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Propagation of *Talinum cuneifolium* L. (Portulacaceae), an Ornamental Plant and Leafy Vegetable, by Stem Cuttings

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ABSTRACT

This paper reports on the propagation of *Talinum cuneifolium* through stem cuttings. It is popularly known as Ceylon spinach and is used as a leafy vegetable in the tropics. In the present study, stem cuttings of *T. cuneifolium* were successfully propagated in soil and hydroponic media. Propagated plants flowered extensively within a week under both conditions. Stem cuttings of plants in hydroponic media initiated adventitious roots within a week. The growth and development of vegetative propagules, flowers and adventitious roots were determined at different periods of the experiment. Our study indicated that *T. cuneifolium* can be propagated through stem cutting within 7-days of culturing. Furthermore, hydroponically grown shoots with adventitious roots can serve as an ideal experimental system for toxicity bioassays (rhizofiltration) in the field of environmental research.

Keywords: Ceylon spinach, hydroponic, rhizodegradation, rhizofiltration, soil multiplication, toxicity bioassay

INTRODUCTION

The commercial production of ornamental plants is of a great economic importance in worldwide agriculture. The ornamental industry has widely applied *in vitro* propagation for large-scale plant multiplication of elite superior varieties. The propagation through shoot cutting of herbaceous plants has wide applications in ornamental plant production and the interest in ornamental or foliage plant production is increasing rapidly as a result of their economic importance and their diversity. In soil, propagation through seed germination is a way to develop *Talinum* plants but the propagation rate is slow. It has been reported that peony (*Paeonia*) plants, producing offspring through seed germination, can maintain the superior characteristics of parent plants (Halda and Waddick 2004). The soil system may affect seedlings because of variable availability of nutrients and soil texture. It is reported that in *Talinum*, soil nutrients like phosphorus, potassium and nitrogen in different combinations and pH cause variations in stem weight, leaf width or leaf area

