ference (2). Lily bulbs are marketed by circumference, and such an increase, if realized, would be of appreciable economic importance. The occasional failure of bulb size to increase in reponse to hand deflowering (8) or application of the flower aborting agent CIPC (isopropyl-*m*-chlorocarbanilate) (17) suggests that other factors, such as alternate sinks or hormonal regulation may affect bulb growth after removal of flowers.

Literature Cited

1. Biale, J.B. 1975. Fruit ripening and senescence of flowers and leaves. Physiol. Veg. 13:701–708.
2. Blaney, L.T. and A.N. Roberts. 1959. Grading methods compared for lily bulbs. Oregon Orn. Nursery Dig. 3:3–4.
3. Blaney, L.T. and A.N. Roberts. 1965. Flower count compared to total bulb weight. Oregon Orn. Nursery Dig. 9:1–2.
4. Cockran, F.D. and T.L. Vereen. 1939. Some factors affecting flowering and bulb production in the Creole Easter lily. Proc. Amer. Soc. Hort. Sci. 37:1031–1033.
5. Coorts, D.C., J.B. Gartner, and J.P. McCollum. 1965. Effect of senescence and preservative on respiration in cut flowers of *Rosa hybrida*, 'Velvet Times'. Proc. Amer. Soc. Hort. Sci. 86:779–790.
6. Gould, F.G. 1953. Blights of lilies and tulips, pp. 611–617. In: Plant diseases, the Yearb. of Agr. USDA, Washington, D.C.
7. Kuc, R. and M. Workman. 1964. The relation of maturity to the respiration and keeping quality of cut carnation and chrysanthemum. Proc. Amer. Soc. Hort. Sci. 84:575–581.
8. Masaaki, K. 1974. Studies on the flower bud abortion in Easter lily (*Lilium longiflorum* Thunb.) after application of surfactants. Bul. Veg. Orn. Crops Res. Sta. Japan, Ser. C, 1:83–90.
9. Nakamura, R.R., R. Ito, and K. Hirano. 1975. Changes in the respiration rate of cut flowers. Sci. Rep. Faculty Agr., Okayama Univ., Japan 46:29–37.
10. Nichols, R. 1973. Senescence of the cut carnation flower: respiration and sugar status. Proc. Amer. Soc. Hort. Sci. 48:111–112.
11. Nichols, R. and L.C. Ho. 1979. Respiration, carbon balance and translocation of dry matter in the corolla of rose flowers. Ann. Bot. 43:19–25.
12. Niimi, Y. and H. Torikata. 1979. Changes in cytokinin activities, photosynthesis and respiration of the grape flower clusters during their development. J. Jap. Soc. Hort. Sci. 47:301–307.
13. Siegleman, H.W. 1952. The respiration of rose and gardenia flowers. Proc. Amer. Soc. Hort. Sci. 59:496–500.
14. Siegleman, H.W., C.T. Chow, and J.B. Biale. 1958. Respiration of developing rose petals. Plant Physiol. 33:403–409.
15. Stimart, D.P., D.J. Brown, and T. Solomos. 1983. Development of flowers and changes in carbon dioxide, ethylene, and various sugars of cut *Zinnia elegans* Jacq. J. Amer. Soc. Hor. Sci. 108:651–655.
16. Wang, Y-T. and P.J. Breen. 1983. Flower removal alters the distribution of current photosynthates in Easter lily, *Lilium longiflorum* Thunb. HortScience 18(4):609 (Abstr.)
17. William, R.D., L. Riddle, and W. Schroeder. 1982. Disbudding lilies using different formulations of CIPC. Hort. Weed Control Rpt., 1982. Dept. of Hort., Oregon State Univ., Corvallis.

HortScience 19(5): 703–705. 1984.

In Vitro Propagation of African Marigold

S.L. Kothari and N. Chandra

Department of Botany, University of Rajasthan, Jaipur 302 004 India

Additional index words. African marigold, adventitious shoot buds, plant regeneration, negatively geotropic green roots

Abstract. **Leaf segments of *Tagetes erecta* L. (African marigold) were cultured on Murashige and Skoog (MS) basal medium supplemented with 6-benzyl-aminopurine (BA), kinetin, 3-indoleacetic acid (IAA), 3-indolebutyric acid (IBA), and naphthaleneacetic acid (NAA). Regeneration of a large number of adventitious shoot buds was observed on a medium with BA (3 to 5 mg/liter) + IAA (1 mg/liter). When shoot buds plus some callus were subcultured on the same medium, additional buds differentiated for up to 2 years. The shoot buds elongated when subcultured on a medium with BA (2 mg/liter) + 0.5 mg/liter gibberellic acid (GA_3). Shoots with sufficiently developed internodes were rooted on filter paper bridges in culture tubes on MS liquid medium with IBA (0.5 mg/liter) + GA_3 (0.5 mg/liter) to obtain complete plantlets with well-developed root and shoot systems. Abnormal negatively geotropic green roots formed on a medium with kinetin (1 to 4 mg/liter) + IBA (1 to 4 mg/liter).**

