Flower Morphology in Blueberry Species and Hybrids

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Additional index words. *Vaccinium ashei*, *V. corymbosum*, *V. constablaei*, *V. simulatum*, corolla length, corolla width, corolla aperture, style length, anther-to-stigma distance, insect pollination

Abstract. Several morphological features of *Vaccinium ashei* Reade, *V. constablaei* A. Gray, their F_1 hybrids, *V. simulatum* Small, and southern highbush blueberry (*V. corymbosum* L. hybrids) flowers were compared in Gainesville, Fla. Desirable characteristics that could increase the extent of honeybee pollination, such as a large corolla aperture and a short anther-to-stigma distance, were common in *V. constablaei* but not in *V. ashei*. F_1 (*V. ashei* × *V. constablaei*) hybrids were generally intermediate between the two parents. Thus, it appears that *V. constablaei* could be used to breed *V. ashei* cultivars with improved flower morphology. *Vaccinium simulatum* and *V. constablaei* flowers were similar in all features. The corollas of southern highbush blueberry flowers were wide and had wide apertures, but the distance between stigma and anther pore was also large.

Fruit set in blueberries may be related to the flower structure and attractiveness to pollinating insects (Shutak and Marucci, 1966). The blueberry flower is morphologically adapted to facilitate cross-pollination by bees. The distance between the stigma and the anther pores from which the pollen is discharged, together with the stigma shape, with angled sides like an inverted funnel, reduce within-flower self-pollination. The anthers closely surround the pistil, but in the absence of insect visitation, few pollen grains fall on the stigma of the same flower (Shutak and Marucci, 1966).

Few studies have attempted to relate morphological features of the blueberry flower to fruit set. Eck and Mainland (1971) measured various flower parts from 35 *V. corymbosum* (highbush) cultivars and observed that fruit set was negatively correlated with distance between stigma and anther pore. High fruit set was also associated with short corollas that were wide at the middle and narrow at the base. Flowers with these characteristics possibly were more attractive to pollinating insects and easier to pollinate. Long corolla tubes with narrow apertures are typical of rabbiteye blueberries and may reduce pollination by hindering honeybee entry. Improvement in the shape of rabbiteye flowers should be possible, given the large variation among rabbiteye clones for most flower structural features (Lyrene, 1994a). In this study, we compared several flower morphological features of *V. ashei*, *V. constablaei*, their F_1 hybrids, southern highbush blueberry (*V. corymbosum* hybrids), and *V. simulatum* to obtain information that might be useful in breeding blueberry cultivars with

flower shapes more favorable for pollination by honeybees.

Materials and Methods

Flower features were determined for 23 *V. ashei*, 12 *V. constablaei*, 13 *V. ashei* × *V. constablaei* F_1 hybrids, four southern highbush blueberry (*V. corymbosum* hybrids), and one *V. simulatum* clones in Mar. 1993. The *V. constablaei* and *V. simulatum* flowers originated from dormant branches cut on 23 Dec. 1992 from plants growing wild at altitudes above 1600 m in the mountains of western North Carolina near the Shining Rock Wilderness Area south of Asheville. *Vaccinium constablaei*, which was growing on an open meadow with few or no trees, was extremely abundant, with hundreds of clones 1.1 to 1.7 m tall, each of which had hundreds of densely packed canes that formed colonies 4 to 10 m in diameter. The *V. simulatum* plants, which were growing within 2 km of the *V. constablaei*, were 2 to 3 m tall and produced only a few major canes. Enough dormant branches were taken from each clone to provide 50 to 100 inflorescence buds containing several hundred flowers. The cut branches were enclosed in plastic bags with wet paper towels to maintain moisture, and were immediately placed on ice in a chest (0 °C). On 4 Jan. 1993 they were transferred to a cold room (7 °C). At the time of collection, the branches had already received many hours of chilling.

The bags were opened and checked every 2 weeks, and, if necessary, the wet paper towels were replaced. On 4 Feb., the bases were recut and the branches were placed in a greenhouse in 4 L plastic buckets, each containing 1.5 L of a preservative solution [sucrose (5%) + citric acid (5.93×10^{-4} M) + 8-hydroxy quinoline hemisulfate salt (5.93×10^{-4} M)]. Once a week, the bases of the branches were recut and the preservative solution was renewed. The greenhouse was unheated except by the sun during

the day and by heaters that kept the temperature above 5 °C at night.