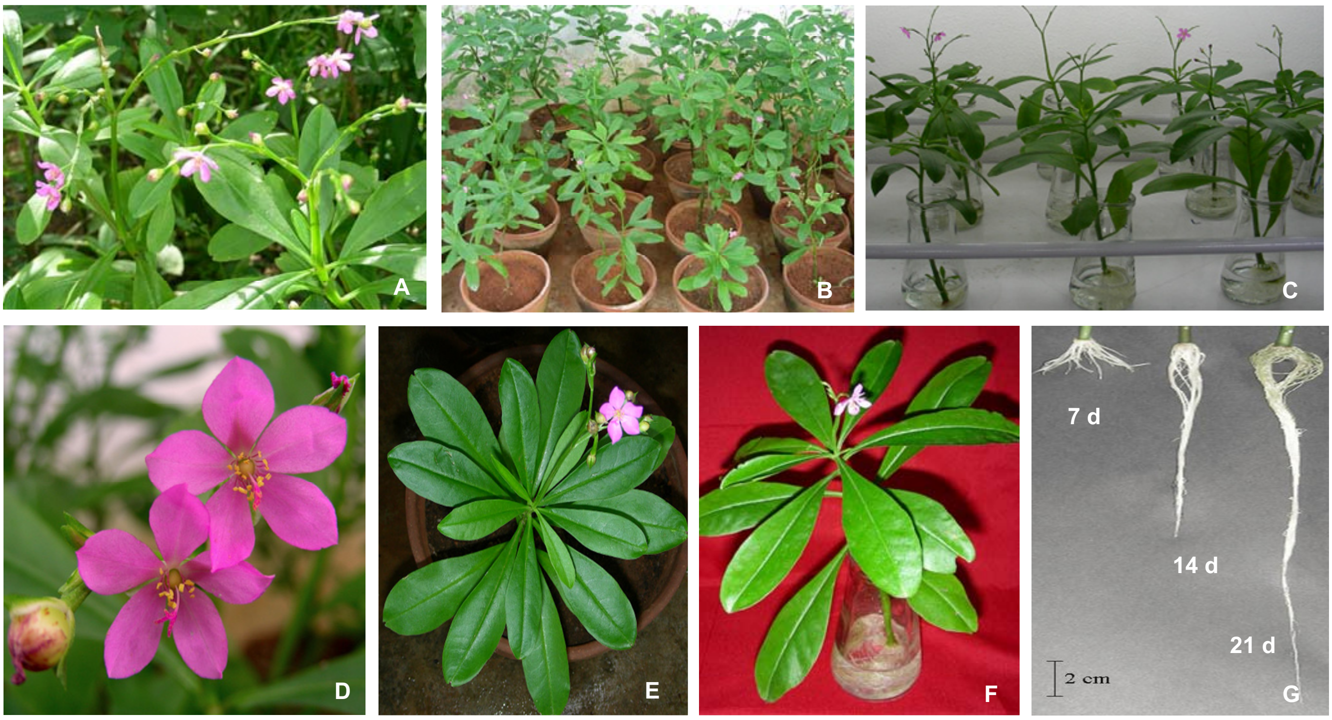

Fig. 1 Propagation of *T. cuneifolium* in field, greenhouse and hydroponic conditions. (**A**) Plants with widespread flowers and seeds in field. (**B**) Plantlets grown in the pots under greenhouse condition. (**C**) Plants in hydroponic media in laboratory conditions with adventitious roots, vegetative propagules and flowers. (**D**) Flowers of *T. cuneifolium* plant. (**E**) Plant in soil culture at 21-days. (**F**) In hydroponic media in laboratory conditions with adventitious roots and flower at 21-days. (**G**) Roots developed under hydroponic media at 7-, 14- and 21-days.

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Table 1 Geographical distribution and economic importance of different species of *Talinum*.

Species	Geographical distribution	Economic importance	References
T. calcaricum	America, Columbia, Georgia	Ornamental	Harris and Martin 1991
T. calycinum	America	Ornamental	Harris and Martin 1991
T. cuneifolium	Africa, America, Arabia, India	Ornamental, medicinal, vegetable	Lakshminarayana *et al.* 2007; Raju *et al.* 2007
T. mengesii	America, Georgia	Ornamental	Black and Murdy 1972
T. paniculatum	Trinidad,	Ornamental, medicinal	Foxcroft *et al.* 2008
T. parviflorum	America	Ornamental	Harris and Martin 1991
T. teretifolium	America, Georgia	Ornamental	Black and Murdy 1972
T. triangulare	Africa, America, Arabia, India, Japan	Ornamental, saponins, vegetable	Khoda *et al.* 1992; Fasuyi 2007

(Ukpong and Moses 2001). *In vitro* propagation, by means of stem cuttings, is a common tool for rapid expansion and multiplication of plants and also has been widely established for various ornamental plants. *In vitro* propagation is a key tool in plant biotechnology that exploits the totipotency of plant cells. The main advantage of *in vitro* (vegetative) propagation is to produce a large number of vegetative propagules, adventitious roots and leaves. It is a reliable way of multiplying plants in a short period of time (Jain and Ochatt 2010). The regeneration of adventitious roots in excised leafy plant shoots is a crucial phenomenon and rooting ability depends on many endogenous and exogenous factors such as species, type of cuttings, composition of nutrient medium, season of propagation, environment and propagation system (Németh 1986; Hartmann *et al.* 2002).

Members of the Portulacaceae family i.e. *Portulaca*, *Calandrinia*, *Lewisi* and *Talinum* are known to bear ornamental and attractive flowers (Hickey and King 1981; Foxcroft *et al*. 2008). *Talinum cuneifolium* (Ceylon spinach) is a terrestrial herbaceous plant with erect, simple or branching stems (**Fig. 1A**). It is a small genus with 50 species mostly found in the tropics, subtropics and warmer parts of the world (Hickey and King 1981; Watson and Dallwitz 1992). *T. cuneifolium* is relatively easy to grow and has attractive foliage under an ideal environment and stem cuttings of plants are a ready means of propagation of these plants in both soil and hydroponic media (**Fig. 1B**, **1C**). *T. cuneifolium* flowers and fruits throughout the year and flowers are small and showy, open in the morning and close in the late afternoon (**Fig. 1D**). Apart from its ornamental value, *Talinum* species are widely used as leafy vegetable plants in the tropics including India, Arabia, Africa and America (**Table 1**). In south-eastern States of Nigeria, *Talinum* is being cultivated and considered as a highly desirable vegetable plant (Ukpong and Moses 2001). Almost all its parts are useful and used as traditional medicine and leaves are a good source of antioxidant micronutrients (xanthophyll carotenoids) such as lutin (89.79 mg/100 g DW) and zeaxanthin (1.22 mg/100 g DW), in higher concentration, in the leaves (Khoda *et al.* 1992; Lakshminarayana *et al.* 2007). *T. cuneifolium* shows close resemblance with *T. triangulare* in habit, gross morphological characters and base chromosome numbers but differ in only pollen morphology. The former belonging to pollen type III and later to pollen type II (Nyanayo 1992).

