepals and by its rather broad succulent leaves (Austin 1979).

Jacquemontia reclinata (Figure 1) is a perennial vine with a stem branched at the base and a stout taproot (Small 1934), although we have also observed 1.4 cm (0.56 in) diameter and < 2 m (6.6 ft) long roots protruding in a radial direction and a fibrous root mass (~15 x

10 cm [6 x 4 in]) close to the surface (Figure 2). Branches are slender and elongate, widely radiating, woody at the base, and may twine over other plants (Small 1934; Robertson 1971). The leaves are entire, spirally arranged, more or less succulent, and finely pubescent, at least when young (Small 1934). *Jacquemontia reclinata*'s white to light-pink flowers may be present from November to May (Robertson 1971) or year-round (Austin and others 1991). Twenty-two species of insects—mostly small bees and bee flies, and less frequently wasps, butterflies, and ants—are known to visit the flowers and may pollinate them (Pinto-Torres 2004). Few seedlings or young plants are found near adults in the wild. Although rarely observed, seedlings are typically found in the shade of adjacent shrubs and trees, and they send lateral stems from their rootstocks into more exposed areas when maturity is reached.

Currently, there are 10 known populations of *J. reclinata*, which range in size from 1 to 245 plants. These populations are within a 145-km (90-mi) stretch of coastline in southeast Florida, between Dade and Palm Beach counties (Wunderlin 1998; USFWS 1999; Coile 2000). Within its range, *J. reclinata* is found on disturbed or open sunny areas of coastal sand dunes, mostly on the crest and lee side (Austin 1979), and on open areas of maritime hammocks (Robertson 1971) from 1 to 9 m (3.3 to 29.7 ft) above sea level (Maschinski and others 2003). It may also invade and restabilize the above habitats after a tropical storm, hurricane, or fire disturbance (USFWS 1999).

The primary habitat of this species has been destroyed by commercial and residential development, including parking lots and picnic areas, and by beach erosion (USFWS 1999). In some locations, *J. reclinata* is negatively affected by competition with native and nonnative species (USFWS 1999). About 700 individuals remain in the wild, most of which (79%) occur in 3 populations (Maschinski and others 2005). The small geographical range, habitat fragmentation, low recruitment rate, and small population sizes coupled with stochastic events, seriously threaten *J. reclinata*'s persistence. These factors led to listing *J. reclinata* as federally endangered in 1993.

Fairchild Tropical Botanic Garden has developed propagation methods to aid in conservation and recovery efforts of *J. reclinata.* Knowledge of germination and propagation protocols is required to produce plants for reintroduction and augmentation, as well as to establish and maintain *ex situ* populations of living plants and seeds. Here we present our standard propagation conditions for *J. reclinata* based on the results of seed and vegetative propagation experiments.

SEED PROPAGATION

Jacquemontia reclinata fruits suitable for propagation are light brown-colored capsules about 4 to 5 mm (0.16 in) long, generally with 4 seeds (2.5 to 3 mm [0.11 in] long) per capsule (Robertson 1971), which are shed throughout the flowering season. Our experience is that there are 343 seeds per gram.

From our work, we have discovered several things about *J. reclinata* seeds that help us achieve more successful germination. First, we must ascertain seed source because different germination behavior and requirements and storage effects have been identified between cultivated and wild seedlots (see sidebars), as is the case with other species (Rojas-Arechiga and others 2001; Singh and Deswal 2002). Second, seeds from cultivated and wild plants can be stored successfully under a variety of conditions, and short-term storage can actually increase germination (see storage sidebar). Our preferred storage technique for seeds from cultivated plants is 3 mo at 12 °C (54 °F) and from wild plants is 1 mo at ambient conditions (23 °C [73 °F] and 60% to 84% relative humidity). For a longer storage time (for example, 42 mo), we recommend using a –20 °C (–4 °F) freezer. Third, the yellow pigment released from seeds during soaking does not appear to affect germination, nor does soaking seeds in fresh water or salt water (see germination sidebar).

Germination of seeds is easily attained on commercially available soilless media (for example, Metromix 360, VJ Growers Supply, Apopka, Florida) or Fairchild's standard potting mix (10% perlite, 40% Canadian peat, 20% composted bark, and 30% sand). In pots, soil is gently tapped down to create a flat surface and then watered. After the excess water drains, nonsoaked seeds are sown and top-dressed with about 1 cm (0.4 in) of chicken grit or sand. The top-dressing grit reduces compaction from watering, reduces moss and weed growth, and prevents seeds from washing out of the pot (Carrara 2005). Adding pea-sized rock (The Bushel Stop, Lake Worth, Florida) to the medium in a 1:1 ratio (v:v) increases aeration and drainage and reduces plant mortality by the root rot pathogen *Fusarium solari.* To minimize plant roots intertwining and then being disturbed during transplanting, we prefer pots over flats and transplant seedlings before they are 9 wk of age.

