resulted in a decrease in growth of dark grown calli. This is especially apparent in treatments receiving cytokinin alone, but can be seen in a comparison of most weight gains in the light to the same treatment in the dark (Table 2).

Our results indicate that substantial levels of ethylene may be accumulated in sealed vessels where tissue is kept for an extended period of time, and that the type and concentration of growth regulating compounds employed may strongly influence ethylene synthesis. The synergistic effect on ethylene production is not unique to rose callus tissue and has been observed by other workers (6). Different auxins and cytokinins, although resulting in equally good growth, elicit various degrees of ethylene evolution, but growth of rose callus tissue was not adversely affected by increased ethylene levels.

Although at higher cytokinin concentrations fresh weight gain of calli was greater in light than in darkness, no statistical difference between light and dark treatments with regard to ethylene evolution was observed. This suggests that the reduced growth in dark treatments was not related to increased ethylene accumulations.

Table 2. The effect of BA, DMAA and zeatin riboside with IBA or NAA on callus weight gain maintained in light (L) or darkness (D).

		Callus weight gain (mg)									
			BA			DMAA			Zeatin riboside (mg/liter)		
Auxin	Light	0(mg/liter)	5	10	15	5	10	15	5	10	15
0	L	17a[z]	207b	292d	0a	217b	150b	197b	170b	130b	129bc
0	D	32a	18a	0a	16a	46a	0a	10a	4a	30a	0a
IBA	L	271b	269bc	336d	5a	340c	371d	271c	287c	316c	327d
IBA	D	280b	52a	89b	8a	290bc	188bc	207bc	178b	270c	158c
NAA	L	393c	318c	185c	0a	452d	410d	424d	423d	262cb	305d
NAA	D	313b	230b	132bc	0a	348c	229c	243bc	172b	191b	76b

[z]Column means separated by HSD at P = 5%; means of 12 samples, 4 weight gain replications at 3 auxin concentrations.

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In Vitro Propagation of *Hosta sieboldiana*[1]

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Abstract. **Plants were initiated from florets of *Hosta sieboldiana* (Lodd.) Engler cultivars cultured on a modified Murashige-Skoog medium containing 0.5 – 2.5 mg/liter naphthaleneacetic acid (NAA) and 0.5 – 2.5 mg/liter 6-benzylamino purine (BA) and grown in the light. These plants could be separated and proliferated on a similar medium to give large numbers of plants. Plant propagated from 'Frances Williams' had a chimeral separation into green and gold sports as well as retaining the chimeral organization.**

The hosta or plantain lily is a perennial plant popular for established gardens or other shady areas in the landscape. Plants are usually propagated by crown division, thus, introduction of a new cultivar can take several years. An underground bud or "eye" can be used to propagate Hosta by tissue culture (1), however this excision reduces the growth potential of scarce new cultivars and using underground structures increases the contamination rate of explants.

Young emerging inflorescences have been a good explant source for *in vitro* propagation of other herbaceous perennials (3, 4), but young inflorescences were tried unsuccessfully with hosta (2). The florets of the inflorescence proved a satisfactory explant source for *Hosta sieboldiana,* if the flower scapes were allowed to develop until the bottom florets were separate and 0.5 – 1 cm in length. Other workers (1, 6, 8) have reported that shoot tips, florets and scape slices have worked for other hosta species.

Rapid multiplication of 3 cultivars of the large-leaved *Hosta sieboldiana,* 'Helen Doreo', 'Frances Williams', and 'Frances Williams Gold Sport', and the separation of the chimeral 'Frances Williams' was achieved with the following techniques.

Flower scapes were excised as the bottom florets became separated and 0.5 – 1 cm in length but were still tightly closed. Larger florets on more mature scapes will not proliferate plants readily. The florets still attached to the scape were disinfected in a solution of 0.5% sodium hypochlorite (10% Clorox) and 0.1% polyoxyethylene sorbitan (Tween 20) for 15 min, rinsed with sterile water, transferred to a sterile surface, and removed from the scape by cutting just below the ovary. The florets were transferred to 2.54 cm tubes, pressed half way into an agar medium, and placed under Cool White fluorscent light ca. 3 klx for 18 hr a day. Earlier studies with other herbaceous perennials (3, 4) indicated greatest plant proliferation when the flower tissue was placed in the dark with an appropriate medium for callus production. The tissue was transferred to a new medium in the light for plant proliferation. This technique reported earlier for *H. sieboldiana* cultivars caused only 10-15% of the florets to produce plants (2). Starting the florets *in vitro* in the light resulted in 50-75% of the florets producing plantlets. Papachatzi et al. (6) obtained callus from *H. plantaginea* 'Grandiflora' in the dark in a high NAA medium and formed shoots by transferring the callus to a high BA medium in the dark. These shoots then had to be transferred to a medium with no growth