The aim of this study was to propagate *T. cuneifolium* both in soil and hydroponic media. Further, we studied the influence of soil and hydroponic media on the growth rate of vegetative propagules and regeneration potential of *T. cuneifolium* plants. The study was performed to explore the stem cutting method in order to promote additional experimental applications.

MATERIALS AND METHODS

Plants

T. cuneifolium is a succulent perennial that inhabits soil communities along the perimeter of rock outcrops. Materials required for these experiments were collected from a field bank maintained as stock plant at the University of Hyderabad, India. Stem cuttings were excised from the branching portion by hand with a sharp razor blade. The average cutting diameter of each plant shoot was 30-50 mm and shoot height ranged from about 17-22 cm. The rooting test of each plant species involved three replication, each consisting of 8 cuttings. Immediately after excision from the stock plants, the cuttings were propagated in soil and hydroponic media without any wetting agent treatment.

Propagation in soil

Stem cuttings of *T. cuneifolium* plants were potted in 5-L cement pots filled with a mixture of red soil and sand (pH-6.8) in a greenhouse with an average daily minimum temperature of 23°C and a maximum of 31°C; the relative humidity ranged between 60-75%. Inside the greenhouse the photosynthetic photon flux density (PPFD) ranged from 900-1200 μmol m^{-2} s^{-1} with an average sunlight of 6- h day^{-1}. Potted *T. cuneifolium* plants were watered daily and examined for the formation of flowers and vegetative propagules.

Propagation in hydroponic medium

T. cuneifolium stem cutting were placed in a conical flask containing 100 ml of 10% modified Hoagland's solution (Hoagland and Arnon 1950) under a 16- h photoperiod at 28 ± 2°C in laboratory conditions. Plants were kept for 3-weeks for acclimatization and regeneration of adventitious roots. The pH of the solution was maintained at 4.8. Nutrient solution was replaced every 3-4 days to provide a fresh dose of nutrient elements and to avoid algal growth. Algal culture binds to the root outer surface and reduces the absorption of nutrient elements and inhibits root growth.

Rating of propagules and rooting in hydroponic medium

Adventitious root formation in *T. cuneifolium* stem cuttings was calculated after a rooting period of 7-, 14- and 21-days. Rooting of cuttings was not always successful and the percentage of non-surviving cuttings was calculated at 7-days. Cuttings that survived were used to analyze the mean number of vegetative propagules, mean root length and fresh weight (FW) at three different sampling periods (7-, 14- and 21-days) to evaluate the commercial quality of stem cuttings.

Statistical analysis

The experiments were repeated three times and the data presented corresponds to the mean values ± S.E. (standard errors) of three replicates (Microsoft Office 2003). Significant differences of these data were calculated using one-way analysis of variance (ANOVA) and Tukey's test (SIGMASTAT 9.0). P values < 0.05 were considered significantly different and denoted by different letters.

RESULTS AND DISCUSSION

Propagation in soil

Talinum is a succulent herb that grows in shallow soil habitats with rock exposures. Members of this genus are considered as rock outcrop species, many of which grow on granite, serpentine, sandstone, and limestone rocks (Black and Murdy 1972; Ware and Pinion 1990). During the expe-

Table 2 Analysis of number of flowers and vegetative propagules in propagated stem cutting in soil medium and root length, root fresh weight and vegetative propagules in hydroponic media after 7-, 14- and 21-days, respectively.

