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BREEDING OF NEW *HIPPEASTRUM* CULTIVARS USING DIPLOID SPECIES I. THE F-1 EVALUATION

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Abstract. **An amaryllis breeding program using diploid species not well represented in current commercial tetraploid cultivars has been underway since 1988. Objectives are to develop evergreen cultivars with attractive foliage and fragrant flowers of novel floral form and coloration. F-1 progeny between *Hippeastrum papilio (Amaryllis papilio* Ravenna[1]) and, respectively, *H. lapacense (A. lapacensis* Cardenas), *H. cardenasianum (A. cardenasiana* Traub & Doran), and *H. vittatum* Herbert 'Tweedianum' have been successfully produced, and selections made during their first flowering season (February through June, 1990). Over two hundred progeny have been evaluated. Sibling, interhybrid and new primary hybrid crosses with *H. papilio, H.* 'Tucamanii', *H. brasilianum (A. brasiliana* Traub & Doran), *H. fragantissimum (Amaryllis fragrantissima* Cardenas) [these latter three all white, heavily fragrant, trumpet-flowered species], and *H. reticulatum* Herbert var. *striatifolium* Herbert as the seed and pollen parents have been generated. Ten to twenty percent of the seedlings from all new crosses are being treated with colchicine in an attempt to induce polyploidy.**

Hippeastrum Herbert, the amaryllis, consists of ca. 60 entirely New World species (26). The species are concentrated in two main centers of diversity: in eastern Brazil and the central- southern Andes of Peru, Bolivia and Argentina, on the eastern slopes and adjacent foothills. Little of this genetic diversity is represented in modern amaryllis hybrids. Primary hybrids were produced from a relatively small number of species, mainly *H. vittatum* Herbert, *H. leopoldii* Dombrain, *H. pardinum* (Hook. f.) Lemaire, *H. reginae* Herbert, *H. puniceum* (Lam.) Kuntze and *H. aulicum* Herbert (2, 10, 21, 24, 25). *Hippeastrum* x 'Johnsonii,' generally acknowledged as the first amaryllis hybrid, was a primary hybrid of *H. vittatum* and *H. reginae* (25). Commercial breeding efforts have always emphasized large flower size which is attributable genes from *H. leopoldii* and *H. pardinum* (2, 24). Commercial breeding efforts subsequent to the initial flurry of primary hybridization over a century ago has largely been concentrated among the hybrid populations, leading to a greater complexity of parentage (with little documentation) and dilution of many of the unique characteristics of the original component species (2, 3, 10, 21, 24).

Using a number of interesting diploid species, we have begun a breeding program directed towards the following goals: novel attributes of flower form (e.g.: trumpet or long-tubed perianth, novel pigmentation patterns), reintroduction of fragrance, high quality evergreen foliage, repeat bloom, and more than 3 flowers per scape. Concurrent with our breeding program, we are establishing a collection of *Hippeastrum* germplasm in association with Brazilian and domestic colleagues. Attempts to increase the supply of rare species via tissue culture are underway as well. Ultimately, we endeavor to develop a breeding gene pool largely unrelated to the genetic stock now represented in the commercial amaryllis production industry, largely dominated by the Dutch.

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[1]Following the decision of the Nomenclature Committee of the International Association for Plant Taxonomy, *Hippeastrum* Herbert is recognized as properly applied to the neotropical genus discussed in this paper, and *Amaryllis* L. as a monotypic genus of South Africa. As formal transfers into *Hippeastrum* of many species described as *Amaryllis* have yet to be made, the valid name and authority is given in parenthesis wherever necessary.

Materials and Methods

Reciprocal F-1 progeny between *H. papilio (Amaryllis papilio* Ravenna) and, respectively, *H. lapacense (A. lapacensis* Cardenas), *H. cardenasianum (A. cardenasiana* Traub & Doran), and *H. vittatum* Herbert 'Tweedianum' were accomplished in 1988. Five hundred seedlings were planted in a trial field in May, 1989 that receives full sun from 11 AM to sunset, with approximately 10% of each cross maintained in containers under 63% shade.

