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Ageratum L. 'John Eustice': A New Vigorous Lavender–blue Flowered Summer Annual

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The genus *Ageratum* is comprised of ≈29 species in the Asteraceae that are native to the Americas (Johnson, 1971). *Ageratum houstonianum* (commonly called ageratum or floss flower) is the predominant commercially grown species and is marketed and sold primarily as a spring bedding plant and to a lesser degree as a cut flower (Stephens, 2006). Floss flower can be a short-lived perennial in frost-free areas (USDA cold hardiness zones 9 and warmer) and in practice is grown as a summer annual. Ageratum does well in part to full sun and in a range of soil types with moderate fertility and moisture. Most bedding plant cultivars have a compact plant habit (typically growing 10 to 25 cm tall) with capitulums held close to the foliage. Cut flower cultivars have been selected for taller-growing plants (typically growing 40 to 60 cm tall) with strong stems and longer internodes that more prominently elevate and display the inflorescences.

Ageratum houstonianum is native to Mexico and Central America and has escaped cultivation and naturalized in multiple warm regions of the world (Johnson, 1971; Stephens, 2006). Wild collections and most cultivars are diploid ($2n = 2x = 20$), whereas some cultivars are polyploid (Johnson, 1971; Sakata® Ornamentals, 2013; Stephens, 2006). Sporophytic self-incompatibility is present in this species and is the result of a single locus with a linear dominance series of alleles (Stephens et al., 1982). Self-incompatibility and also nuclear controlled male sterility provide breeders with effective tools to facilitate controlled crosses (Stephens, 2006) and are particularly useful because individual florets are very small and tedious to emasculate.

Ageratum houstonianum and its relatives produce terminal inflorescences with capitula containing only disk florets and are typically arranged in a compound cyme. Disk florets in a single capitulum open in concentric rings from the perimeter to the center over ≈1 week. The corolla of an individual floret is funnelform with typically a white base and colored tip. The corolla ends in five deltoid lobes and the adaxial and abaxial sides are typically similar in color. However, in some genotypes, lobe color can be markedly different on either side leading to a bicolored effect to the capitulum between open and unopened florets. The style is prominent and forked and typically the same color as the adaxial side of the corolla lobes. Plants produce oppositely arranged ovate, deltoid, or slightly cordate leaves on vegetative growth and transition to alternate arrangement as the stem produces reproductive tissue.

Ageratum houstonianum is generally easy to grow, floriferous, and it does not typically have significant pest issues, all of which contribute to its popularity. Currey et al. (2011) list several annual bedding plant species categorizing them for floral induction, and the ageratum cultivars surveyed were all facultative long-day plants and supplemental irradiance did not impact induction. Scheduling ageratum to produce flowering and salable plants is therefore straightforward and relatively easy for growers. Periodic challenges that can be encountered when producing ageratum include root rot diseases in overly saturated soils and infestations of thrips and whiteflies in the greenhouse. Outdoors in amenable soil with adequate light, water, and drainage, ageratum is typically a very dependable ornamental. Ageratum is also quite resistant to herbivory including Japanese beetles, a problematic insect in the eastern United States that feeds veraciously on a wide host range of landscape plants (U.S. Department of Agriculture, 2004). Ageratum cultivars are available in a range of flower colors including lavender–blue, lavender, pink, white, purple, and mauve, but the most popular color of ageratum is lavender–blue. Lavender–blue cultivars are particularly popular because that is a difficult color to find in other flowering bedding plants, especially ones adapted to flowering continually in regions with long warm summers.

In the past decade, elite genotypes of primarily compact growing *A. houstonianum* have been introduced as vegetatively propagated cultivars and are included in popular branded plant lines. Examples include the Artist® (Proven Winners®) and Patina™ (Syngenta®) series of ageratum, which are widely available in independent garden centers and box stores as bedding plants in the United States in spring. Typically sold in ≈10-cm pots or in mixed patio containers, vegetatively propagated ageratum cultivars are typically grown to a relatively large size and tend to be more expensive than the seed grown cultivars typically sold as smaller plants in packs. However, exceptional performance for traits like heat tolerance, strong continual bloom, or very symmetric plant habit in many vegetatively propagated ageratum genotypes provides added value and helps justify the extra expense.

