

Materials and Methods

Three popular *Spathiphyllum* cultivars, Viscount, Taylor's Green and Petite were obtained from a local commercial nursery. Plants had been grown in 6 inch pots to the stage where they were about to be treated with GA₃ to induce flowering (12 to 14 weeks prior to sale). Thirty plants of each cultivar were transferred to the research greenhouse at MREC 2 weeks before treatment. Plants were held on raised benches in a shade painted greenhouse (1200-1500 ft-c.) cooled with fan and pad to maintain temperatures between 95°F maximum and 70° minimum. On August 7th, plants received a single foliar spray at 0, 125 or 250 ppm GA₃. Tween 20 at 1 drop/L was added as a wetting agent. Plants were randomized in a complete block design and were observed weekly. Once flowering began, number of opened flowers was recorded weekly until the end of the experiment at 14 weeks.

Results

Fourteen weeks after GA₃ treatment, none of the 0 ppm plants had flowered (Table 1). 100% flowering was observed in Petite and Taylor's Green at both 125ppm and 250 ppm. Only 60% of Viscount flowered at 125 ppm but all Viscount treated at 250 ppm flowered. At 14 weeks after treatment Petite averaged 9.0 flowers per plant at 125 ppm and 10.2 flowers per plant at 250 ppm. Taylor's Green averaged 6.1, and 8.6 flowers per plant at 125 and 250 ppm GA₃ respectively. Viscount averaged 1.6 flowers per plant at 125 ppm and 6.9 flowers per plant at 250 ppm GA₃.

Discussion

Results indicate that both cultivar selection and rate of GA₃ application affect flowering response of *Spathiphyllum*.

Table 1. The percent flowering and total number of flowers per plant produced on three *Spathiphyllum* cultivars 14 weeks after treatment with three rates of gibberellic acid.

Cultivar	GA ₃ conc	% Flowering	Total no. flowers
Petite	0	0	0 a*
Petite	125	100	9.0 b
Petite	250	100	10.2 b
Taylor's Green	0	0	0 a
Taylor's Green	125	100	6.1 b
Taylor's Green	250	100	8.6 c
Viscount	0	0	0 a
Viscount	125	60	1.6 a
Viscount	250	100	6.9 b

*Means, of each cultivar, followed by different letters are significantly different at the 5% level; Duncan's multiple range test.

Two cultivars, Petite and Taylor's Green, flowered well at 250 and 125 ppm GA₃ although both produced more flowers at the higher rate. Viscount on the other hand did not flower well at the lower 125 ppm rate. Results also show that Petite produced more flowers than Taylor's Green or Viscount at all levels of GA₃ treatment when comparing each level individually. Results indicate that if growers desire to use lower rates of GA₃, flowering response of each cultivar must be evaluated.

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Proc. Fla. State Hort. Soc. 113:170-172. 2000.

SOMATIC EMBRYOGENESIS IN *LIMONIUM LATIFOLIUM* (SM.) O. KUNTZE, PLUMBAGINACEAE

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Additional index words. Tissue culture, micropropagation, plant regeneration.

Abstract. We are interested in studying certain stress tolerance traits in perennial sea lavender *Limonium latifolium*, a member of the halophytic plant family Plumbaginaceae. This species is

important in ornamental horticulture for fresh and dry cut flowers. Plant regeneration via somatic embryogenesis is an efficient route to propagate genetically uniform plants for conventional breeding and genetic transformation. We have developed a protocol to induce somatic embryogenesis from 5-day old cotyledon and hypocotyl explants. The explants were cultured on agar-solidified Murashige and Skoog's medium supplemented with 3% (wt./v) sucrose, 1 mg.l⁻¹ 2,4-D and 0.1 mg.l⁻¹ kinetin. Globular embryogenic calli developed in two weeks and differentiated into the cotyledonary stage embryos. Experiments are in progress to optimize in vitro embryo maturation to the seedling stage. This is the first report of somatic embryogenesis in *L. latifolium*.

Many members of the plant family Plumbaginaceae are important ornamental species. Especially, *Limonium* species (also known as *Statice*) are cultivated for fresh and dry cut

Florida Agricultural Experimental Station Journal Series No. N-01991. Acknowledgments: M. A. was supported by the Fulbright Commission, Cairo, Egypt (Grant #462/99). ²Corresponding author. E-mail brath@gnv.ifas.ufl.edu ³Current address: Genetics Department, Cairo University, Cairo, Egypt.

Table 1. *In vitro* plant regeneration in various members of the Plumbaginaceae. Abbreviations: MS = Murashige and Skoog (1962) medium LS = Linsmaier and Skoog (1965) medium, 6-BA = 6-Benzyl aminopurine, DAP = 6- γ -Dimethylallylaminopurine, 2,4-D = 2,4-Dichlorophenoxy acetic acid, IAA = Indole-3-acetic acid and IBA = Indole-3-butyric acid.