Results and Discussion

Five crosses were evaluated during their first flowering, less than 18-24 months from seed. Flowering began in February 1990, peaked in April and continued sporadically through July. With the advent of cooler night temperatures in October-November, a number of the progeny are producing scapes. Over two hundred seedlings have been evaluated. Since primary interest is on clones capable of flowering without a period of dormancy, irrigation has never been withheld from the trial field. Results of the F-1 are summarized below and in Table 1. Overall selection criteria include flower size, flower number (≥ 3 being desireable), repeat bloom, and leaf persistance.

H-1: *H. papilio* x *lapacense*: "butterfly" form, red to maroon freckling and striations overlying a white to green background. A few clones have upper and lateral tepals deeply suffused with red. Most show the red picotee of the papilio parent. Foliage quality is very good, dark green and persistant. Selection criteria: pure red rather than maroon, pure white background, wide petals.

Table 1. Summary of the 1990 Amaryllis F-1 Evaluation (As of 7/1/90).

CROSS	TOTAL EVALUATED	Scapes Produced					Number of Clones With 3 or More Flowers Per Scape	Grade[z]		
		1	2	3	4	5		Good	Fair	Poor
H-1	54	31	21	2			15	12	40	2
H-2	53	19	30	4			9	14	32	7
H-4	39	16	13	9	1		4	15	21	3
H-5	27	13	10	1	2	1	7	4	20	3
H-8	33	16	12	1	4		8	7	25	1

[z]Evaluations based on selection criteria described in text.

H-2: *H. lapacense* x papilio: similar to H-1 in form, color, and foliage, but a fair number more lightly striated over the white or green background. Selection criteria as for H-1.

H-4: *H. vittatum* 'Tweedianum' x *papilio*: quite variable, about half with trumpet-shaped flower, the other half more open-faced. Most with red picotee. Several have a unique purple pigmentation over a white background. Some very robust and vigorous growers. Foliage quality good, lighter green than H-1 or H-2. Selection criteria: trumpet shape, purple color, white background.

H-5: *H. papilio* x *vittatum* 'Tweedianum:' similar to H-4. Most tend to show picotee from *papilio* parent. Long-lasting flower of heavy substance. Selection criteria: spreading petals, white background.

H-8: *H. cardenasianum* x *papilio*: very elegant flower form, semi-star. Pink to maroon striations over a white to light green background. Excellent foliage quality, including some very compact clones. Flowers of medium substance. Selection criteria: less green in background, clear red or pink pigmentation.

Primarily inter-hybrid crosses have been made using oustanding selections from the F-1 progeny. Second generation crosses between H-8 selections and outstanding progeny of H-1 and H-2 respectively are expected to yield promising results. Despite partial self-compatibility in *H. papilio,* all progeny have proven self incompatible, thus preventing access to F-2 segregation. For reasons yet undetermined, most sibling crosses failed to set seed. New hybrid populations have been generated with *H. papilio, H.* 'Encore' and 'Pink Ambrosia' (the latter two heavily fragrant, trumpet-flowered hybrids); a number of hybrid selections from the breeding program of Fred Meyer, Escondido, CA involving *H.* 'Tucamanii' (an undescribed species), *H. brasilianum (A. brasiliana* Traub & Doran), *H. fragantissimum (Amaryllis fragrantissima* Cardenas), all fragrant, trumpet-flowered species; and *H. reticulatum* Herbert var. *striatifolium* Herbert (a dwarf species with a novel and cross transmitable white leaf midrib) as the seed and pollen parents. As of this writing, 38 seconds generation crosses have been accomplished and nearly 2000 seedlings are in production. We estimate flowering these seedlings in fall and/or spring of 1992. A number of the *H. papilio* x *reticulatum* var. *striatifolium* hybrids are exhibiting the white midrib of the pollen parent. The new leaves of these hybrids emerge an attractive bronze to almost red color. A new trial bed will be developed in early 1991 under 30% shade.

In crosses now setting, approximately 10-20% of the germinated seedlings have been or will be treated with colchicine using the methods of Williams (27) in an attempt to induce tetraploidy in some of the progeny. Germinated seedlings are inverted to the midpoint of the seedling bulbil in 0.05% colchicine in agar for 24 hours. Root tip mitotic metaphase preparations will be used for determination of ploidy level (20).

