Effects of Self-pollination and Cross-pollination of *Vaccinium darrowii* (Ericaceae) and Other Low-chill Blueberries

Dario J. Chavez and Paul M. Lyrene

Horticultural Sciences Department, University of Florida, P.O. Box 110690, 2135 Fifield Hall, Gainesville, FL 32611

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Abstract. Partial to complete self-incompatibility is normal in most *Vaccinium* L. (Ericaceae) species. Wild blueberry plants of several Florida provenances and species were self- and cross-pollinated in a greenhouse free of pollinators. Fruit set of *V. darrowii* Camp (2x), *V. corymbosum* L. (4x), *V. arboreum* Marsh (2x), and F₁ (*V. darrowii × V. corymbosum*) hybrids was higher after cross-pollination than after self-pollination. Partial to complete self-incompatibility was present in *V. darrowii*, *V. corymbosum*, and their tetraploid F₁ hybrids. The three *V. arboreum* clones tested were fully self-incompatible. Intra- and interpopulation crosses in *V. corymbosum*, *V. darrowii*, and *V. darrowii × V. corymbosum* hybrids were highly successful, and self-pollination reduced all fertility parameters. Advanced selections of *V. corymbosum* were the most self-compatible clones tested, possibly because self-compatibility has been increased by breeders selecting for reliable fruit set in large fields planted with one or a few clones. One southern highbush selection and two F₁ hybrids had fruit set of more than 70% after self-pollination. These plants could be potentially used to breed plants that could be planted in single blocks providing reliable yield.

In the genus *Vaccinium* L., there are no fundamental sterility barriers between homoploid members of the same phylectic section (subgenus) (Darrow et al., 1949; Meader and Darrow, 1944; Sharpe, 1953; Sharpe and Darrow, 1959). Diploid, tetraploid, and hexaploid species exist within several sections of the genus. In section *Cyanococcus* L., which includes the cultivated blueberries, partial to complete self-incompatibility and interfertility between homoploid species has allowed formation of interspecific hybrid swarms (Camp, 1942; Vander Kloet 1983, 1988). Studies of self-pollination and cross-pollination in several *Vaccinium* species have provided varying results. In most cases, partial to complete self-incompatibility was present, particularly in wild or “unimproved” plants (Bailey, 1938; Coville, 1921; Meader and Darrow, 1944; Merrill, 1936; Merrill and Johnston, 1940; Morrow, 1943; White and Clark, 1939). In cultivated highbush blueberry (tetraploid *V. corymbosum* L. and its hybrids), cross-pollination usually produces earlier ripening berries, increased berry size, and higher fruit set. Fewer fully developed seeds were found in self-pollinated berries. Partial to complete self-incompatibility, slower pollen tube growth, or collapse of ovules after fertilization could be factors that produced these results (Morrow, 1943).

Meader and Darrow (1944) studied the crossing behavior of several hexaploid rabbiteye (*V. virgatum* Aiton) varieties. Fruit set was higher after cross-pollination. Berry weight and seed weight per berry were always higher after cross-pollination than after self-pollination. In the same study, several section *Cyanococcus* species from different ploidy levels were self- and cross-pollinated. Tetraploid forms of *V. virgatum* were found to be self-sterile but gave a fruit set of 1% to 27% when cross-pollinated with tetraploid *V. corymbosum*. *Vaccinium tenellum* Aiton (2x) gave a fruit set below 29% when selfed, but when crossed with a different clone of the same species, had over 80% fruit set. *Vaccinium darrowii* Camp (2x) set 15% when selfed. *Vaccinium darrowii*, crossed with other diploid species (*V. pallicium* Aiton and *V. elliottii* Chapman), gave a fruit set of 71.5% and 62.2%, respectively. Berries formed after pollinating *V. darrowii* flowers with *V. elliottii* pollen ripened earlier than those produced by self-pollination of *V. darrowii*. However, *V. myrsinites* (4x) produced two-thirds less fruit set when crossed with other tetraploid species than it did when self-pollinated. Fruit set of self-pollinated *V. myrsinites* Lam. averaged 79%. Krebs and Hancock (1990) reported that fruit set was significantly greater after cross-pollination of *V. corymbosum* cultivars. They proposed that variable fertility after self- and cross-pollination in cultivated highbush was produced by early-acting inbreeding depression (seed abortion). They proposed that the degree of self-incompatibility depended on the level of zygotic inbreeding, which depended on the parents that were mated. Reduced self-fertility was attributed to homozygosity for sublethal mutations at loci controlling embryo development or to loss of heterotic interactions at these loci (Hokanson and Hancock, 2000; Krebs and Hancock, 1991). Diploid *V. darrowii* has been extensively used in breeding blueberries, but most crosses have involved only one *V. darrowii* clone, Fla. 4B. *Vaccinium darrowii* in Florida is quite variable. Lyrene (1986) described three types of *V. darrowii* in Florida: 1) the Florida panhandle race, a petit form with highly glaucous leaves, which matches Camp’s original description of the species (see Camp, 1945); 2) the Ocala Forest race, a tall form with shiny green leaves (glaucous on new growth flushes); and 3) the Istokpoga race, found at the southern end of the species range in the Florida peninsula, a highly variable population with short and tall plants. Introggression from a diploid highbush species (*V. fuscatum*) was clearly occurring in the Florida peninsula. The use of a wider range of *V. darrowii* accessions in breeding would provide beneficial diversity in the cultivated gene pool.