The *V. ashei* and *V. corymbosum* clones used in the experiment were cultivars or advanced selections from the Univ. of Florida breeding program. The F_1 *V. ashei* × *V. constablaei* hybrids had been produced several years earlier by hand crosses, and had been grown for 2 years in a high-density field nursery in Gainesville, Fla. The *V. constablaei* parents of these F_1 plants were from the U.S. Department of Agriculture, National Clonal Germplasm Repository in Corvallis, Ore., and the *V. ashei* parents were advanced selections from the Univ. of Florida blueberry breeding program. In Dec. 1992, the *V. ashei*, *V. corymbosum*, and F_1 hybrid plants were dug from field nurseries, potted, and placed in a dark chamber at 7 °C for 6 weeks. In February, they were removed from the cooler and placed in the same greenhouse in which the *V. constablaei* and *V. simulatum* branches were being forced. Ten fully opened flowers were assessed per clone. The following features were evaluated: corolla length, measured from the deepest indentation of the calyx to the apex of the corolla; corolla width, representing the widest diameter at the middle of the corolla tube; corolla aperture, which was the mean diameter of the opening at the apex of the corolla tube; style length from the top of the ovary to the distal end of the stigma; anther-to-stigma distance, measured from the pore of the anthers up to the stigma; overall stigma position in relation to the corolla apex; and color of the petals.

A possible source of error in this experiment was the comparison of flowers forced on cut branches (*V. constablaei* and *V. simulatum*) with flowers opening on potted plants for the other taxa. This procedure was necessary because the mountain species cannot be grown to maturity in the field in Florida. A few fall flowers that have been observed on *V. constablaei* and *V. simulatum* seedlings grown in the field in Florida were morphologically similar to flowers on the cut branches forced in the greenhouse. Lyrene (1994b) found that flower morphological parameters were little affected by the amount of chilling the dormant plants received before flowering or by the temperature in the forcing environment.

Statistical analyses were performed for all characteristics, except stigma position and color of the petals, using a completely randomized design with varying numbers of replicates. For each parameter, the clone mean was obtained by averaging the measurements from the 10 flowers per clone. The taxa were then compared using the variance among clones within taxa as the error variance. When analysis of variance indicated significant differences among taxa for a parameter, taxa means were compared using Tukey's mean separation procedure.

Results and Discussion

Flowers of *Vaccinium ashei*, *V. constablaei*, their F_1 hybrids, *V. corymbosum* (southern highbush), and *V. simulatum* differed in mor-

Received for publication 26 Sept. 1997. Accepted for publication 10 Feb. 1998. Florida Agricultural Experiment Station Journal Series Number R-05990. The cost of publishing this paper was defrayed in part by the payment of page charges. Under postal regulations, this paper therefore must be hereby marked advertisement solely to indicate this fact.

Table 1. Flower morphological features of *Vaccinium ashei*, *V. constablaei*, their F₁ hybrids, *V. corymbosum* (southern highbush), and *V. simulatum*. All measurements in mm.

Vaccinium taxon	No. clones examined	Corolla			Style length	Anther-to-stigma distance
		Length	Width	Aperture		
<i>V. ashei</i>	23	9.1 a ²	6.6 b	2.7 b	10.0 a	2.8 a
F ₁ ³	13	7.2 b	6.2 bc	3.3 ab	7.9 b	1.4 b
<i>V. constablaei</i>	12	5.3 c	5.2 c	3.8 a	5.2 c	0.7 b
<i>V. corymbosum</i>	4	8.9 a	7.9 a	3.6 a	9.1 a	2.9 a
<i>V. simulatum</i> ⁴	1	4.8	6.1	4.1	5.1	0.4

²Mean separation within columns by Tukey's test $P \leq 0.05$.

³F₁ hybrid of *V. ashei* x *V. constablaei*.

⁴Data from *V. simulatum* are not included in the statistical analyses because of insufficient replication.

Table 2. Number of clones having various corolla colors for *Vaccinium ashei*, *V. constablaei*, their F₁ hybrids, *V. corymbosum* (southern highbush), and *V. simulatum*.

Vaccinium taxon	No. clones examined	Corolla color			
		White	Light-yellow	White w/ yellow veins	Red-yellow
<i>V. ashei</i>	23	22	1	0	0
F ₁ ²	13	4	7	2	0
<i>V. constablaei</i>	12	1	11	0	0
<i>V. corymbosum</i>	4	2	1	0	1
<i>V. simulatum</i>	1	0	1	0	0

²F₁ hybrid of *V. ashei* x *V. constablaei*.

Table 3. Stigma position in relation to the corolla apex of flowers of clones of *Vaccinium ashei*, *V. constablaei*, their F₁ hybrids, *V. corymbosum* (southern highbush), and *V. simulatum*.