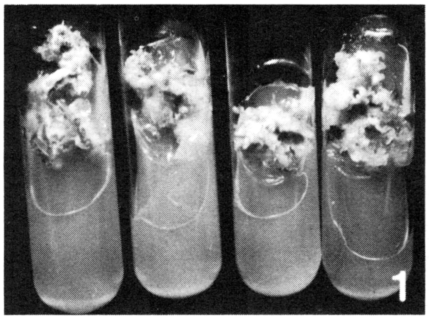

Fig. 1. Formation of shoot buds from callus from leaf explants on MS + BA (5 mg/liter) + IAA (1 mg/liter) after 20 days of culture.

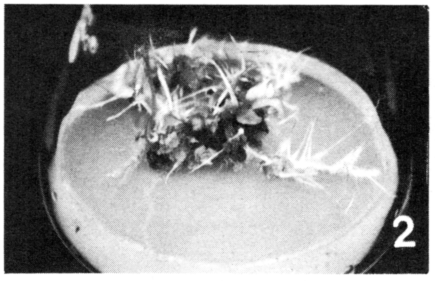

Fig. 2. Shoot and root formation from leaf segments on MS + BA (3 mg/liter) + IAA (5 mg/liter).

Plant tissue and organ culture as a technique for the mass propagation of selected plants is now established for many plant species (1, 5, 6, 7, 9), but only a few species in the Asteraceae family have been propagated using tissue culture (2, 3, 10, 12). *In vitro* multiplication of ornamental plants through tissue culture is advantageous since the plants produced may be genetically identical and disease free (6). The present report describes regeneration of plants of *Tagetes erecta*, an important ornamental plant, through leaf culture. Organogenesis in *Tagetes* from hypocotyl and cotyledons has been reported (8).

Leaves taken from plants growing in the botanic garden of the Univ. of Rajasthan, Jaipur campus, India, were surface sterilized for 5 min in a 0.1% $HgCl_2$ solution and washed 3 times in sterile distilled water. Leaf segments (1.5 × 0.6 cm) were cultured aseptically on 0.8% agar (Difco Bacto) solidified MS basal medium (11) supplemented with various concentrations of auxins and cytokinins. The pH of the medium was adjusted to 5.8 before autoclaving at 1.06 kg/cm^2. All cultures were incubated at 28°C under 24 hr illumination from cool-white fluorescent tubes and incandescent bulbs (1000 lx). Wide neck 100 ml Erlenmeyer flasks and test tubes (25 × 150 mm and 25 × 200 mm) were used as culture vessels. A 40 ml medium was dispensed in each flask and 20 ml in a tube. Throughout this paper, basal medium is denoted as MS followed by hormonal levels in mg/liter in brackets. All experiments were repeated 3 times.

The responses of leaf segments to different concentrations and combinations of BA, kinetin, IAA, IBA, and NAA are given in Table 1. The formation of complete plantlets

Received for publication 5 Mar. 1984. The authors are thankful to J.M. Widholm, Dept. of Agronomy, Univ. of Illinois, Urbana IL 61801, for his helpful suggestions and critically reading the manuscript. The cost of publishing this paper was defrayed in part by the payment of page charges. Under postal regulations, this paper therefore must be hereby marked *advertisement* solely to indicate this fact.

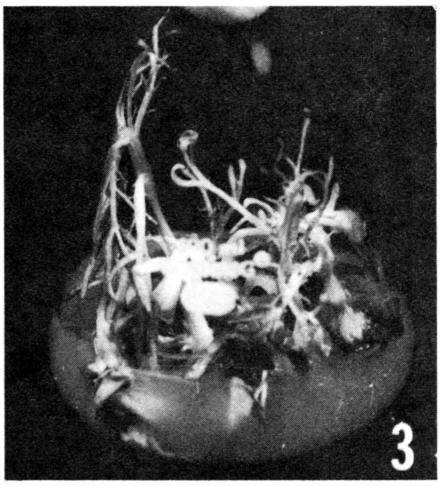

Fig. 3. Shoot buds together with some callus subcultured on MS + BA (2 mg/liter) + GA_3 (0.5 mg/liter), showing increase in size and number.