*Vaccinium arboreum* (sparkleberry) is a diploid species that is abundant and widespread in the southeastern United States. Because it is in section *Batodendron* Marsh. and does not readily make fertile hybrids with cultivated blueberries, which are in section *Cyanococcus*, it has not been much used in breeding. Recently, genes from *V. arboreum* have been shown to be useful in breeding (Brooks and Lyrene, 1995; Lyrene, unpublished data), and more information on the pollination biology of this species is needed. The purpose of this research was to determine levels of self-compatibility within the populations of several *Vaccinium* species and to search for plants with high self-compatibility that could be used in breeding self-compatible southern highbush cultivars.

Materials and Methods

Self- and cross-pollination experiments. Self- and cross-pollination were conducted using plants of *V. darrowii* (2x), *V. arboreum* (2x), *V. fuscatum* (2x), *V. corymbosum* (4x), and F₁ (*V. darrowii × V. corymbosum*) hybrids. The diploids, *V. darrowii*, *V. fuscatum*, and *V. arboreum* clones, were propagated from wild. *Vaccinium darrowii* clones came from the vicinity of Sumatra and Wilma in the Florida panhandle and from the Florida peninsula in the vicinity of Lake Istokpoga. *Vaccinium fuscatum* clones (a Florida diploid highbush taxon resembling *V. corymbosum*) were propagated from plants growing in a disturbed wetland near Davenport
in the central Florida peninsula. Softwood cuttings from the selected wild plants were rooted in a 1:1 mixture of sphagnum peat and perlite. Rooted cuttings were transplanted to 4-L nursery containers filled with sphagnum peat and perlite (1:1). The plants were maintained in containers in a greenhouse at the University of Florida, Gainesville, FL. *Vaccinium arboreum* plants were grown from open-pollinated seeds collected in Clay County in northeastern Florida. The seedlings were transplanted to a high-density nursery at the University of Florida Plant Science Unit in Citra, FL, in May. In November, 15 plants were dug with a root ball of soil and transplanted to 8-L nursery containers using sphagnum peat. The containers were placed in a greenhouse in Gainesville, FL.

Tetraploid southern highbush selections from the Florida breeding program were selected from a field nursery, potted in 8-L nursery containers of sphagnum peat, and placed in a greenhouse in Gainesville, FL. For simplicity, these tetraploid products of the breeding program will be designated *V. corymbosum*, although other species are included in their genetic background. Tetraploid *Vaccinium darrowii × V. corymbosum* hybrids were selected from crosses made at the University of Florida. The F₁ hybrids were grown in a field nursery for 8 months and then selected and transplanted to 8-L nursery containers of sphagnum peat and maintained in a greenhouse in Gainesville, FL. Self- and cross-pollination experiments for southern highbush selections, F₁ hybrids (*V. darrowii × V. corymbosum*), *V. darrowii*, *V. fuscatum*, and *V. arboreum* clones were made. The results from the individual crosses of each type were averaged and compared.

