Plant Propagation for the Breeding Program at Chicago Botanic Garden©

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INTRODUCTION

The Chicago Botanic Garden, The Morton Arboretum, and the nursery consortium Ornamental Growers' Association of Northern Illinois (OGA) are the corporate members of the Chicagoland Grows®, Inc., plant introduction program. Founded in 1986, the introduction program is dedicated to the development, testing, and introduction of landscape plants to the industry, gardens of the Midwest, and comparable climates in the USA, Canada, and Europe. In support of Chicagoland Grows, Chicago Botanic Garden initiated a perennial plant breeding program in 1995, which has developed to date the Prairieblues™ false lupins, the Meadowbrite™ coneflowers, and other selections.

Plant propagation at Chicago Botanic Garden supports the breeding program in two general ways. The breeding program produces seed from controlled crosses, which are then germinated in-house and grown on for further breeding. Once individual plants are selected for testing as potential introductions, then vegetative propagation protocols are applied to ensure the genetic integrity of the propagated plants. The remainder of this paper will present the seed and vegetative propagation protocols we have utilized for selected genera in the breeding program. The Chicago Botanic Garden is in northern Illinois in U.S.D.A. Zone 5a, for comparison of propagation dates in other regions.

SELECTED GENERA IN THE BREEDING PROGRAM

Asclepias (Milkweed) Propagation (*Asclepias tuberosa* and *A. purpurascens* Hybrids)

1. Seed. Ripe seed was collected from dehiscing fruit of various species and several hybrids in early autumn. The actual seeds are separated from the silky pappus (the parachutes). Seed viability is easy to discern in *Asclepias* as the linear embryos are readily visible by candling the seeds over a light source. We air-dried seeds for several days to several weeks, then cold stratified in damp sand at about 35-38°F for 12 weeks. Seed was sown in PT128 trays in March in a plug mix with a light covering of vermiculite and then placed under mist on 70°F bottom heat. Seedlings began to emerge within a few weeks. Move the seedlings along quickly and plant them out the same year as in-ground establishment is difficult if their taproots become at all potbound. Plants will bloom the following year, some in the first year.

2. Root Cuttings. A group of hybrid milkweed plants were grown for 3 years in a raised sand bed filled with a coarse builder's sand and leaf compost mix. This planting medium ensured ease of digging up roots. Dormant roots were then dug out of the raised bed in March. The roots used for propagation were cut into pieces approximately 1 to 3 in. in length and approximately ½ to 1 in. in diameter. Small, dormant, adventitious shoots were visible on some of the root pieces. The root cuttings were potted in 2½-in. pots in a well-draining bark mix and put into a greenhouse at a minimum of 65°F air temperature. Shoots emerged within 1 month-plus from 70% of the pots. As a side note, the roots remaining in the raised bed responded to their disturbance by sending up a proliferation of shoots that summer. This technique works well for the maintenance and propagation of selected milkweed clones. Plants will bloom the following year, some in the 1st year.

Symphyotrichum, Aster, Propagation (syn. *Aster ericoides*, *A. novae-angliae*, *A. oblongifolius*, and Hybrids)

1. Seed. Ripe seed is collected from mid-October to early November. Luckily our northern native asters are adapted to ripening their seed quickly after fall flowering, and seed maturation is not affected by intermittent light frosts or freezes. The seeds are air-

dried for upwards of several weeks, manually separated from their bristly pappus, then mixed with moist sand in small plastic envelopes, and stratified at 35-38°F for 12 weeks. The seeds are then sown in March in PT128 trays in a plug mix with a light covering of vermiculite and placed under mist on 70°F bottom heat. Seedling emergence occurs within a few weeks, and plants are large enough to plant out that summer. All the plants bloom the first year.

2. Cuttings. Cuttings are taken in late May into late June before flower buds are initiated. The basal portion of each cutting is dipped for 5 s in an aqueous solution of 1,250 ppm K-IBA and then stuck in a rooting medium of perlite and peat moss (2:1, v/v) in a CP72 plug tray. Cuttings are maintained in a fog house at 96% relative humidity and 68°F minimum air temperature with supplemental bottom heat at 75°F. Rooting is typically 100% within weeks. When cuttings are stuck in the rooting medium, it is imperative at least one or two vegetative nodes are stuck into the medium as it is from these buried nodes that an overwintering bud is formed. Otherwise, plants will have a much lower survival rate the following spring. Cuttings can also be stuck directly in larger SVD-2 and SVD-3 pots, which allow greater medium depth under the base of the deeply stuck cutting. Rooted cuttings will form good-sized flowering plants the same year.

