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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on the Aroid *Epipremnum aureum* 'Marble Queen' (2), whose variegation is of the type which lacks pigmentation (unpublished observations), indicates that as the level of variegation increases, plant growth decreases.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

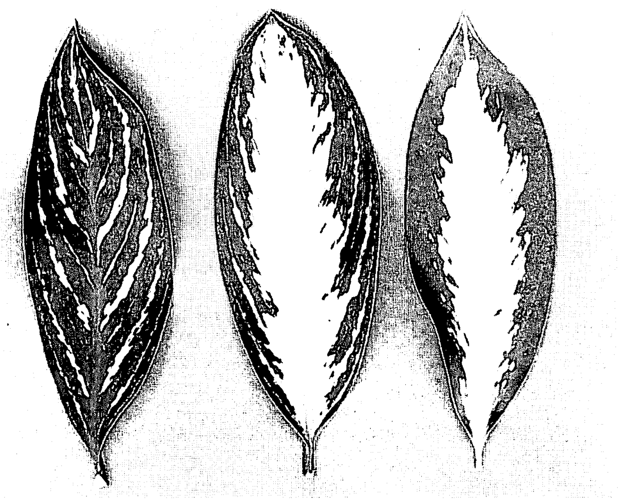

Fig. 1. Variegation patterns of *Aglaonema nitidum* (L to R) 'Curtisii', 'Curtisii' + 'Ernesto's Favorite', and 'Ernestos Favorite'.

at the apical growing point. Ten leaf discs, 0.5 cm diameter (0.20 cm^2) were taken from each variegated leaf, five from within and 5 adjacent to a variegated area, using a #2 cork borer. The hybrids expressing both parental patterns were sampled where the patterns overlapped and in green areas. Only five leaf discs were taken from leaves of non-variegated plants.

Total chlorophyll content was determined using modifications of procedures by MacKinney (6) and Arnon (1): each group of five discs was placed into vials with 5 ml of cold acidified (10% HC1) methanol and dark extracted for 48 hr at –15C. Optical density was measured at 652 nm using a spectrophotometer (Spec 20, Bausch and Lomb) and readings were converted to $mg \cdot l^{-1}$ using the formula:

$$(OD \times 1000)/34.5 = mg \cdot l^{-1}.$$

Anatomical studies. Tissue sections containing both green and variegated areas were removed from leaves of each phenotype, fixed in F.A.A., dehydrated and embedded in paraffin. Sections were cut on a rotary microtome at 10 microns, and stained in toluidine blue O.

Table 1. Chlorophyll levels in variegated and nonvariegated areas of leaves from *Aglaonema nitidum* 'Curtisii' and 'Ernesto's Favorite' and their hybrids.

Variegation Phenotype	Variegation Genotype	Chlorophyll Concentration[z] Green	Chlorophyll Concentration[z] Variegated	Row Sig.[y]
Parental Types				
Curtisii	Vv	10.03	9.35	NS
Ernesto's Favorite	$V^{ef}v$	9.89	9.53	NS
Hybrids/Phenotypes				
Curtisii	$V^{c}v$	9.67	9.31	NS
Ernesto's Favorite	$V^{ef}v$	8.99	8.71	NS
Curtisii & Ernesto's Favorite	$V^{c}V^{ef}$	9.17	8.91	NS
Green (Non-Variegated)	vv	8.84		
Column Significance[y]		NS	NS	

[z]Concentration given as $mg \cdot l^{-1}$
[y]Significance determined using f-test at 5% level; NS = Not significant

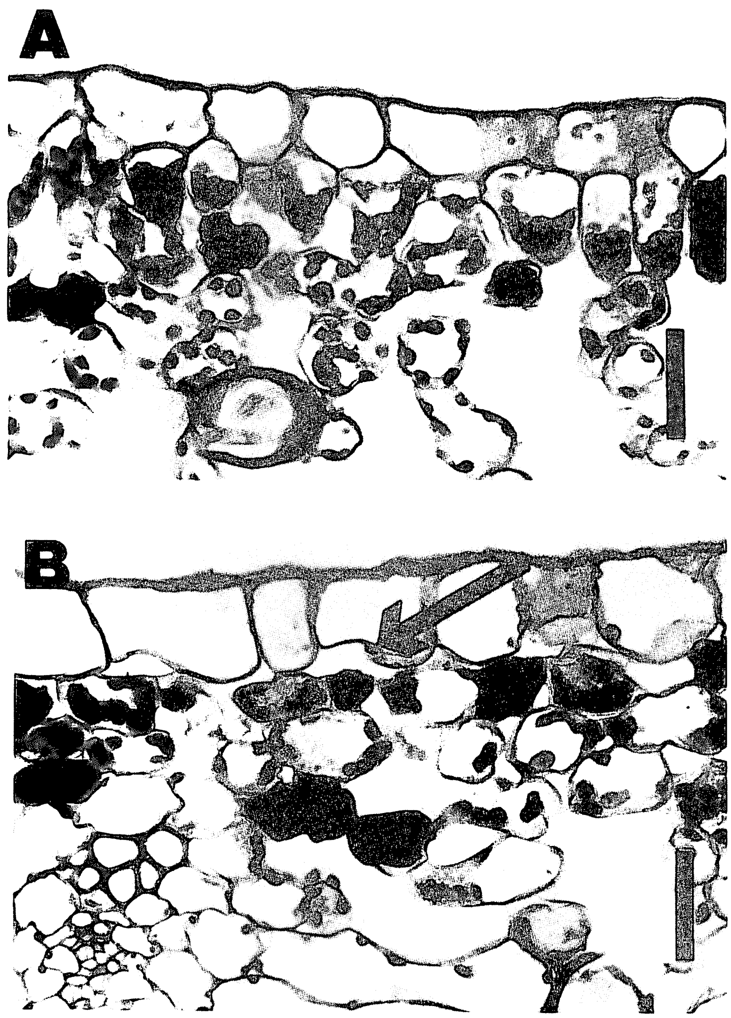

Fig. 2. Cross-sections of *Aglaonema nitidum* 'Curtisii' leaves: A. non-variegated tissue, B. variegated tissue, arrow indicates air space. Scale bar = 50 μm.