Table 1. Morphogenetic responses of leaf segments of *Tagetes erecta* with different concentrations of cytokinins and auxins on MS medium.

MS ± (mg/liter)	Morphogenetic response[z]	Cultures responding (%)
BA (1.0) + IAA (1.0)	R −	20
(3.0)	R −	40
(5.0)	R −	80
BA (3.0) + IAA (1.0)	S + +	100
(3.0)	S +	100
(5.0)	S + R −	80
	R −	20
BA (5.0) + IAA (1.0)	S + +	100
(3.0)	S −	100
(5.0)	S −	40
Kinetin (1.0) + IAA (1.0 to 3.0)	R +	100
Kinetin (3.0) + IAA (1.0 to 3.0)	R + NGR −	100
Kinetin (5.0) + IAA (1.0 to 3.0)	R −	100
Kinetin (1.0 to 4.0) + IBA (1.0 to 4.0)	R − NGR + +	100

[z]NGR, negatively geotropic roots; R, roots, S, shoot buds −, poor development; +, good development; + +, excellent development.

from cultured leaf segments was accomplished in 4 stages:

1. Induction of shoot buds. Leaf segments cultured on media containing BA (3 to 5 mg/liter) + IAA (1 to 5 mg/liter) showed prolific adventitious shoot bud differentiation via callus formation. Leaf segments swelled during the initial 6 days of culture, and callus formation started from the entire leaf, but particularly from the cut ends and the margins. Shoot buds appeared on the 12th day and continued to form for 3 weeks. Within a culture period of 30 days, the entire callus was covered with shoot buds (Fig. 1). High cytokinin/auxin ratio for the induction of shoot buds was first suggested by Skoog and Miller (13). Some roots also were formed on a medium with BA (3 mg/liter) + IAA (5 mg/liter) after the formation of shoot buds in the same culture flask (Fig. 2). On this medium, 2 of the 10 culture flasks showed early differentiation of roots instead of shoot buds. Shoot formation may have been prevented by these early differentiated roots.

Kinetin (1 to 5 mg/liter) in combination with the auxins (IAA, IBA, or NAA at 1 to 5 mg/liter) failed to induce any shoot bud differentiation. Rhizogenesis, however, occurred from the surface of leaf segments on media with kinetin (1 mg/liter) + IAA (1 to 5 mg/liter). Root formation was suppressed when the kinetin concentration was increased to 5 mg/liter and the IAA concentration lowered to 1 mg/liter. Abnormal development of negatively geotropic green roots from the entire surface of the leaf was observed on media with kinetin (1 to 4 mg/liter) + IBA (1 to 4 mg/liter) after 10 days of incubation, and in 30 days the leaf was covered with green roots growing apogeotropically.

2. Multiplication of shoot buds. Shoot buds differentiated in stage 1 were subcultured every 14 days on the same medium in which differentiation occurred (BA, 3 mg/liter + IAA, 1 mg/liter) as long as regeneration from callus continued. This proliferation of adventitious buds continued for more than 2 years. On this medium, shoot buds remained small, i.e., internodal length did not increase.

3. Elongation of shoot buds. Shoot buds from stage 2 subcultured on a medium with BA (2 mg/liter) grew in diameter size but did not elongate, but in the same medium with GA_3 (0.5 mg/liter) added, shoot length increased markedly. A well developed shoot system with elongated internodes was formed in 14 days (Fig. 3). Shoot elongation in the presence of GA_3 also has been reported in tobacco (4) and *Chrysanthemum* (2, 3) callus cultures. However, neoformation of buds has been reported to be inhibited by GA_3 (14). The elongation of shoots was essential for obtaining complete plantlets, since shoots without sufficiently developed internodes produced callus upon transfer to the rooting medium.

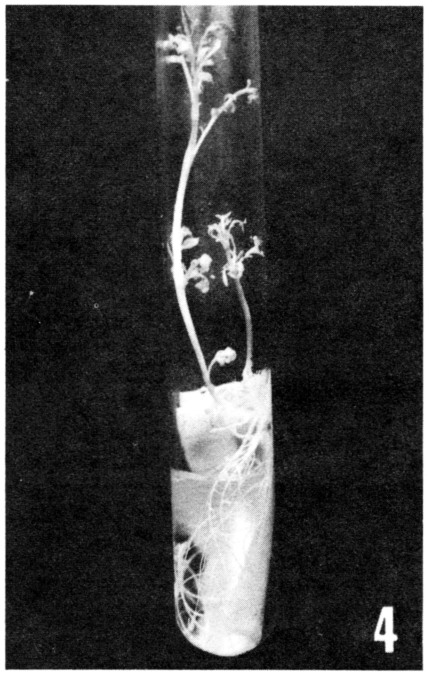

Fig. 4. Rooting of *in vitro* produced shoots on MS + GA_3 (0.5 mg/liter) + IBA (0.5 mg/liter).