***Baptisia*, False Lupins, Propagation (Multiple Species and Hybrids of the Prairieblues™ Series)**

1. Seed. Because we generally sow a limited amount of seed, we used sand paper to scarify each seed individually. *Baptisia* seed tend to have softer seed coats than other members of the pea family (Fabaceae), notably woody legumes such as *Cercis*, which can be scarified with concentrated sulfuric acid. Whatever method is used to scarify the seed, care must be taken not to cut all the way through the seed coat and into the underlying embryo and cotyledon tissue. After scarification, the seeds are soaked in water for 24 h then cold stratified in damp sand at about 35-38°F for 4 weeks. Seed is then sown in February in PT128 trays in a plug mix with a light covering of vermiculite, and then placed under mist on 70°F bottom heat. Seedling emergence is staggered over the next several months, likely due to varied seed coat thickness and possibly other dormancy factors. Plants are potted and planted out in spring of the following year and will be 2 to 3 additional years to blooming size.

2. Cuttings. Shoot-tip cuttings are taken as the vegetative growth is nearly finished and while the stems are still pliable, but the terminal is nearly finished elongating. In northern Illinois, this will typically be about mid-June. We take fairly long shoot tip cuttings, 3 in. in length, shorter for the more compact selections. The critical key to success is to stick the cuttings with at least one, and possibly two, unbranched nodes inserted into the rooting medium. One of these dormant axillary buds will become the overwintering bud in the pot the first winter; if there isn't an axillary bud in the rooting medium, the chances of plant emergence the following spring drops dramatically (Conner and Bir, 2001).

For rooting we use 1,250 K-IBA for a standard 5 s dip. A well-drained medium is optimal, such as a peat and perlite (1:1, v/v) mix (Note: this mix may dry out too readily after rooting, may want to modify slightly). Use a fairly deep pot for rooting since there is a good 1 to 1.5 inches of stem inserted into the medium. A plug tray won't work well as the basal end of the cuttings will be too deep in the plug, where it can be waterlogged. We have used an SVD-2 pot, 32 to a tray, for years quite successfully. Several of our licensed nurseries use SVD-3 or SVD-4 pots or their equivalent and stick two cuttings to a pot to ensure they have a fuller looking plant the following year. Treated cuttings are placed under mist. Most cuttings will root in 3 weeks though there will always be some laggards. The earlier you can root the cuttings, the higher the rooting percentages, and overwinter survival will be greater. Overwinter in a minimum heat polyhouse and keep them a bit on the dry side. We have rooted cuttings as late as August, but then had difficulty overwintering them. A small percentage of the plants will bloom the following year, and all of them should bloom the 2nd year.

***Echinacea,* Coneflower, Propagation (*E. purpurea*, *E. tennesseensis*, *E. paradoxa* and Hybrids of the Meadowbrite™ Series)**

1. Seed. Ripe seed is collected from late August to early October, species dependent. Bag the inflorescences during seed ripening to prevent goldfinches from stripping the seed. The seed is air-dried for upwards of a month, cleaned, then cold-stratified in damp sand at 35-38°F for 12 weeks. Seed is then sown in March in PT128 trays in a plug mix with a light covering of vermiculite. The trays are placed under mist on 70°F bottom heat. A quick flush of germination tends to occur within several weeks, then sporadic germination afterwards. The more complex interspecific hybrid seed no doubt has more complex dormancy mechanisms derived from their different parent species, leading to more sporadic and complicated germination. Seedlings can be planted out in the first year or grown on for planting the 2nd year. Some seedlings will bloom the 1st year, and all will do so the 2nd year.

2. Tissue Culture. Field-grown plants are dug up in late autumn, potted in a well-draining soilless medium, and then stored in a minimum heat polyhouse for several months. Plants are then greenhouse-forced into new growth starting in January-February of the following year. The most efficient explant is leaf petiole tissue from expanding basal leaves. The leaf petioles are surface disinfested for 12 min. in an aqueous 20% commercial bleach solution then rinsed for 10 min. in sterilized distilled water. The leaf petioles are then aseptically cut into 0.5-1.0-cm-long segments and placed on initiation medium abaxial (back) side down. The initiation medium consists of Murashige and Skoog (MS) basal medium and vitamins, 30 $g \cdot L^{-1}$ sucrose, 2.5 µM or 0.556 $mg \cdot L^{-1}$ of benzyladenine (BA), 0.5 µM or 0.113 $mg \cdot L^{-1}$ of potassium salt of NAA (K-NAA), adjusted to a pH = 5.7 to 6.3, and solidified with 7 to 9 $g \cdot L^{-1}$ agar. If the explants are sufficiently juvenile, adventitious shoots should appear on the cut ends and on the top of the explant in 3-4 weeks. If adventitious shoots are really small or there is no response after 4 weeks, subculture to fresh initiation medium for another 4-week period. Once shoots are initiated, they can be transferred to multiplication medium for continued shoot growth and axillary proliferation. The multiplication medium is the same as the initiation medium except the K-NAA is eliminated and the BA is decreased to 0.5 µM or 0.111 $mg \cdot L^{-1}$. Shoots can be divided and recultured on fresh multiplication medium every 3-4 weeks. Plant growth regulator-free medium works best for shoot rooting. Addition of IBA or NAA to either multiplication or rooting medium will stimulate callus growth, which should be avoided as it interferes with proliferation and rooting. All cultures were maintained in incubators with 12 to 14 h of light/day and 20-22°C. Rooted shoots are easy to acclimate in the greenhouse under fog or mist and on bottom heat.