In February of each year, the highbush plants and F₁ hybrids were chilled for more than 1000 h in a walk-in cooler at 4 °C with no light, after which they were placed in a greenhouse. The *V. arboreum*, *V. darrowii*, and *V. fuscatum* clones, which are evergreen and have no chilling requirement, were not chilled. Each *V. darrowii* plant was divided into two sections to compare self- and cross-pollination as described by Morrow (1943). Flowers that had opened previously were removed. Flowers were emasculated before pollination. Pollinations were made in a greenhouse free of pollinators. For cross-pollination, new flowers were pollinated with pollen from a different clone of the same species with the exception of *V. fuscatum*, which was cross-pollinated with *V. arboreum* pollen. For self-pollination, new flowers were pollinated with pollen from the same plant. Pollen was collected by tapping the male flower on thethumbnail. The stigma of emasculated flowers were touched by the thumbnail. For each test, 100 to 250 flowers were self-pollinated and 100 to 250 flowers were cross-pollinated.

Berries were harvested when fully ripe. Berry weight and number of seeds per berry were determined for the first 20 berries ripe from each cross. Seeds were classified as plump and round (large seeds) and shriveled and shrunken (small seeds). Seeds from additional berries were removed using a food blender. Seeds were washed free of pulp and skins with water, dried on a benchtop at room temperature, and stored in coin envelopes at 5 °C. Seeds were planted in Nov. 2007 on the top layer of 4-L nursery containers of sphagnum peat. The containers were kept in a greenhouse with intermittent mist for 2 to 3 months until germination was complete. Seedlings from each cross were counted.

First-pollination to first-ripening interval was calculated from the first day of pollination to the first day of berry ripening. Average pollination-to-ripening interval was calculated from the mean day of pollination to the mean day of harvesting. Fruit set percentage, berry weight, number of seeds per berry, number of plump seeds per pollinated flower, pollination-to-ripening interval, average pollination-to-ripening interval, and number of seedlings per pollinated flower were determined for self- and cross-pollinated flowers.

An index of self-incompatibility (ISI) was calculated by dividing the average number of plump seeds per pollinated flower obtained after self-pollination by the same following cross-pollination. A score of 0 to 0.2 was classified as self-incompatible, a score of 0.2 to 1 as incompletely compatible, and a score of 1 as self-compatible (Zapaya and Arroyo, 1978). Means for fruit set percentage were separated using the χ² “test of independence” with a significance level of 5%. Means for berry weight, number of seeds per berry, number of plump seeds per pollinated flower (PPF), number of seedlings per pollinated flower (SPF), pollination-to-ripening interval, and average pollination-to-ripening interval for the different treatments were separated using least square means by Tukey’s test with a significance level of 5%. Data analysis was performed using the GLM and FREQ procedures of SAS (Statistical Analysis System Version 9.1; SAS Institute, Cary, NC). In the χ² tests for fruit set, each pollinated flower was considered an observation with two possible responses: set or nonset. The null hypothesis of the χ² “test of independence” was that the ratio of set to nonset was the same for the two crosses being compared.

### Results

**Fruit set (percentage).** Fruit set for *V. darrowii* was less than 30% after self-pollination and more than 70% after cross-pollination (Table 1). For *V. arboreum*, fruit set was 0% after self-pollination and ranged from 3.3% to 54.2% after cross-pollination (*V. arboreum × V. arboreum*) (data not shown). For *V. corymbosum* selections, fruit set after self-pollination averaged 39.4% (range for five crosses, 9.8% to 74.1%). Fruit set of *V. corymbosum* after cross-pollination averaged 82.8% (range for 74 crosses, 37.1% to 94.3%, data not shown).

Fruit set for self-pollination of 17 F₁ (*V. darrowii × V. corymbosum*) hybrids ranged from 0% to 90.3%. Two hybrid plants had a selfed fruit set over 80% (data not shown).

Fruit set after cross-pollination of F₁ (*V. darrowii × V. corymbosum*) hybrids was 59.8% for one cross and 98.2% for a second. Two of the three *V. fuscatum* clones tested had fruit set over 50% after self-pollination. Fruit set after self-pollination did not differ significantly between *V. fuscatum* and southern highbush selections (Table 1). Mean fruit set of *V. darrowii*, *V. corymbosum*, and their F₁ hybrids after self-pollination was lower than after cross-pollination (*P < 0.05*) (Table 1).

**Pollination-to-ripening interval.** First-pollination to first-ripening intervals and average pollination-to-ripening intervals did not differ significantly for self- and cross-pollination in either *V. darrowii* or *V. fuscatum* (Table 1). In southern highbush selections and in F₁ (*V. darrowii × V. corymbosum*) hybrids, first-pollination to first-ripening interval was longer after self-pollination than after cross-pollination (*P < 0.05*). For southern highbush selections, average pollination-to-ripening was also significantly longer after self-pollination (Table 1).