4. Rooting of the shoots. Shoots from stage 3 were cut from the base and placed on filter paper bridges in culture tubes with a liquid medium for rooting. On a medium with IBA (0.5 to 3 mg/liter) alone, roots developed, but callus also was formed from the base of the stem. GA_3 alone (0.5 mg/liter) in the medium enhanced root formation, but the root system that developed was not prolific. When GA_3 (0.5 mg/liter) and IBA (0.5 mg/liter) both were added in the medium, roots were formed from the base of the shoots without any callus formation within 7 days in all the culture tubes. In 18 days, normal looking plantlets with well-developed root and shoot systems were obtained (Fig. 4). These plantlets then were transferred to a sugar-free medium in culture tubes on filter paper bridges with the same hormonal level and subsequently transferred to soil where 20% of them survived.

Literature Cited

1. Conger, B.V. 1981. Cloning agricultural plants via *in vitro* techniques, CRC Press, Boca Raton. Fla.
2. Earle, E.D. and R.W. Langhans. 1974. Propagation of *Chrysanthemum in vitro*. 1. Multiple plantlets from shoot tips and the establishment of tissue cultures. J. Amer. Soc. Hort. Sci. 99(2):128–132.
3. Earle, E.D. and R.W. Langhans. 1974. Propagation of *Chrysanthemum in vitro*. 2. Production growth and flowering of plantlets from tissue cultures. J. Amer. Soc. Hort. Sci. 99(4):352–358.
4. Engelke, A.L., H.Q. Hamzi, and F. Skoog. 1973. Cytokinin-gibberellin regulation of shoot development and leaf form in tobacco plantlets. Amer J. Bot. 60:491–495.
5. Holdgate, D.P. 1977. Propagation of ornamentals by tissue cultures, p. 18–43. In: J. Reinert and Y.P.S. Bajaj (eds.). Applied and fundamental aspects of plant cell tissue and organ culture. Springer-Verlag, Berlin.
6. Hughes, K.W. 1981. Ornamental species, p. 5–50. In: B.V. Conger (ed.). Cloning agricultural plants via *in vitro* techniques. CRC Press, Boca Raton, Fla.
7. Hussey, G. 1978. The application of tissue

culture to the vegatative propagation of plants. Sci. Prog. 65:185–208.

8. Kothari S.L. and N. Chandra. 1982. Induction of negatively geotropic roots in cultures of *Tagetes patula* L. Curr. Sci. 51:238–239.
9. Murashige, T. 1974. Plant propagation through tissue cultures. Annu. Rev. Plant Physiol. 25:135–165.
10. Murashige T., M. Serpa, and J.B. Jones. 1974. Clonal multiplication of *Gerbera* through tissue culture. HortScience 9(3):175–180.
11. Murashige, T. and F. Skoog. 1962. A revised medium for rapid growth and bioassays with tobacco tissue cultures. Physiol. Plant. 15:473–497.
12. Roest, S. and G.S. Bokelmann. 1973. Vegetative propagation of *Chrysanthemum cinerariaefolium in vitro*. Sci. Hort. 1:120–122.
13. Skoog, F. and C.O. Miller. 1957. Chemical regulation of growth and organ formation in plant tissue cultured *in vitro*. Symp. Soc. Expt. Bio. 11:118–131.
14. Thorpe, T.A. and D.D. Meier. 1973. Effects of gibberellic acid and abscisic acid on shoot formation in tobacco callus cultures. Physiol. Plant. 29:121–124.

HortScience 19(5): 705–707. 1984.

The Influence of Chlormequat and Daminozide on Net Photosynthesis, Transpiration, and Photorespiration of Hybrid Geranium

A.M. Armitage, Z.P. Tu[1], and H.M. Vines
Department of Horticulture, University of Georgia, Athens, GA 30602

Additional index words. *Pelargonium* ×*hortorum*, gas exchange, cycocel, B-nine, growth regulators

Abstract. **Foliar application of 0.3% CCC applied to geranium (*Pelargonium* ×*hortorum* Bailey) resulted in increased net photosynthesis (Pn), transpiration (Tr), chlorophyll concentration, and reduced photorespiration as measured by post lower illumination CO_2 burst (PLIB) compared with plants treated with 0.5% SADH. Pn and Tr rates were enhanced beginning 2 to 3 days after the CCC treatment compared with control plants and remained elevated for a least 4 to 5 more days.**