***Phlox* Propagation, Moss Phlox, Summer Phlox (*P. subulata* Selections, *P. glaberrima*, *P. maculata*, *P. paniculata*, and Hybrids)**

1. Seed. Seed is collected any time from June into September, species or hybrid dependent. The spring-blooming moss phlox, such as *P. subulata* and *P. nivalis*, can ripen their seed in as little as 30 days; whereas the summer blooming *P. maculata* and *P. paniculata* tend to take 45 days or longer to mature their seed. Seed is cleaned, air-dried for at least several days, and then stored under refrigeration until sown. In late September, the seed is sown in PT128 trays in a plug mix with a light covering of vermiculite. These are placed under mist on 65°F bottom heat for a month. The trays are then moved to a minimum heat quonset that is kept above freezing, but experiences daily temperature fluctuations, especially on sunny days. Seeds from some of the spring blooming moss phlox adapted to very early growth or to high elevations will sporadically germinate while in the quonset. They are kept damp by hand-watering and the germinated seedlings removed to a greenhouse for growing on. The flats are then brought back into the mist house after 12 weeks. Usually there is a rapid flush of germination within a week when the flats are brought back to the mist house, then several more weeks of sporadic germination. The flats are given another cold treatment after 12 weeks in the mist house by putting them into a refrigerator at 35-38°F for 8 weeks; then they are moved back to

the mist house. The flats are also put back in the minimum heat quonset for a 2nd winter. Each time the flats are moved back to a warm environment there will often be another immediate small flush of germination followed by a few sporadic-emerging seedlings.
2. Cuttings. Shoot-tip cuttings are given a 5-s dip in 1,250 ppm K-IBA; the cuttings are stuck in perlite and peat moss (2:1, v/v) in CP72 cell trays and then placed under mist. The spring blooming moss phlox prefer cooler temperatures for rooting, and so bottom heat is not advised. These cuttings should be taken immediately after the plants finish blooming, typically in May for us. Cuttings from the summer-blooming taxa can be taken from June into October, as long as there is active shoot growth for shoot tip cuttings. These are treated as above and placed on 75°F bottom heat. Rooting for all types typically approaches 100% in about 4 weeks. The spring blooming types are typically grown on for planting out the following year, whereas the summer flowering taxa can be planted out the same year if rooted early enough or overwintered for planting the following year. All of these types will bloom the following year.

***Polygonatum* 'Prince Charming' (Solomon's Seal)**
1. Division. Brent Horvath from Intrinsic Perennial Garden in Hebron, Illinois, the originator of this selection, sells bare-root dormant divisions taken from stock plants in January to March before the plants emerge. A one or two-eye division with 1-2 in. of rhizome are recommended for a 4-in. pot. Active plants can also be divided over summer any time after they are done flowering. Plants will form nice small flowering clumps the following year.

***Tradescantia* 'Tough Love' (Spiderwort)**
1. Division. Field-grown plants dug in the fall and stored in a minimum heat quonset have been divided in March then potted into a well-draining bark mix in 3½-in. pots. These are kept in the greenhouse at 65°F and are all well rooted after about a month. Plants will bloom the same year and will be large enough to divide again by the following year. Stock plants can be kept in pots year-round for early spring division. Fall division of freshly dormant plants would likely work as well.

***Veronica*, Groundcover Speedwells ('Whitewater' and Hybrids)**
1. Seed. Seed is collected in July to August, taxon-depending. They are air-dried, then extracted from the small heart-shaped fruit, and stored under refrigeration until sown. Seed is sown March in PT128 trays in a plug mix with a light covering of vermiculite, then placed under mist on 70°F bottom heat. Germination takes place within a few weeks. Seedlings can be grown on for planting out the same year or the following year. The seedlings should all bloom the second year from seed. This protocol has worked for spike speedwells (*V. spicata*, *V. spicata* subsp. *incana*, etc.)
2. Cuttings. Shoot-tip cuttings can be taken from field grown or container grown stock plants as long as the shoots are in active growth. Cuttings have been rooted in March, May, June, and August with equal success. Other months would likely work. The basal end of shoot-tip cuttings approximately 2 in. in length are treated with a 5-s dip in an aqueous solution of 1,250 ppm K-IBA. Cuttings are then stuck in CP72 pots (cell packs 72 per flat) containing a medium of perlite, peat, and sand (2:1:1, by vol.). A rooting medium perlite and peat (2:1, v/v) is also as effective. Pots are placed in a fog greenhouse with bottom heat of 75°F and air temperature of 70 to 80°F. All the cuttings root within weeks. Plants are container grown until the following year, when they should all bloom.
3. Division. While we have not attempted division with these plants, the fact that their shoots readily layer into the ground suggests these can be lifted and divided, probably best immediately after blooming in late spring.

Literature Cited
Conner, J.L. and Bir, R.E. 2001. Propagating *Baptisia* 'Purple Smoke' and keeping it alive. Proc. SNA Res. Conf. 46:400-401.