	Propagation period		
	7-days	**14-days**	**21-days**
In soil medium			
Flowers (per plant)	0.63 ± 0.15 a	1.45 ± 0.15 ab	2.04 ± 0.23 bc
Mean propagules (%)	50 ± 10 a	129 ± 12 ab	208 ± 15 bc
In hydroponic media			
Mean roots fresh weight (g)	0.155 ± 0.01 a	0.56 ± 0.02 b	0.93 ± 0.04 c
Mean root length (cm)	3.61 ± 0.23 a	10.85 ± 0.5 b	17.45 ± 0.5 c
Mean propagules (%)	68 ± 12 a	144 ± 13 ab	256 ± 23 c

* Mean values ± S.D. of three replicate (n = 8). The values at each time point not sharing a common letter are not significantly different ($P < 0.05$) between groups within a row as determined by analysis of variance (ANOVA) and Tukey's multiple range test.

riment, potted plants were observed daily to understand their ability to acclimatize in soil. In the soil system survival rate of the plant cuttings was 87% at 7-days. Potted cuttings were healthy and flowered within a week of culture (**Fig. 1E**). In the soil system, stem cuttings of 21- and 14-days showed a 3.2- and 2.3-fold increase, respectively in the number of flowers more than 7-days stem cuttings. Similarly, stem cuttings propagated for 21-days showed a 1.4-fold increases in the number of flowers more than stem cuttings propagated for 14-days (**Table 2**). *Talinum* is predominantly self-pollinated or vegetatively propagated. Thus, for 21-days the vegetative propagules showed a 4.2- and 1.6-fold increase more than 7- and 14-days, respectively. Furthermore, 14-days cuttings showed a 2.6-fold increase in the number of propagules more than 7-days cuttings (**Table 2**). Results of our observation confirmed that *T. cuneifolium* plants are mostly multiplied through vegetative propagation and produced numerous vegetative propagules on their senescing stem tips. The number of flowers and vegetative propagules were increased and directly dependent on propagation periods (**Table 2**). Shoot tip explants are routinely used for the propagation of ornamental plants, including *Ardisia japonica* (Roh *et al.* 2005), *Begonia tuberosus*, *Ranunculus asiaticus* L., *Dianthus caryophylus* L., *Jasminum officinale* L (Jain and Ochatt 2010), *Ebenus cretica* (Hatzilazarou *et al.* 2001), *Pelargonium x hortorum* (Druege *et al.* 2007), *Rosa hybrida* L. (Bredmose and Hansen 1995) and *Zantedeschia albomaculata* (Chang *et al.* 2003). The sustainable growth of *T. cuneifolium* makes continuous demands on soil properties and it is necessary to restore the nutrients and increase the sustainability of soil by the application of nutrients and organic manure. Soil multiplication is a powerful tool for large-scale propagation of ornamental plants and further, having application for phytoremediation. Phytoremediation is a cost-effective, nonintrusive technique and an emerging green technology that uses the ability of certain plant species to remove toxic metals from the soil. It has been reported that *Portulaca oleracea*, another member of the Portulacaceae family, can survive for a longer periods in spite accumulating more toxic metals than reported hyperaccumulators such as *Heli-anthus annus* and *Brassica juncea* in the presence of an electroplating effluent (Jenita *et al.* 2010).

Propagation in hydroponic medium

In laboratory conditions, *Talinum* plants can be maintained effortlessly in hydroponic media. Plants were observed with adventitious roots and flowers in hydroponic media (**Fig. 1F**). Results of our study showed that the propagation of stem cuttings in hydroponic media and laboratory conditions generally produced a high survival rate of 94% at 7-days. The higher percentage of survival of cuttings showed that plants grew easily in hydroponic media. Plants grown well in the nutrient solution and root primordia at the base of stem cuttings were observed within one week of culture. Root parameters such as length and FW of adventitious roots were enhanced and increased exponentially over time (**Table 2**). In hydroponic media, stem cuttings of 21- and 14-days showed a 6.2- and 3.0-fold increase, respectively in the root length more than 7-days stem cuttings. Similarly, stem cuttings propagated for 21-days showed a 1.6-fold increase in the root length more than stem cuttings propagated for 14-days (**Fig. 1G**). Furthermore, 21-days root FW showed a 1.6- and 6.2-fold increase more than 14- and 7-days stem cuttings, respectively. Similarly, stem cuttings of 14- days showed a 3.7-fold increased in the root FW more than stem cuttings of 7-days (**Table 2**). Almost no variation in the root diameter was observed. After 7-days, the presence of longer roots and higher root FW of regenerated stem cuttings indicated the rapid development of adventitious roots. In terms of vegetative propagules, 21-days stem cutting showed a 3.8- and 1.8-fold increase more than 7- and 14-days stem cuttings, respectively (**Table 2**). The above parameters (root length, root FW and vegetative propagules) of stem cuttings showed minimum values after 7-days indicating that the initial week is required for acclimatization of plants stem cuttings in hydroponic media. Our results suggested that the formation of any vegetative propagules and adventitious roots in *T. cuneifolium* cuttings was promoted by hydroponic media and growth conditions.