Results and Discussion

Total chlorophyll levels whether from variegated or nonvariegated tissue, and regardless of phenotype, were not statistically different (Table 1).

Microscopic and macroscopic examination of leaf sections showed air spaces between the epidermis and the mesophyll in the variegated tissues and absent in the green tissues (Fig. 2). This observation was illustrated by the fact that the epidermis over variegated tissue, being more loosely attached, could be easily peeled off in large segments as compared to green tissues where epidermal peels were not possible.

The presence of such air spaces causes incoming light to be scattered at the air-cell interface prior to reaching the chloroplasts and results in a portion of incoming light being reflected back through the epidermis creating the distinctive silvery-grey markings of the upper leaf surface. Light reflected back in this manner would be unavailable for photosynthesis.

Microscopic analysis also showed that chloroplasts in variegated tissues were more randomly distributed than in green tissues. The mesophyll cells in the green tissue had a pallisade-like layer, whereas the variegated area was composed of isodiametric parenchyma cells. Cellular and chloroplast arrangement in green areas of *A. nitidum* correspond to that in *Alocasia macrorrhiza* grown in high light treatments, while the cellular and chloroplast arrangement in variegated areas corresponded to that in leaves of *Alocasia macrorriza* grown in low light (3). Whether variegation in Aglaonema diminishes its ability to utilize light, provides for greater efficiency under rainforest conditions or has some other function remains to be determined. It is possible that variegated leaves of *Agloanema* may have the ability to utilize whatever light is able to penetrate the canopy of the rainforest, be it low intensity diffuse light or high intensity sun flecks.

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VEGETATIVE PROPAGATION OF FLORIDA NATIVE PLANTS: V. *PRUNUS* SPP.

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Additional index words. Prunus caroliniana (Cherry Laurel), *P. serotina* (Black Cherry), and *P. umbellata* (Hog Plum).

Abstract. **Species of the genus *Prunus*, particularly *P. serotina* Ehrh. (black cherry), have long been recognized as excellent landscape, timber, and wildlife plants. Their propagation by seeds, which requires 120 days of cold stratification, has been known for many years. However, propagation by cuttings has not been reported. There appears to be a direct correlation between flowering/leaf expansion time and root initiation. The two species reported in this study differ in one major respect: *P. serotina* flowers after leaf expansion in April, whereas *P. umbellata* Ell. (hog plum) flowers before the leaves emerge in March. Consequently, best rooting in black cherry is obtained in March, after leaf expansion but before flowering, while that of hog plum is in April, after flowering and leaf expansion. In both cases, IBA treatment promoted rooting of cuttings.**

Several *Prunus* species, but particularly *P. serotina* Ehrh. (Black Cherry), have long been recognized as good timber trees and wildlife plants (8). *Prunus serotina* is a large (50 to 110 ft.) attractive deciduous tree with ascending branches, dark shiny green foliage, and small but numerous white flowers on long racemes. It is a common plant in much of Florida, characteristic of upland hardwood hammocks but also in mixed hardwood and pine, and flatwoods (1, 14). Despite excessive fruit production which may become a minor nuisance when ripe, it is considered an excellent plant for landscaping and for revegetation of disturbed sites because of its wide adaptability to adverse edaphic conditions. The reddish-brown wood has a close grain and is particularly suitable for furniture manufacturing, interior trims, veneers, and tool handles (8). In contrast, except for occasional use in making jelly and jams from the fruit, *P. umbellata* Ell. (Hog Plum) has litte economic value but is an excellent landscape plant because of its early spring flowering, crooked trunk, and spreading branches. The plant is deciduous, and the comparatively large white flowers in umbellate (hence the name) inflorescences appear before the leaves emerge. The large (1/2 to 2/3 inch) fruit is red to black and matures in late summer, providing food for birds and other wildlife (8). A third Florida native species, *P. caroliniana* (Mill.) Ait. (Cherry Laurel), a densely foliated evergreen species, is among the most attractive of all *Prunus* species for landscaping because of its dense foliage and wide adaptability. Although the seeds of all three species have a tendency toward spontaneous germination, *P. caroliniana* has a much greater inclination toward weediness. For this reason and because its mode of propagation is well-known (7) it was not included in this study.

Other than the recent enumeration of propagation methods for several *Prunus* species by Dirr and Heuser (7), only a few studies have dealt with this economically and environmentally important genus. Spellerberg (13) examined the effect of rooting conditions, addition of CO_2, extended daylength, and GA_3 treatment of *P. triloba* cuttings. Shoot growth was enhanced by all treatments, but rooting was not significantly improved. Cuttings of *P. persica* (Peach), were reported to root best in June when taken from the branches that were bent in the previous January and cuttings treated with 3000 and 4000 ppm IBA (9). Hardwood cuttings of *Prunus* 'Golden Queen' had 85% rooting when taken during winter, treated with 1000 ppm IBA, and kept on bottom heat at 23° C for six weeks (12). It has been shown in earlier studies (2, 11) that rooting of *Prunus* spp. is dependent on the time of year when cuttings are taken, usually in late spring or early summer, with the

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