Growth regulators are used for the control of height in the production of hybrid geranium. Three of the major growth regulators used commercially in floriculture are succinic acid, 2-2 dimethylhydrozide (SADH, B-nine, alar, daminozide), 2 chloroethyl trimethylammonium chloride (CCC, chlormequat, cycocel), and α cyclopropyl-α-(4-methyoxyphenyl)-5-pyrimidine-methanol (Arest, ancymidol). Ancymidol and CCC restricted internode elongation and overall height in many cultivars of hybrid geranium (1, 10, 13); however, CCC is less costly than ancymidol and is normally recommended for height control (4). One of the responses of hybrid geranium to foliar application of CCC is accelerated flowering (1, 8, 10) compared with control plants. Miranda (10) found that flowering was accelerated regardless of the number of applications, or the time of application of CCC.

Work with hybrid geranium has shown that time to 1st flower anthesis was correlated with the area of the 1st 6 to 8 leaves (3), and that high light intensity accelerated flower initiation (1, 12). The influence of light intensity on flowering time is thought to be correlated to increased carbon fixation, and this fixation might result in rapid initiation (2). Jensen (9) reported that CCC accelerated flowering in hybrid geranium by reducing the light requirement for flower initiation.

Ferree and Hall (6) found that a single application of SADH had no measurable influence on photosynthesis or transpiration of apples leaves. Halfacre et al. (7) reported that SADH treatment resulted in decreased photosynthesis rates.

The object of this research was to determine the influence of CCC and SADH on photosynthesis, transpiration and photorespiration in hybrid geranium 'Sprinter Scarlet'.

'Sprinter Scarlet' plants were grown in a glass greenhouse at 21 ± 3°C night and 28 ± 5° day temperature. Plants were grown in 1 peat: 1 vermiculite (v/v) medium and were watered to saturation with 200 ppm N of 15N–7P–12.5K at each irrigation. Soil was leached every 4th to 5th irrigation with tap water to reduce soluble salt accumulation. Plants were grown for 9, 10, or 11 weeks (1st, 2nd, and 3rd experiments) in the greenhouse prior to application of growth regulators. At that time, 5 plants in each treatment were sprayed to run off with 5000 ppm SADH, 3000 ppm CCC, or distilled water (control). First measurements were made as

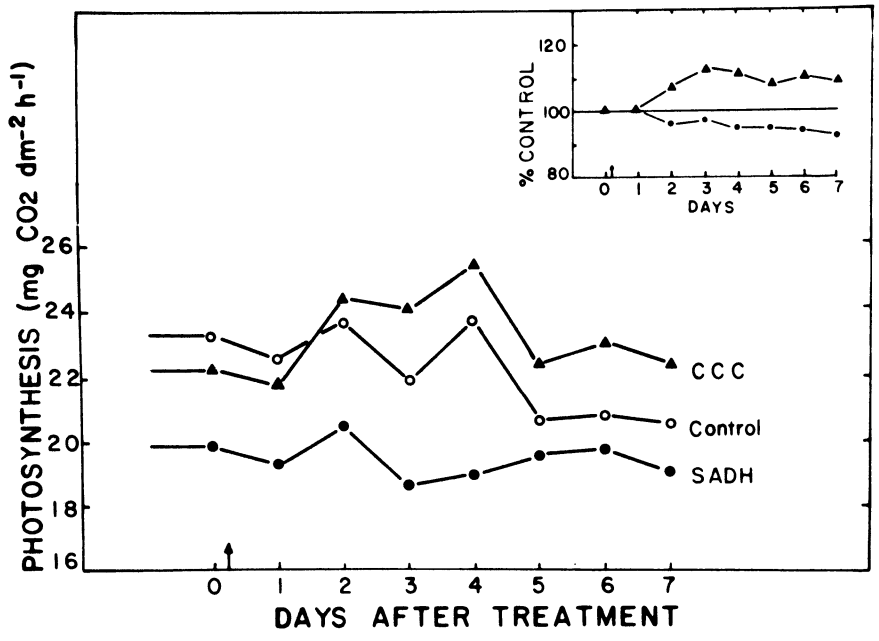

Fig. 1. The influence of CCC and SADH on net photosynthesis of 'Sprinter Scarlet'. Treatments were applied where arrow indicates.

Received for publication 3 Feb. 1984. The cost of publishing this paper was defrayed in part by the payment of page charges. Under postal regulations, this paper therefore must be hereby marked *advertisement* solely to indicate this fact.

[1]Recipient of a study stipend from the Chinese Agriculture Ministry. Permanent address: Rice Research Institute, Guangdon Academy of Agriculture Sciences, Guangzhou, Guangdong, The People's Republic of China.