**Berry weight.** Mean berry weight of *V. darrowii* and F₁ (*V. darrowii × V. corymbosum*) hybrids was lower after self-pollination than after cross-pollination (*P < 0.05*). Mean berry weight of southern highbush selections and *F. fuscatum* clones was higher after self-pollination than after cross-pollination (*P < 0.05*) (Table 2).

**Seeds and seedlings per flower.** Number of PPF after self-pollination of *V. darrowii* ranged from 0.06 to 3.32, depending on the clone that was selfed. SPF ranged from 0.02 to 0.32. Cross-pollinated *V. darrowii* gave PPF ranging from 3.39 to 37.96 and SPF ranging from 1.23 to 9.83 (data from individual crosses not shown). For PPF and SPF, self-pollination of *V. darrowii* gave lower values than cross-pollination (*P < 0.05*; Table 2). SPF of *V. darrowii* after cross-pollination was ≥20 times higher than after self-pollination (*P < 0.05*). The average number of seeds per berry was lower when *V. darrowii* clones were self-pollinated compared with cross-pollination (*P < 0.05*). Few plants were produced from self-pollinated *V. darrowii* seeds. The ISI was 0.08 for *V. darrowii*, classified as self-incompatible.

After cross-pollination of *V. arboreum*, PPF ranged from 0.46 to 6.78 and SPF ranged from 0.10 to 1.80 compared with zero values for self-pollinated *V. arboreum* (data from individual crosses not shown). Self-pollinated *V. arboreum* flowers abscised a few weeks after pollination. The three *V. arboreum* clones tested appeared to be completely self-incompatible with an ISI score of 0.

In southern highbush selections, PPF for self-pollination ranged from 0.52 to 8.13 compared with 3.08 to 11.50 for cross-pollination (data from individual crosses not shown). Number of large seeds per berry and total number of seeds per berry were lower for self-pollination than for cross-pollination (*P < 0.05*). Cross-pollinated berries had fewer small seeds compared with self-pollinated berries (*P < 0.05*). The ISI score
for the southern highbush selections was 0.38, classified as incompletely compatible. In *V. darrowii × V. corymbosum* hybrids, PPF ranged from 0.00 to 12.22 for self-pollination and from 8.69 to 21.90 for cross-pollination (data from individual crosses not shown). Two F₁ hybrid plants gave a PPF higher than 7.00 when self-pollinated. PPF and seeds per berry after self-pollination were lower than after cross-pollination ($P < 0.05$). The ISI score for the F₁ hybrids was 0.22, classified as incompletely compatible (Table 2). SPF for the *V. darrowii × V. corymbosum* hybrids and for *V. fuscatum* were not significantly different and for *V. fuscatum* were not significantly different between cross- and self-pollination experiments ($P < 0.05$) (Table 2). The ISI score for *V. fuscatum* was 0.46, classified as incompletely compatible.

### Discussion

Coville’s (1921, 1937) first attempts to self-pollinate highbush blueberry cultivars failed. Self-pollinated plants ripened few berries, and those that ripened contained few seed. In cross-pollinations among different plants of the same species, he obtained many berries with numerous seeds. Krebs and Hancock (1991) attributed the reduced fertility of highbush blueberry after self-pollination to postzygotic events that aborted most selfed seeds. In our study, fruit set of *V. darrowii, V. corymbosum, V. arboreum*, and F₁ (*V. darrowii × V. arboreum*) hybrids after cross-pollination was higher than after self-pollination. Differences in fruit set between self- and cross-pollination were similar to those reported by Coville (1921), but the degree of self-incompatibility varied among the taxa.

Although the experiments were not specifically designed to compare the length of the fruit-development period among taxa, it is clear that *V. arboreum*, a fall-ripening species, has a very long fruit development period; that southern highbush selections, which have been selected for early ripening, have a short fruit-development period; and *V. darrowii*, the Florida native species used in breeding to impart southern adaptation to highbush cultivars, is intermediate.

No previous self-pollination studies have been reported in *V. arboreum*. In our study, complete self-incompatibility was observed in the three clones of *V. arboreum* that were tested. Self-pollinated *V. arboreum* flowers abscised a few weeks after pollination. In addition, several *V. arboreum* clones that flowered in a greenhouse in Gainesville, FL, without access to pollination set no berries. Under similar conditions, some clones of *V. corymbosum, V. darrowii, V. fuscatum*, and F₁ (*V. darrowii × V. fuscatum*) natural hybrids set a few berries (unpublished data).