Origin

An ageratum breeding program was initiated in 2002 by John Eustice and David Zlesak in St. Paul, MN, which led to the development of 'John Eustice'. Several seed-propagated *A. houstonianum* cultivars were acquired (e.g., members of the Hawaii series and 'Blue Danube' and 'Blue Mink') as well as wild-collected germplasm as parental material. The wild species genotypes used as parents were obtained from multiple sources including a potted plant of *A. littorale* A. Gray purchased from a Florida native plant nursery and multiple seed accessions that were not *A. houstonianum*. The identity of these wild species obtained as seed could not be confidently determined by the authors. Interspecific hybrids were generated between the *A. houstonianum* cultivars and the various lavender–blue flowered, generally tall-growing *Ageratum* spp.

The primary objective of the breeding program leading to the development of 'John Eustice' was to combine the very strong butterfly attraction, glossy foliage, vigorous growth, and loose branching habit that elevates and separates the capitula found in some of the wild species parents with the wide range of flower color, early flowering, larger capitulum size, and attractive plant habit of commercial *A. houstonianum*. Superior F_1 and subsequent advanced generation genotypes were selected, vegetatively propagated through herbaceous stem cuttings to preserve them, and used as parents in successive generations. During the first few years of the breeding program, pedigree breeding was used. Crosses were strategically made to obtain progeny possessing combinations of unique and desirable traits. This included recovering recessive flower colors to have the full range possible in ageratum, obtaining well-branched plants with desirable growth habits, selecting plants

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with smoother leaves and stems, and prioritizing early and continued flowering.

As the breeding program progressed and led to a collection of genotypes with a combination of desired phenotypes, the primary breeding method shifted from pedigree breeding to primarily recurrent mass selection. Seed was saved and bulked from clonally propagated interspecific advanced selections grown adjacent to each other in outdoor garden beds. The original parents and interspecific hybrids in this breeding program are highly self-incompatible. Viable seeds do not form when plants are isolated indoors without vectors for cross-pollination, but viable seed does form when multiple genotypes are grown near each other outdoors and butterflies and other pollinators are present. By shifting away from a traditional pedigree breeding method, it has become unfeasible to know the exact pedigrees of seedlings in advanced generations. Using recurrent mass selection, seedlings were raised from open-pollinated seed bulked from multiple female parents that shared similar, desirable traits (i.e., glossier foliage, clear and attractive flower colors, early flowering, etc.).

'John Eustice' germinated in April of 2010 from seed collected and bulked from unreleased interspecific lavender–blue flowered selections during the summer of 2009. The population leading to 'John Eustice' was germinated and transplanted into packs indoors and then transplanted into outdoor garden beds in late spring in Woodbury, MN. During the summer of 2010, 'John Eustice' was identified as a superior genotype with exceptional floral traits and plant habit. It was first vegetatively propagated in Sept. 2010 by herbaceous stem cuttings. Over subsequent clonal generations and continued trialing in both containers and ground beds, the valuable characteristics of this genotype proved to be stable.

Fig. 1. 'John Eustice' ageratum 14 Sept. 2013 in an outdoor garden bed in Roseville, MN, from a 5 July 2013 planting date displaying abundant and well-spaced capitula.

Description and Performance

'John Eustice' is unique from other vegetatively propagated ageratum cultivars because of a combination of multiple valuable traits. They include: a vigorous, mounded, and slightly spreading plant habit; relatively larger plant size than most bedding plant ageratum cultivars; well-branched plant habit with strong stems that resist lodging; lavender–blue capitulums held prominently above the foliage; relatively glossier foliage than typical ageratum cultivars; strong butterfly appeal; early and continued flowering throughout the growing season; and a colorful and long-lasting cut flower (Fig. 1). When used as a cut flower, blooms have remained attractive for ≈7 to 10 d, a typical and consistent duration the authors found for ageratum generally.