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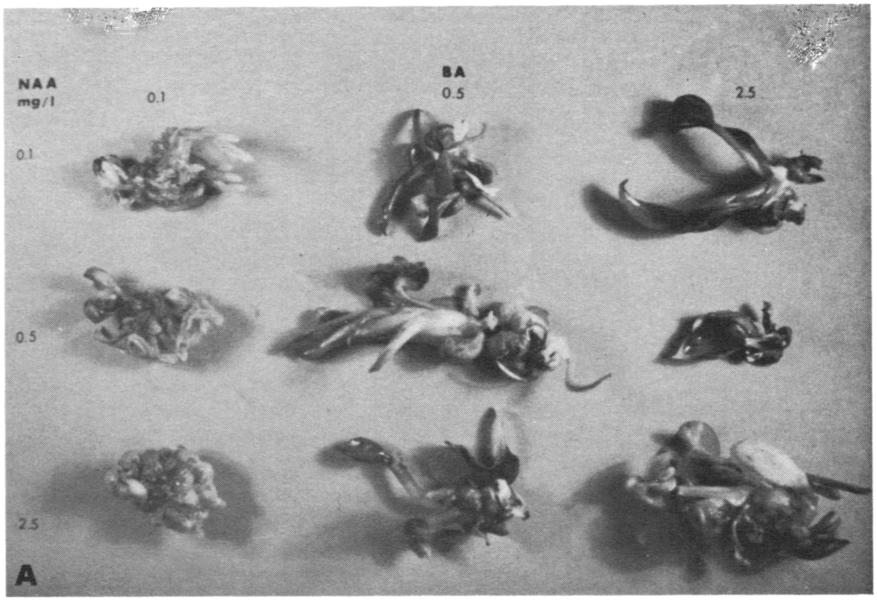

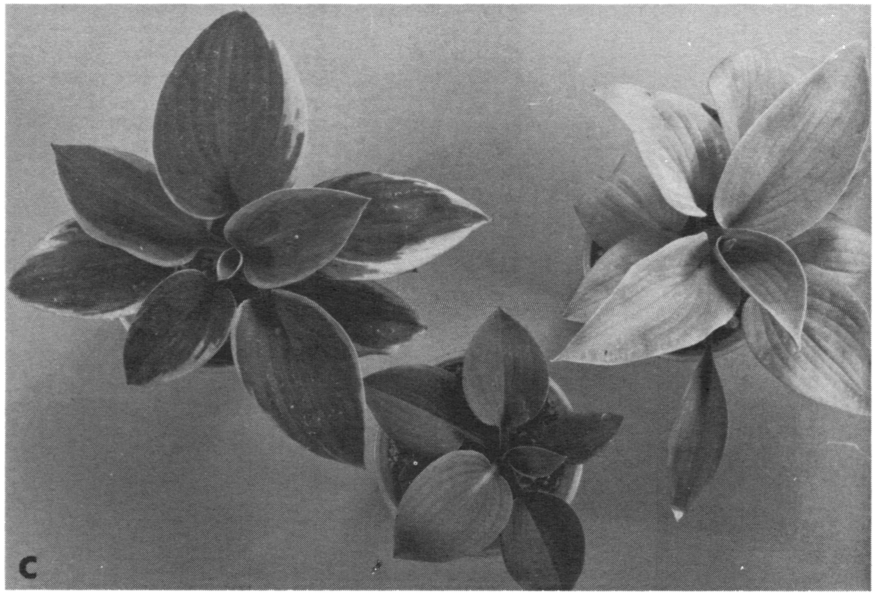

Fig. 1. (A) The influence of NAA-BA combinations on the development of plantlets and other organs from 'Frances Williams' hosta, (B) Reculturing plantlets and shoots from 'Helen Doreo' florets to transplantable sized (C) The types of plants from 'Frances Williams' florets; normal chimera type on left, green type in center and gold sport on the right.

regulators in the light to form chlorophyll and roots.

A Murashige-Skoog high salt medium (5) containing modifications of 300 mg/liter KH_2PO_4, 160 mg/liter adenine sulfate and 500 mg/liter casein hydrolysate with an auxin-cytokinin growth regulator complex was used for the hosta florets. The appropriate ranges of concentration of NAA and BA which gave good plantlet proliferation are illustrated in Fig. 1A. NAA at 0.5 – 2.5 mg/liter and BA at 0.5 – 2.5 mg/liter gave good plantlet production. The production of shoots and plantlets took 8-12 weeks. These were then separated from the flower tissue and placed on the same medium with 0.5 mg/liter NAA and 0.1 mg/liter BA for further plant development (Fig. 1B). The clumps of plantlets which developed were transferred a number of times, separating and potting off the largest plants and returning the other for reculture. Several hundred to a few thousand plants can be cultured from the florets of a single scape in one year.

When the chimeral hosta, 'Frances Williams' was cultured with these techniques, 45% of the plants obtained were variegated, 45% were a gold sport and 10% were a green sport (Fig. 1C). The gold sports of hosta are useful landscape types and as reported earlier by Vaughn et al. (7) the gold sports of hosta grow as well as the regular chimera or all green types. The gold sports do sunburn more easily and should be grown in more shady locations.

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