Superior selections from the breeding program will be increased through tissue culture using the twin-scaling method (1, 6, 22, 23) and tested for both pot crop and landscape potential. One H-8 clone has yielded vigorous rooted plantlets in 4 weeks from a twin scale expant. Irradiation of aseptic subcultures of the superior F-1's and second generation progeny will also be attempted (16).

It is imperative that a programming schedule for flower induction and development be formulated before any cultivar is released from the program. There is literature on plant growth regulator (4, 18, 19), environmental (5, 7, 9, 11, 12, 13, 14, 17) and nutritional (8, 15) effects on flower forcing in *Hippeastrum,* expressly relating to the complex hybrid material that currently dominates the trade. Experiments will be conducted to determine the protocols necessary to force selections from the breeding program. Additional studies underway include an investigation into the relationship between ammonium nutrition and susceptibility to bulb rot diseases.

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CHLOROPHYLL LEVELS AND ANATOMY OF VARIEGATED AND NONVARIEGATED AREAS OF *AGLAONEMA NITIDUM* LEAVES

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Abstract. **Chlorophyll level determinations and anatomical analyses of leaf tissue from two variegated *Aglaonema* cultivars and eight of their hybrids were performed to compare variegated versus non-variegated leaf tissue. Plants with four phenotypes (three distinct variegation patterns and one non-variegated type) were studied. No differences in cholorphyll levels were detected. Anatomical observations showed that variegation was due to intercellular air spaces between the upper epidermis and the mesophyll.**

Aglaonema, members of the family Araceae, are native to Southeast Asia where they grow in the low light of the tropical rainforest floor. Several cultivars are used as ornamental foliage plants because they possess attractive foliar variegation patterns and are well-adapted to light levels encountered in interior environments.

Previous genetic studies have shown that five distinct foliar variegation patterns in Aglaonema are controlled by the V-gene and its series of codominant alleles (4). Individual variegation patterns are determined by separate alleles, any two of which could be expressed in a single plant to produce compound patterns. The homozygous recessive genotype (vv) yields nonvariegated plants (Table 1). Most foliar variegation can be characterized into two basic categories. True variegation is demonstrated by foliage having areas of little or no pigmentation, while other types are due in part to anatomical features, i.e. hairs, scales, or air spaces in the cuticle or epidermal cells.

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Research performed on the Aroid *Epipremnum aureum* 'Marble Queen' (2), whose variegation is of the type which lacks pigmentation (unpublished observations), indicates that as the level of variegation increases, plant growth decreases. Additionally, the higher the degree of variegation the greater the sensitivity to chilling temperatures.

Few reports regarding the nature of foliar variegation in *Aglaonema spp.* are available (5,7). This study was conducted to determine if variegation in the cultivar named was due to differences in chlorophyll levels and/or anatomical features.

Materials and Methods

Two cultivars of *Aglaonema nitidum* (Jack) Kunth, each with prominent foliar variegation patterns, and eight plants of their hybrids were chosen for this study. *A. nitidum* 'Curtisii' displays variegation consisting of silvery-grey stripes overlaying the leaf veins, while *A. nitidum* 'Ernesto's Favorite' has a broad silvery-grey swath occupying the central longitudinal portion of the leaf (Fig. 1). The eight hybrid plants included two representatives of each of four different variegation patterns from crosses of 'Curtisii' and 'Ernesto's Favorite'. The patterns included: 1. parental 'Curtisii'; 2. parental 'Ernesto's Favorite'; 3. combinations of both parental patterns; and 4. non-variegated. Mature stock plants with a minimum of 2 stems were grown in a shaded greenhouse under natural daylength with maximum light levels of 230 $\mu mol \cdot s^{-1} \cdot m^{-2}$ (1200 Ft.C) and a temperature range of 18 to 32°C (65 to 90°F). Plants were maintained in 20 cm diameter pots (3.86 liter vol) and fertilized with 10 g Osmocote™ (19-2.6-10 N-P-K; 19-6-12 N-P_2O_5-K_2O) (Grace/Sierra Co., Milpitas, CA) every three months.

Chlorophyll Content. Leaf samples for chlorophyll analysis were collected as follows: leaves of the same physiological age, one from each of two separate stems, were sampled from each pot. Sampled leaves were fully mature and fourth down from the newest expanding leaf