Species	Explant	Basal medium	Growth regulators	Regeneration technique	Reference
<i>L. sinuatum</i>	axillary bud	LS	0.6 mg.l ⁻¹ 6-BA	Meristem culture	Harazy et al., 1985
<i>L. perezii</i>	protoplasts	MS	1 mg.l ⁻¹ each of 2,4-D, 6-BA and NAA	Organogenesis	Kunitake and Mii, 1990
<i>L. estevei</i>	nodal segment	MS	1 mg.l ⁻¹ IBA and 0.1 mg.l ⁻¹ 6-BA	Meristem culture	Martin and Perez, 1992
<i>L. perigrinum</i>	leaf discs	MS	1-2 mg.l ⁻¹ thidiazuron	Organogenesis	Seelye et al., 1994
<i>L. dufourei</i> , <i>L. calaminare</i> , <i>L. gibertii</i> , <i>L. dichotomum</i> and <i>L. catalaunicum</i>	nodal segment	MS	1 mg.l ⁻¹ IBA and 0.1 mg.l ⁻¹ 6-BA	Meristem culture	Martin and Perez, 1995
<i>L. thiniense</i>	Shoot tip	MS	0.1 to 0.5 mg.l ⁻¹ 6-BA or 1-2 mg.l ⁻¹ kinetin	Meristem culture	Lledo et al., 1996
<i>L. altaica</i> x <i>L. caspium</i>	Shoot primordia	MS	0.1 mg.l ⁻¹ each of BA and NAA	Meristem culture	Matsumoto et al., 1997
<i>L. cavanillesii</i>	Inflorescence stem pieces	MS	2-5 mg.l ⁻¹ kinetin, 5 mg.l ⁻¹ DAP or 0.1 mg.l ⁻¹ 6-BA	Meristem culture	Amo-Marco and Ibanez, 1998
<i>P. zeylanica</i>	Leaf & stem	MS	1 mg.l ⁻¹ 6-BA and 0.25 mg.l ⁻¹ IAA	Organogenesis	Rout et al., 1999
<i>L. bellidifolium</i> and <i>L. sinuatum</i>	Cotyledon & Hypocotyl	MS	1 mg.l ⁻¹ 2,4-D and 0.1 mg.l ⁻¹ kinetin	Somatic embryogenesis	Aly and Rathinasabapathi (2000)

flowers (Harada, 1992). Advantages include their availability in a range of bright colors and their use for enhancing floral arrangements both as fresh-cut and dried flowers.

Currently, most *Limonium* that are supplied to the international cut flower market come from Kenya, Israel and Holland (Janson and Reid, 1999). Superior cultivars are micropropagated in Japan, Holland and other countries. Since *Limonium* species are suitable for cultivation in Florida both in open fields and greenhouses, there is yet under-explored potential for their cultivation in Florida for supplying to the cut flower market.

An important feature of this family is their high level of tolerance to salinity and drought. We therefore employ *Limonium latifolium*, a perennial static, to study certain metabolic adaptations to stress tolerance (Rathinasabapathi et al., 2000). Moreover, some members of this family were reported to contain medicinal compounds (e.g., Devi et al., 1999).

Tissue culture techniques have been applied only to a limited extent in the Plumbaginaceae. A literature review indicated that certain *Limonium* species were regenerated using tissue culture, mostly via meristem culture techniques or organogenesis (Table 1). Recently, we have developed a rapid and efficient protocol to induce somatic embryogenesis in *L. bellidifolium* and *L. sinuatum* (Aly and Rathinasabapathi, 2000). This article describes a modified protocol that successfully induced somatic embryogenesis in *L. latifolium*.

Material and Methods

Limonium latifolium seeds were purchased from Park Seed Co. (Greenwood, SC, U.S.A.). Methods to disinfect and germinate the seeds and inoculation of explants and media preparation were conducted as described by Aly and Rathinasabapathi (2000). Hypocotyl and cotyledon explants were cultured on Murashige and Skoog medium (MS, Murashige and Skoog, 1962) supplemented with 1 mg.l⁻¹ 2,4-D and 0.1 mg.l⁻¹ kinetin and 3% (wt./v) sucrose, pH 5.7 and solidified with 0.8% (wt./v) agar. For maturation, embryogenic calli were transferred to MS medium supplemented with 0 and 0.1 mg.l⁻¹ kinetin and 3% (wt./v) sucrose.