The growth of *T. cuneifolium* is directly dependent on the growth of roots in hydroponic media because the growth and survival of terrestrial plants are related to the potential ability of roots to absorb water and nutrient elements from the soil or growth media (Eapen *et al.* 2005). The advantages of water culture are that plant roots are suspended only in water medium and avoid soil, containing salts and soil-borne microorganisms. They may interrupt nutrient uptake and plant growth by precipitating salts or affecting the pH of the solution. On the other hand, hydroponic culture provides only liquid nutrient medium to plants and a generous availability of nutrients promote rapid plant growth.

Hydroponically cultivated adventitious roots of several terrestrial plants were used for rhizofiltration, a process used for absorbing or precipitating toxic metals effectively from a polluted site (Dushenkov *et al.* 1995). Adventitious roots of *Talinum* plants have the potential to absorb heavy metals and might be useful for a new rhizofiltration system. *Talinum* plants could absorb heavy metals and were used for absorption experiments of heavy metals i.e. Pb, Cd, Ni, Cu (Rajkumar *et al.* 2009). Absorption of heavy metal content induces some ultrastructure modifications in root tissues concerning the increase in cell wall thickness (Probst *et al.* 2009). Heavy metal accumulation in plant tissues causes the synthesis of small cysteine-rich peptides, phytochelatins (PCs) or metallothioneins (MTs), which bind to the metals and form peptide-metal complexes and detoxify them. These properties develop a characteristic to absorb more heavy metals in the plant. Probst *et al.* also reported that roots of *Vicia faba* absorbed more metals than stems and leaves. The order of metal accumulation was root > leaf > stem for all heavy metals except for Pb and Cd where the order was root > stem > leaf (Probst *et al.* 2009). The methods of propagation in hydroponic media have a significant influence on the production of adventitious roots and new leaves. Very rapid development of adventitious roots in *Talinum* plants indicates that a one-week period required for the establishment of a potential plant propagation system for rizhofiltration. Furthermore, with the help of adventitious roots we can study the effect of foliar retention, absorption of heavy metals and also various aspects of abiotic stresses in root tissues.

Talinum cuneifolium can be grown in soil and hydroponic media. It can be used as an indoor and outdoor ornamental plant. Thus, *T. cuneifolium* with high vegetative propagation potential would serve as an ideal experimental system for different fields of plant science research.

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REFERENCES

Black CB, Murdy WH (1972) The evolutionary origin of *Talinum teretifolium* Pursh. *Botanical Gazette* **133 (4)**, 405-410

Bredmose N, Hansen J (1995) Regeneration, growth and flowering of cut rose cultivars as affected by propagation material and method. *Scientia Horticulturae* **64**, 103-111

Chang HS, Chakrabarty D, Hahn EJ, Paek KY (2003) Micropropagation of calla lily (*Zantedeschia albomaculata*) via *in vitro* shoot tip proliferation. *In Vitro Cellular and Devlopmental Biology – Plant* **39**, 129-34

Druege U, Baltruschat H, Franken P (2007) *Piriformospora indica* promotes adventitious root formation in cuttings. *Scientia Horticulturae* **112**, 422-426

Dushenkov V, Kumar PBAN, Motto H, Raskin I (1995) Rhizofiltration: the use of plants to remove heavy metals from aqueous streams. *Environmental Science and Technology* **29**, 1239-1245

Eapen D, Barroso ML, Ponce G, Campos ME, Cassab GI (2005) Hydrotropism: root growth responses to water. *Trends in Plant Science* **10 (1)**, 44-50