Once sown, pots are placed on a greenhouse bench that receives 30% sun under South Florida ambient conditions (25 °C to 35 °C [77 °F to 95 °F], 54% to 87% relative humidity, ~11 to 12 hours photoperiod) and watered 3 times per wk. Seeds germinate readily under these greenhouse conditions, indicating the lack of primary dormancy in this species. *Jacquemontia reclinata* seeds do not require light to germinate; however, we recommend the use of light because seeds exposed to light have more uniform germination. We found that 1-y-old seeds placed on filter paper in Petri dishes, exposed to 30% sunlight or kept dark by wrapping the dish with aluminum foil, and placed under greenhouse conditions described above, yielded the same germination (40%) after 15 d.

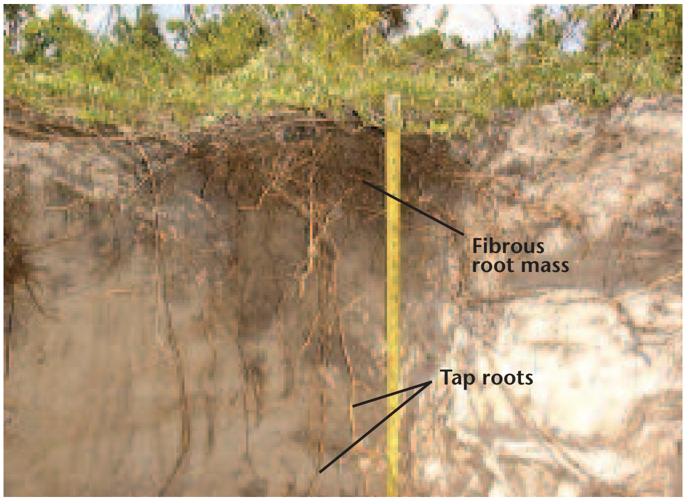

Figure 2. Jacquemontia reclinata root system. Photo by Samuel J Wright

STORAGE

By monitoring germination of freshly harvested seeds from cultivated individuals, we found germination commenced 1 wk after sowing, and average percentage germination ranged from 45% to 93%, whereas seeds from wild plants had an average percentage germination of zero to 85%. We found that seeds from cultivated and wild plants stored 1 mo at ambient conditions had average emergence of 25% and up to 91%, respectively. Seeds from cultivated plants cold-stored (12 °C [54 °F]) for 1 mo had 35% emergence. Seeds from cultivated plants given 3 mo of ambient or cold storage emerged at 76% and 81%, respectively, suggesting an after-ripening effect. Conversely, the highest emergence from seeds from wild plants after 3 mo of storage at 23 °C (73 °F) was only 35%. Under much longer storage times (61 to 62 mo) at 23 °C (73 °F), germination of seeds from cultivated plants dropped to 19% to 42%, consistent with the hypothesis that viability declines over time in all species (Baskin and Baskin 1998). However, we discovered that *J. reclinata* seeds are orthodox and can be successfully stored at –20 °C (–4 °F), yielding germination rates of 50% to 90%. Seeds from cultivated plants having moisture contents of 5%, 12%, and "fresh" (harvest time moisture content), which are then stored at –20 °C, 12 °C, and 23 °C for 42 mo, had germination ranging from 46% to 97%, but none of the treatment combinations were significantly different. Therefore, we packaged 620 seeds in foil envelopes (Barrier Foil Products, UK), sealed them with a hot iron (Walters 2004), and sent them for long-term freezer storage at the National Center for Genetic Resources Preservation in Fort Collins, Colorado.

VEGETATIVE PROPAGATION

Seeds may not always be available because of seasonality. Also, because of changing environmental factors (for example, canopy closure), existing *J. reclinata* habitat may not have suitable conditions for flowering and seed production. Because of these factors, genetic preservation of declining populations may be possible only through clonal propagation. Therefore, it is critical to develop vegetative propagation methods.