During the summer of 2013, a field trial was conducted to compare the growth and flowering characteristics of six commonly available lavender–blue *A. houstonianum* cultivars with 'John Eustice' (Table 1). 'Agpatblue' (Patina™ Blue) and 'Agsantis' (Artist® Blue) are vegetatively propagated, patented cultivars and were purchased as young plants. The additional comparison cultivars (Blue Horizon, Field's Blue, Hawaii Blue, and Tycoon Blue) are commercially propagated by seed. To standardize the trial, cuttings were taken and rooted of seed-propagated cultivars so everything going into the comparative trial was propagated using the same method. Going into the trial, plants were 4 weeks old or older. Before planting, all plants were cut back to ≈5 cm. Planting occurred on 5 July 2013 in Roseville, MN, using a randomized complete block design with three blocks and one replication of a row of four adjacent plants of a cultivar per block. Plants were planted 25.4 cm apart both between and within rows. The soil was a sandy loam. The bed was mulched with ≈5 to 7 cm of hardwood chips, irrigated as needed (plants received at least 2.54 cm of water per week from either rainfall or irrigation), and fertilized at the recommended rate with a granular quick-release fertilizer (10N–4.4P–8.3K; Ideal™ All-purpose Fertilizer; Eau Claire Cooperative, Eau Claire, WI).

Data from the cultivar comparison were collected 14 and 15 Sept. 2013. Data were taken on the two most developed plants per replication (each replication consisted of a row of four plants). The data were averaged over the two plants per replication to get a single value per trait per replication for descriptive statistics, analyses of variance, and mean separation (Duncan's multiple range test; $P \leq 0.05$). Data collected on an overall plant basis were plant height, plant width (widest diameter was recorded along with the diameter perpendicular to it and the two values were averaged), and number of stems ending in inflorescences with at least one capitulum containing an open floret. Representative samples of three measurements per plant were taken for: the number of capitula per inflorescence, diameter of capitula possessing open florets, length from the top of the inflorescence to the first node with opposite leaf arrangement, leaf blade length and width, petiole length, and internode length between the first two adjacent nodes with opposite leaf arrangement below the inflorescence (Table 1). Color data were recorded for unopened and opened florets using representative half open capitula of each cultivar and the color charts from the Royal Horticultural Society (2001) (Table 2). Ploidy was determined for each cultivar through direct chromosome counts of root tip cells at metaphase using the procedure outlined in Zlesak et al. (2005).

'John Eustice' and 'Blue Horizon' were significantly larger-growing than the other ageratum cultivars and had inflorescences displayed significantly higher above their foliage (Table 1). Comparing the two largest growing cultivars, 'John Eustice' had more inflorescences per plant than 'Blue Horizon' and leaf blades that were smaller, both valuable traits for use of 'John Eustice' as a cut flower cultivar. 'John Eustice' had significantly more capitula per inflorescence than all the other cultivars. When estimating the overall number of capitula per plant (multiplying the mean inflorescence number and number of capitula per inflorescence), 'John Eustice' had the most followed by 'Agsantis' and then 'Agpatblu'. The plant habit of 'John Eustice' was more similar to the cushion habit of the compact cultivars, which had capitula displayed over most of the surface of the hemispherical plants (Figs. 1 and 2). 'Blue Horizon' had blooms displayed primarily at the top of the plant (Fig. 2).

The color varied between unopened and opened florets for these cultivars all marketed as having blue flower color (Table 2). 'Blue Horizon' maintained the most consistent lavender–blue color from unopened to opened florets. Most of the rest of the cultivars possessed a relatively similar lavender–blue color between unopened and opened florets, including 'John Eustice' (Fig. 3). 'Agpatblu' and 'Agsantis' displayed the greatest color differences between unopened and opened florets with opened florets being lavender–blue and unopened florets being lavender–pink.

Chromosome counts revealed that 'John Eustice' and all but one of the other cultivars were diploid ($2n = 2x = 20$). 'Blue Horizon' was triploid ($2n = 3x = 30$), which is consistent with its marketing (Sakata® Ornamentals, 2013). Polyploids compared with their diploid counterparts often display