Research Note

ZANTEDESCHIA AETHIOPICA *PROPAGATION BY TISSUE CULTURE*[1]

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Zantedeschia aethiopica (L.) K. Spreng (white calla lily) and *Zantedeschia* spp, of the Araceae family, are popular cut flowers (Tjia, 1987). They are produced and marketed as flowering potted plants bearing attractive spathes, commonly referred to as flowers (Corr and Widmer, 1991). Although today's average sale price per cut calla lily at the farm gate in Puerto Rico is $2.00, few ornamental growers produce it. Growers in Puerto Rico produced 3.2 ha of cut flowers whereas estimated cut flower imports amounted to $1.8 million in 1993-94 (P. R. Dept. Agric., Agric. Statistics, personal communication). Puerto Rico has the potential, with its tropical climate and skilled labor, to produce cut flowers locally. One crop that can reduce our cut flower imports is calla lily.

Soft rot, caused by the bacteria *Erwinia* spp. (Cohen, 1981; Cortés-Monllor, 1990), adversely affects calla lily production in Puerto Rico. This bacteria causes particularly severe losses during vegetative propagation. At the Adjuntas Substation (subhumid, temperature range from 22.2 to 24.4°C, average annual precipitation 189.23 cm), over 90% of the plants propagated by rhizome died because of this disease (Dr. Mildred Zapata, phytopathologist, personal communication). To reduce bacteria disease, calla lily plants can be propagated by tissue culture (Hartman, 1974; Knauss, 1976). The objective of this study was to modify tissue culture methods to obtain an appropriate technique for the production of calla lily plants.

At the Plant Tissue Culture Laboratory of the Río Piedras Research Center, two disinfection methods were tested. In method 1 (standard method used in our tissue culture laboratory), five calla lily rhizomes were washed thoroughly and roots and leaves were removed with a knife. Excised and dissected rhizome buds were surface-disinfected with sodium hypochlorite (10%) and two drops of Tween 80[5]/100 ml for 10 min, followed by two to three rinses with sterile distilled water containing 120 mg/L of citric acid. Treated buds were then rinsed three times with sterile distilled water. Excess water from the buds was absorbed with filter paper in sterile petri dishes. In method 2, five dissected rhizomes were dipped in 95% ethanol for 1 min and flamed twice (Cohen, 1981). This surface disinfection procedure was repeated twice during rhizome bud dissection, a modification of Cohen's method.

On 15 June 1993, all buds were transferred to Murashige and Skoog medium (1962) supplemented with inositol (100 mg/L), thiamin.HCL (0.4 mg/L), sucrose (30 g/L), benzy-

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ladenine (BA) (3 mg/L) and agar (6 g/L) (standard medium for calla lily micropropagation, Cohen, 1981). The pH of the medium was adjusted to 5.8. Tissue cultures were grown in 28-ml glass tubes or 153-ml glass jars at 26-28°C under 16 h of fluorescent cool white light (1722 lumen · m^2). Propagules were transferred to fresh medium every three weeks.

Treatment of rhizomes by disinfection method 1 was not effective. Eighty percent of the rhizome buds did not grow because of bacteria contamination, whereas 80% of those treated by method 2 grew effectively in the medium. But after six weeks in the medium, all propagule shoots developed a chlorotic pigmentation. The standard medium (Cohen, 1981) was modified by using half strength concentration of major and minor salts and BA at 0.3 mg/L. Propagule shoots turned green after two weeks in the modified medium. Multiplication of the propagules in the medium was slow, so BA was increased to 2 mg/L. The increase in BA improved multiplication rate, but an even further increase in BA (3 mg/L) was tested to maximize calla bud multiplication. This last modified medium was effective for calla lily rhizome bud initiation and multiplication.

Reducing the amount of BA in the medium to 0.1 mg/L induced root formation on calla propagules after three weeks. Calla lily plantlets were removed from the jars and their roots were washed thoroughly to remove the medium and planted in flats containing commercial peat-based medium (Sunshine mix #4)[5]. The peat based medium was drenched with Terraclor 75% WP[5] (0.95 g/L) and Subdue 2E[5] (0.052 cc/L) fungicides after planting. Plants were transplanted successfully in 15-cm pots filled with peat-based medium in a greenhouse at Gurabo Substation. They were also transplanted to concrete boxes (1.22 m × 3.05 m) filled with 1:1:1 mixture of top soil, sugarcane bagasse and sand under 50% shade at Adjuntas Substation. After three months, plants have not shown any visual symptoms of soft rot disease in Adjuntas or Gurabo. Results of this preliminary study show that the modified disinfection method and medium can increase the success of producing calla lily plants by tissue culture.

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