Vaccinium taxon	No. clones with stigma located:				
	Inside or at the corolla apex	At the corolla apex	Exserted beyond or at the corolla apex	Exserted beyond corolla	Total
<i>V. ashei</i>	0	1	10	12	23
F ₁ ²	1	4	3	5	13
<i>V. constablaei</i>	0	12	0	0	12
<i>V. corymbosum</i>	1	1	2	0	4
<i>V. simulatum</i>	0	1	0	0	1

²F₁ hybrid of *V. ashei* x *V. constablaei*.

phology. *Vaccinium ashei* and *V. corymbosum* had larger flowers, as measured by the length of the corolla and style (Table 1). Their large size may have been due, in part, to selection for cultivars with large fruit. *V. ashei* and *V. corymbosum* (highbush) differed significantly from *V. constablaei* and *V. ashei* x *V. constablaei* F₁ hybrids in corolla length and width. Highbush corollas were significantly wider than those of the other taxa. Despite their small size, *V. constablaei* flowers had large corolla apertures, similar to those ofighbush flowers and significantly larger than the apertures of *V. ashei* flowers. As measured by the anther-to-stigma distance (Table 1), the pore at the end of the anther, where the pollen is released, was much closer to the stigma in *V. constablaei* than in the cultivated blueberries *V. ashei* and *V. corymbosum* (highbush). The single *V. simulatum* clone that was sampled had flowers that were short and had a wide corolla aperture and a short anther-to-stigma distance.

The data show that *V. constablaei* flowers have several desirable features, including large aperture and short anther-to-stigma distance. Eck and Mainland (1971) and Lyrene (1994a) suggested that short and wide corollas, short distance between stigma and anther pore and large corolla-tube apertures could make the blueberry flower more amenable to honeybee pollination, possibly resulting in higher fruit

set. The *V. ashei* flower lacked these features but might be altered by crossing with *V. constablaei*. In the present study, mean values for all flower parameters measured in the F₁ (*V. ashei* x *V. constablaei*) hybrids were intermediate between the two parents.

Vaccinium ashei and *V. constablaei* also differed in petal color (Table 2) and stigma position in relation to the corolla apex (Table 3). *Vaccinium ashei* clones tended to have white petals and stigmas exserted beyond the corolla apex. *Vaccinium constablaei*, on the other hand, had light-yellow flowers and stigmas that were even with the corolla apex. It is not known what effect these two flower characteristics have on insect attraction and pollination in blueberries. F₁ hybrids had some variation in petal color and stigma position relative to the corolla apex as a result of segregation; therefore, selection in the F₁ generation should be possible.

The small corolla tube apertures and stigmas exserted beyond the corolla apex indicated that *V. ashei* flowers were not amenable to pollination by insects such as honeybees that must penetrate the flowers (Cane et al., 1993). Instead, *V. ashei* flowers seem best adapted for pollination by sonicating bees, some of which are native to the *V. ashei* natural habitats. Such insects are very effective in species like blueberries, whose flowers shed pollen through terminal pores in the anthers in

response to vibrations caused by the flight muscles of sonicating insects (Corbet et al., 1988). Sonicating bees are effective pollinators of *V. corymbosum* (highbush blueberries), which also seem to be pollinated efficiently by honeybees (Goodman and Clayton-Green, 1988). Highbush cultivars vary in the attractiveness of their flowers to honeybees. This may be related, in part, to differences in the amount of nectar produced (Jablonski et al., 1985; Marucci and Moulter, 1977). However, honeybee efficiency in pollinatingighbush blueberry can also be explained in part by the large corolla width and wide aperture of the flower (Table 1). These two characteristics make it easier for honeybees to obtain pollen and nectar, consequently increasing pollination. Morphological features of *V. constablaei* flowers, including the large corolla apertures and stigmas at the same level of the corolla apex, could indicate that these flowers are adapted to both sonicating and nonsonicating pollinating insects. *Vaccinium constablaei* and *V. simulatum* are quite distinct in plant form. *V. constablaei* is highly rhizomatous and less than 2 m tall, with hundreds of stems per mature clone. *Vaccinium simulatum* is a taller shrub, growing up to 4 m tall with fewer canes per clone. Both *V. constablaei* and *V. simulatum* are native to the mountains of North Carolina and their flowers are much alike. The flowers of both species may have evolved in harmony with the pollinating insects native to those mountains. The difference in chromosome number (*V. constablaei* is hexaploid and *V. simulatum* tetraploid) and the fact that *V. constablaei* is probably more tolerant of fire and less tolerant of shade are two factors that may help keep the species distinct in nature.

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