Fasuyi OA (2007) Bio-nutritional evaluations of three tropical leaf vegetables (*Telfairia occidentalis*, *Amaranthus cruentus* and *Talinum triangulare*) as sole dietary protein sources in rat assay. *Food Chemistry* **103**, 757-765

Foxcraft CL, Richardson DM, Wilson JRU (2008) Ornamental plants as invasive aliens: Problems and solutions in Kruger National Park, South Africa. *Environmental Management* **41**, 32-51

Halda JJ, Waddick JW (2004) *The Genus Paeonia*, Timber Press, Portland, 187 pp

Harris FS, Martin CE (1991) Plasticity in the degree of CAM cycling and its relationship to drought stress in five species of *Talinum* (Portulacaceae). *Oecologia* **86**, 575- 584

Hartmann HT, Kester DE, Davies FT, Geneve RL (2002) *Plant Propagation: Principles and Practices* (7th Edn), Prentice Hall, New Jersey, pp 363-365

Hatzilazarou S, Economou A, Antoniou T, Ralli P (2001) Propagation of *Ebenus cretica* L. by tissue culture. *Propagation of Ornamental Plants* **1**, 25-27

Hickey M, King C (1981) *100 Families of Flowering Plants*, Cambridge University Press, London, 76 pp

Hoagland DR, Arnon DI (1950) The water-culture method for growing plants without soil. *California Agricultural Experiment Station Circular* **347**, 1-32

Jain SM, Ochatt SJ (2010) *Protocols for In Vitro Propagation of Ornamental Plants. Methods in Molecular Biology* (Vol 589), Humana Press, U.S., 324 pp

Jenita KZ, Kalaivani P, Sathyavathi S, Doble M (2010) *Portulaca oleracea*, a potential candidate for contaminated soil? In: *Proceedings of the 7th International Phytotechnologies conference*, 26-29 September 2010, Parma, Italy, Servizio Editoria e Fotoriproduzione- Settore Economato e Provveditorato, University of Parma, Italy, 188 pp

Kohda H, Yamaoka Y, Morinaga S, Ishak M, Darise M (1992) Saponins from *Talinum triangulare*. *Chemical and Pharmaceutical Bulletin* **40 (9)**, 2557-2558

Lakshminarayana R, Raju M, Krishnakantha TP, Baskaran V (2007) Lutein and zeaxanthin in leafy greens and their bioavailability: Olive oil influences the absorption of dietary lutein and its accumulation in adult rats. *Journal of Agricultural and Food Chemistry* **55**, 6395-6400

Németh G (1986) Induction of rooting. In: Bajaj YPS (Ed) *Biotechnology in Agriculture and Forestry 1. Trees 1*, Springer-Verlag, Berlin, pp 49-64

Nyananyo BL (1992) Pollen morphology in the Portulacaceae (Centrospemu). *Folia Geobotanica et Phytotaxonomica* **27**, 387-400

Probst A, Liu H, Fanjul M, Liao B, Hollande E (2009) Response of *Vicia faba* L. To metal toxicity on mine tailing substrate: Geochemical and morphological changes in leaf and root. *Environmental and Experimental Botany* **66**, 297-308

Rajkumar K, Sivakumar S, Senthilkumar P, Prabha D, Subbhuraam CV, Song YC (2009) Effects of selected heavy metals (Pb, Cu, Ni and Cd) in the aquatic medium on the restoration potential and accumulation in the stem cuttings of the terrestrial plant, *Talinum triangulare* Linn. *Ecotoxicology* **18**, 952-960

Raju M, Varakumar S, Lakshminarayana R, Krishnakantha TP, Baskaran V (2007) Carotenoid composition and vitamin A activity of medicinally important green leafy vegetables. *Food Chemistry* **101**, 1598-1605

Roh MS, Lee AK, Suh JK (2005) Production of high quality *Ardisia* plants by stem tip cuttings. *Scientia Horticulturae* **104**, 293-303

Ukpong EI, Moses JO (2001) Nutrient requirements for the growth of waterleaf (*Talinum triangulare*) in Uyo metropolis, Nigeria. *The Environmentalist* **21**, 153-159

Ware S, Pinion G (1990) Substrate adaptation in rock outcrop plants: Eastern United States *Talinum* (Portulacaceae). *Bulletin of the Torrey Botanical Club* **117 (3)**, 284-290