The 17 F₁ (*V. darrowii × V. corymbosum*) hybrids that were self-pollinated in these experiments were produced from *V. darrowii (2x) × V. corymbosum (4x)* crosses. Approximately 95% of these hybrids were believed to be tetraploids and had high pollen viability determined by staining the pollen with aceto-carmine (Chavez and Lyrene, 2009). The results of cross- and self-pollination of the F₁ hybrids were similar to those for the southern highbush selections and *V. darrowii* clones. One southern highbush clone and two F₁ hybrid plants had fruit set over 70% after self-pollination. The higher self-compatibility of highbush selections is attributed to breeding and selection for plants that set fruit reliably in large clonal plots. High-chill northern highbush blueberry cultivars, which are grown from North Carolina to Michigan, and in the Pacific Northwest, vary in self-compatibility. Those that have become most popular with growers such as ‘Duke’ and ‘Bluecrop’ are highly productive in large fields planted to single clones. Cultivars with low self-compatibility tend to give inconsistent yields from year to year (Lyrene, personal communication).

### Table 1. Results of self- and cross-pollination in Florida *Vaccinium* species measured by fruit set (%), first-pollination to first-ripening interval (pol-ripe interval), and average pol-ripe interval (pol-ripe interval average)

| Cross type | Crosses (no.) | Flowers (no.) | Berries (no.) | Fruit set (%) | Pol-ripe interval (first) | Pol-ripe interval (avg) |
|------------|---------------|---------------|---------------|---------------|--------------------------|-------------------------|
| Cross      | 6             | 1,496         | 1,118         | 74.2 a        | 83.8 a                   | 96.7 a                  |
| Self       | 6             | 1,576         | 331           | 21.3 b        | 95.8 a                   | 114.8 a                 |
| *Vaccinium arboresum* | Cross | 6 | 1,061 | 366 | 151.7 | 158.8 |
| Self | 3 | 547 | 0 | 0.0 |
| *Vaccinium corymbosum* (southern highbush) | Cross | 74 | 10,168 | 8,741 | 82.8 a | 53.7 b |
| Self | 5 | 500 | 188 | 39.4 b |
| *V. fuscatum* hybrids (*V. darrowii × V. corymbosum*) | Cross | 2 | 538 | 426 | 78.9 a |
| Self | 17 | 1,794 | 1,325 | 45.5 b |

| *Vaccinium fuscatum* | VF × VA | 3 | 723 | 279 | 40.2 a |
| Self | 3 | 912 | 402 | 40.8 a |

In *V. fuscatum* crosses, cross-pollinations within the species were not made. Results of crosses between diploid *V. fuscatum* and diploid *V. arboresum* (VA) are presented for comparison.

Pol-ripe interval (first) = number of days from first pollinated flower to first ripe berry.

Pol-ripe interval (average) = number of days from median pollination date to median ripening date.

No data.

Similar letters within a column indicate means not significantly different. Comparisons were made within species. Tukey’s test for pol-ripe interval (first) and pol-ripe interval (average), $α = 0.05$. $χ^2$ “test of independence” for fruit set (%), $α = 0.05$.

### Table 2. Results of self- and cross-pollination in Florida *Vaccinium* species measured by mean berry weight (g), number of large seeds per berry, number of small seeds per berry, total number of seeds per berry, number of plump seeds per pollinated flower (PPF), number of seedlings per pollinated flower (SPF), and index of self-incompatibility (ISI)

| Cross type | Number of clones tested | Berry wt (g) | Large seeds | Small seeds | Total seeds/berry | SPF * | SPF ± | ISI * |
|------------|-------------------------|--------------|-------------|-------------|-------------------|-------|-------|-------|
| Cross      | 6                       | 0.43 a       | 21.2 a      | 11.8 b      | 33.0 a            | 15.28 | 4.44 a | 0.08  |
| Self       | 6                       | 0.27 b       | 4.7 b       | 16.3 a      | 21.1 b            | 1.26  | 0.15  |       |
| *Vaccinium arboresum* | Cross | 6 | 0.31 | 11.4 | 7.1 |
| Self | 3 | — | — | — |
| *Vaccinium corymbosum* (southern highbush) | Cross | 5 | 1.53 b | 31.7 a | 19.9 b |
| Self | 3 | 1.69 a | 7.7 a | 24.1 a |
| *F₁* hybrids (*V. darrowii × V. corymbosum*) | Cross | 2 | 1.24 a | 18.2 a | 30.2 a |
| Self | 17 | 0.94 b | 8.2 b | 14.7 b |
| *Vaccinium fuscatum* | VF × VA | 3 | 0.12 b | 17.3 a | 13.9 a |
| Self | 3 | 0.22 a | 3.7 a | 9.0 b |