Results and Discussion

On the induction medium, both hypocotyl and cotyledon explants responded by producing embryogenic calli characterized by globular structures (Fig. 1). In three independent experiments, each with 30 cotyledon explants, 67%, 53% and 52% of the explants responded. Many of the embryogenic calli developed red coloration. Most cotyledon explants developed globular embryos directly without an intervening callus proliferation. Globular somatic embryos were transferred to the maturation medium after three to four weeks. Bi-polar somatic embryos developed to heart-shaped, torpedo and occasionally to cotyledonary stages.

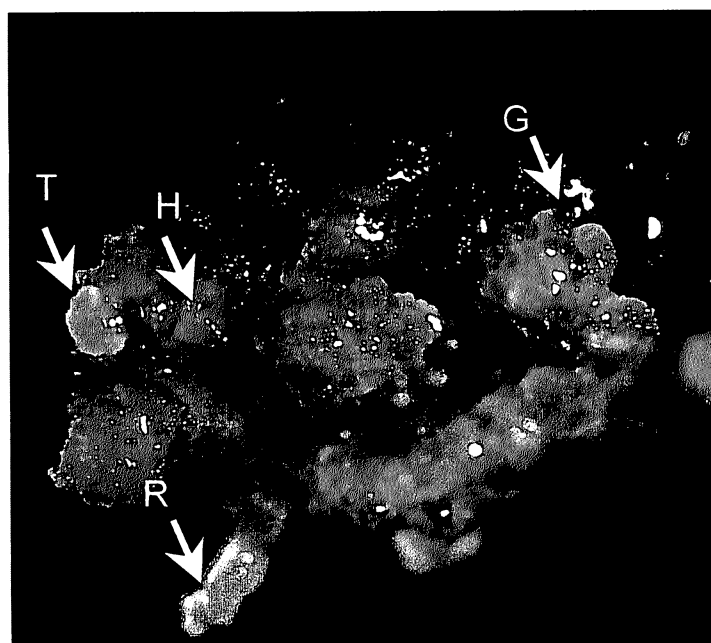


Figure 1. Embryogenic calli induced from a cotyledon explant of *L. latifolium*. Note the globular (G), heart-shaped (H) and torpedo (T) stage embryos and a root (R).

Plant regeneration via somatic embryogenesis is ideal for mass propagation of genetically identical and disease-free plants. It is superior to regeneration through organogenesis since it will avoid chimeric regenerants. Synthetic seed production is advantageous especially with *Limonium* species where seed viability is low or lost rapidly. Also, the method could be adapted to mutant selection and gene transfer techniques (Gray et al., 1995). Experiments are in progress to optimize the maturation and plant regeneration phases of somatic embryogenesis in *L. latifolium*.

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Proc. Fla. State Hort. Soc. 113:172-174. 2000.

STRATIFICATION ENHANCES GERMINATION OF PURPLE CONEFLOWER (*ECHINACEA ANGUSTIFOLIA*) AND ST. JOHN'S WORT (*HYPERICUM PERFORATUM*) SEEDS

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Additional index words. Light, medicinal plants, seed dormancy, seed germination.

Abstract. Usage of purple coneflower (*Echinacea angustifolia*) and St. John's wort (*Hypericum perforatum*) for medicinal, ornamental and landscape purposes has increased significantly in the United States. Seed germination and uniformity of emergence for such herbs were found unsatisfactory without pretreatments of seeds. Three 50-seed replicates of both *Echinacea angustifolia* and *Hypericum perforatum* were stratified in the dark at 5 and 10°C for 5, 10, 20 and 30 days. Germination counts on stratified and non-stratified seeds of both species were recorded on 3, 7 and 10 days in darkness and indirect daylight at 24°C. *Echinacea angustifolia* seeds had the highest percent germination (83%) when stratified at 10°C for 30 days and germinated under indirect daylight, whereas, ger-

mination was less than 50% under dark conditions. Highest germination of *Hypericum perforatum* seeds was attained with stratification at 10°C for 30 days. Stratification at 5°C for 20 days enhanced the rates of germination of both *Echinacea* and *Hypericum* seeds.

The use of purple coneflower and St. John's wort for medicinal purposes (Wijesekera, 1991), ornamental and landscape plantings (Cox and Klett, 1984) has increased significantly in the past few years. These herbaceous perennials are native to North America and constitute the top selling dietary supplements in the U.S. The raw material is used for numerous phytomedicines in Europe and many other countries (McKeown, 1999).

The extracts produced from the purple coneflower (*Echinacea angustifolia* var. *angustifolia*) stimulate the human immune system and are commonly used for treating and preventing the common colds and influenza. On the other hand, the oil and extract produced from St. John's wort (*Hypericum perforatum*) are used as an anti-depressive tonic, an anti-inflammatory and an analgesic to heal damaged tissues and nerves.

In general, the economic production of perennial crops requires high quality seeds with rapid synchronous germination. Propagation from seeds is considered the most efficient

Florida Agricultural Experiment Station Journal Series No. N-01918.
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