Immediately after removing cuttings from the donor plant, which can be done at any time of the day, we dip them first in tap water and then in Rootone® (GardenTech, Lexington, Kentucky), wrap them in a wet paper towel, enclose them in a Ziploc® bag (SC Johnson, Racine, Wisconsin), and place them in an ice-filled cooler until arrival at the nursery facilities. At the nursery, we stick cuttings into perlite, which our research showed would yield a 73% survival rate, whereas sticking cuttings into 3:1 (v:v) Fairchild potting mix (described above) to perlite medium only yielded a 20% survival rate after 6 mo.

LIGHT REQUIREMENT FOR SEEDLING GROWTH

We grow seedlings and cuttings in full sun under the non-limiting soil fertility and moisture conditions of the nursery. Under full sun in these conditions, plants accumulate nearly twice as much root dry mass as plants grown in shade, even though shoot extension rate, total stem length, and shoot dry weight is unaffected by the level of shade (Wright and Fidelibus 2004). Plants with higher root-to-shoot ratios might be more resistant to environmental stresses such as drought, fire, and herbivory.

MYCORRHIZAL REQUIREMENT

Wild *J. reclinata* plants have typical *Arum*-type arbuscular mycorrhizal fungi (AMF) in their roots. AMF significantly increase the dry weight and total phosphorus content of seedlings grown on native soil under greenhouse conditions (Fisher and Jayachandran 2002). *Jacquemontia reclinata* is not an obligate mycotroph because additions of phosphate without AMF also promoted seedling growth (Fisher and Jayachandran 2002). Therefore, for our experiments, we do not add AMF to our pots. AMF inoculation of cultivated plants, however, would be desirable for outplanting into habitats free of natural AMF inoculum, such as coastal dredge fills or restoration of former parking lots or building areas.

SUMMARY

- Seeds germinate in pots with commercially available soilless media placed on a greenhouse bench receiving 30% sun under South Florida ambient conditions.
- Freshly harvested seeds from cultivated and wild plants can germinate easily and a short-term storage period of up to 3 mo can increase germination success. For long-term storage, seeds may be frozen and maintained using orthodox methods.
- Seed-soaking treatments in fresh tap water are unnecessary to achieve germination, and seeds are salt tolerant.
- Cuttings can produce new plants when treated with rooting hormone and planted in perlite.
- Arbuscular mycorrhizae fungi inoculation of cultivated plants is recommended for outplanting into habitats free of natural fungal inoculum.

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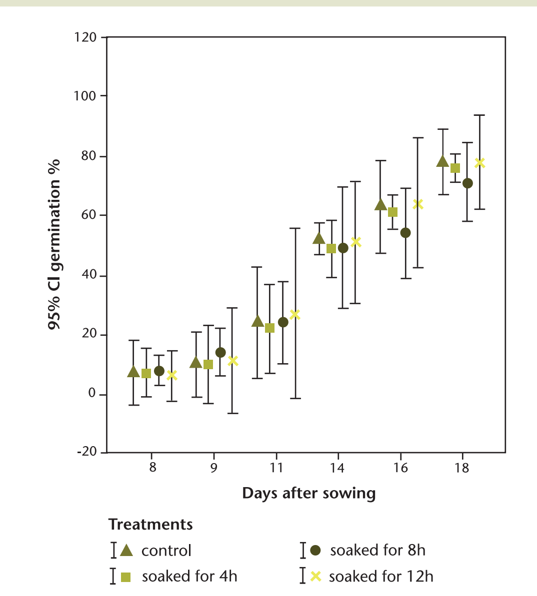
GERMINATION

Although *Jacquemontia reclinata* seeds release a yellow pigment when soaked in water, our work indicates that this chemical does not seem to inhibit germination. Seeds from Fairchild's *ex situ* plant collection, soaked in 10% bleach for 20 min and rinsed with distilled water, were soaked in fresh water for 0 (control), 4, 8, and 12 h or in full-strength seawater for 0, 2, 8, and 24 hours in an incubator (New Brunswick Scientific, Edison, New Jersey) to simulate waves. After soaking, seeds were placed on filter papers in Petri dishes on a greenhouse bench under the conditions described on p 302. All fresh water soaking durations had approximately 75% germination after 18 d. Further, soaking seeds up to 24 h in seawater did not reduce germination percentage or rate compared with that of nontreated seeds, indicating seawater probably does not inhibit germination of *J. reclinata* in the seedbank.



Effect of soaking *J. reclinata* seeds in fresh water for 0, 4, 8, and 12 h, on germination percentage. Four replicates per treatment with 25 seeds each. Repeated measures ANOVA revealed no interaction between soaking treatment and time after sowing ($F = 0.386$, $df = 15$, $P = 0.978$). Bars represent 95% confidence interval for mean.

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