*PPF = Number of plump seeds per pollinated flower. SPF = Number of seedlings per pollinated flower. ISI = Index of self-incompatibility. Self-compatible species score = 1, incompletely compatible species score = 0.2–1, and self-incompatible species score = 0–0.2. *Similar letters within a column indicate means not significantly different. Comparisons were made within species. Tukey’s test, $α = 0.05$. *In *V. fuscatum* crosses, cross-pollinations within the species were not made. Crosses between diploid *V. fuscatum* (VF) with diploid *V. arboresum* (VA) were used for comparisons as cross-pollination experiments. No berries were produced, no data. *Missing data.

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Fruit set, first-pollination to first-ripening interval, average pollination-to-ripening interval, berry weight, number of large seeds per berry, number of small seeds per berry, total number of seeds per berry, and PPF were different for cross- and self-pollination of *V. corymbosum* selections (Table 2) and indicated varying levels of self-incompatibility. Similar results were described by Vander Kloet and Lyrene (1987), who found strong self-incompatibility in wild *V. corymbosum* plants. They found that intra- and inter-population crosses in *V. corymbosum* were equally fertile, but self-pollination resulted in reductions in all fertility parameters. Morrow (1943) observed that after self-pollination in *V. darrowii*, pollen tube growth is slower, ovules may collapse after fertilization, and the number of fully developed seeds is reduced. In another study, *V. darrowii* gave low fruit set when selfed (15%) but high fruit set when cross-pollinated with other diploid species, *V. elliottii* and *V. pallidum* (62.2% and 71.5%, respectively) (Meader and Darrow, 1944).

In our study, cross-pollination increased berry size for *V. darrowii* and F1 hybrids but not for southern highbush selections or *V. fuscatum* clones. This result is surprising because most previous studies in blueberry have shown a positive correlation between berry size and berry seed content. Phenotypic variation between the plants used as female parents may be the reason for this difference. Morrow (1943) reported that cross-pollination increased berry size of three northern highbush cultivars: Scammen, Weymouth, and Dixi. Meader and Darrow (1944) studied 10 rabbiteye (*V. virgatum*) varieties. Without exception, berries were larger after cross-pollination than after self-pollination. In the same study, *V. tenellum* and *V. darrowii* cross-pollinated flowers resulted in larger berries than self-pollinated flowers. All fertility parameters were reduced for self-pollination, including a lower number of seeds per berry that contributed to a smaller berry size.

Average berry weight, number of seeds per berry, and PPF of *V. darrowii* and F1 (*V. darrowii × V. corymbosum*) hybrids were higher when these species were cross-pollinated rather than self-pollinated. Reduction in total number of seeds per berry may have been the result of reduced pollen germination or pollen tube growth, both prezygotic events. Reduction in PPF and SPF may have been the result of embryo abortion after self-pollination, which is a postzygotic event. Both pre- and postzygotic events were believed to be involved with the self-incompatibility reaction. Few plants, with abnormal morphology, were produced from seeds of *V. darrowii*, *V. fuscatum*, and *V. darrowii × V. corymbosum* hybrids after self-pollination (Table 2). Similar results were obtained by Coville (1937). He found that self-pollination of highbush cultivars produced few berries. Most seeds were abnormal and lacked embryos, and few useful plants were obtained from self-pollination. Krebs and Hancock (1990) obtained few plants after self-pollination of *V. corymbosum*. They attributed the low seedling numbers after self-pollination to increased homozygosity of deleterious alleles at loci that are critical to embryo vigor and survival.

In conclusion, some level of self-fertility was seen in some plants of every taxon we tested except *V. arboenum*. One southern highbush selection from the breeding program and two F1 *V. corymbosum × V. darrowii* tetraploid hybrids had fruit set of more than 70% after self-pollination. This indicated the potential for developing low-chill highbush cultivars that would give reliable fruit set when planted in solid blocks containing a single clone.

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