6. Grant, W. F., *Plant biosystematics*, Academic Press, London, 1984, p. 674.

DIRECT PLANTLET FORMATION IN COTYLEDON CULTURES OF *CAPSICUM FRUTESCENS*

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A prerequisite for the improvement of any crop through tissue culture techniques depends upon the success in plant regeneration. Embryos[1,2] and seedling explants[3,4] have been used to raise complete plants in a large number of genera of Solanaceae. *Capsicum frutescens* is an important crop plant and earlier studies on tissue culture of this species concern plant regeneration from seedling explants of a hybrid variety (Bharath)[5] and anther culture[6]. The aim of the present study was to initiate tissue cultures of *C. frutescens* using seedling explants and to establish conditions for successful plant regeneration from cotyledons.

Seeds of *C. frutescens* obtained from the Southern Regional Plant Introduction Station, USA were germinated aseptically. Root, hypocotyl and cotyledon segments, measuring 0.5 to 1.0 cm in length, were excised from 4-week-old seedlings and implanted on Murashige and Skoog's medium (MS)[7]. Various auxins and cytokinins were added to the medium. The pH of the medium was adjusted to 5.8 before the addition of agar (1%). All the cultures were maintained under white fluorescent light for 16/8 h photoperiod at 25 ± 2°C.

All the explants callused efficiently on MS supplemented with 2 mg/l 2, 4-D and 1 mg/l Kn. Callus initiation was first noticed in root segments followed by hypocotyl and cotyledons. Compact, cream coloured calli were produced at the cut ends of cotyledon and hypocotyl. The entire root formed nodular callus which later turned brown. All the three explants implanted on MS + 0.5 mg/l NAA + 2 mg/l Kn or MS + 1 mg/l NAA + 1 mg/l, Kn produced 15–20 roots, in addition to callus. Rooting first appeared in cotyledon cultures, followed by root and hypocotyl segments.

Cotyledon explants showed direct plantlet formation (figure 1) on MS + 1 mg/l NAA + 1 mg/l Kn. However, the percentage of regeneration was low (about 10%). The plantlets were maintained on MS basal medium for 2 weeks (figure 2) and then transferred to a vermiculite irrigated with mineral nutrient solution (figure 3).

Thus it was possible to raise full plants from cotyledons in *C. frutescens* with a single hormonal combination. Efforts are being made to optimize conditions to increase the frequency of regeneration, and make the method efficient for quick multiplication of this crop plant.

Figures 1–3. 1. A four-week-old culture showing direct plantlet formation from cotyledon on MS + 1 mg/l NAA + 1 mg/l Kn; **2.** A five-week-old plant on MS basal medium; **3.** Seven-week-old plant in vermiculite.

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1. Christopher, T., Prolaram, B., Rajam, M. V. and Subhash, K., *Curr. Sci.*, 1986, **55**, 1036.
2. Christopher, T., Prolaram, B., Rajam, M. V. and Subhash, K., *Indian J. Exp. Biol.*, 1987, **25**, 349.
3. Kamat, M. G. and Rao, P. S., *Plant Sci. Lett.*, 1978, **13**, 57.
4. Gunay, A. L. and Rao, P. S., *Ann. Bot.*, 1980, **45**, 205.
5. Gunay, A. L. and Rao, P. S., *Plant Sci. Lett.*, 1978, **11**, 365.
6. Novak, *Z. Flanzensuechi*, 1976, **72**, 46.
7. Murashige, T. and Skoog, F., *Physiol. Plant.*, 1962, **15**, 473.

TOLERANCE OF ALGAL POPULATION IN RICE SOIL TO CARBOFURAN APPLICATION

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FURADAN 3G (3% carbofuran; 2,3-Dihydro-2, 2-dimethyl-7-benzofuranyl *N*-methylcarbamate), the most effective systemic insecticide against rice brown planthopper (*Nilaparvata lugens* Stal.), has been extensively used in rice cultivation. Recent reviews indicate that carbofuran affects soil microbial populations and various elemental transformations, mediated by micro-organisms, of importance to the soil fertility[1,2]. Several workers[3,4] reported its toxicity to pure cultures of algae. However, the influence of carbofuran on native populations of soil algae, a major ecologically beneficial group of soil microflora, has not been studied. An attempt was therefore made to determine the effects of carbofuran on quantitative and qualitative occurrence of algal population in flooded and non-flooded conditions of soil, as existing in rice culture.

Twenty-gram portions of air-dried and sieved black soil (pH 7.4, organic matter 1.82%, total nitrogen 0.044%), collected from a fallow rice field, were taken in test tubes (25 × 150 mm) and treated with aqueous solution of the commercial formulation of carbofuran (50 WP from Rallis India Ltd., Bangalore). Final concentrations were 0, 0.5, 1, 2 and 5 kg a.i. ha^{-1} on a dry weight basis[5]. For each treatment, one set of tubes was maintained at 50% water-holding capacity to provide non-flooded conditions while another set was flooded with distilled water to provide 1:1.25 soil to water ratio[6]. The tubes were incubated at room temperature (27 ± 4°C). After 10 and 20 days of the insecticide treatment, the soil samples, maintained under the two water regimes, were withdrawn for estimation of algae by enrichment culture technique followed by the most-probable number (MPN) method[6]. Fiducial limits for the MPN values of viable cells at the 95% confidence level were also calculated as outlined earlier[5]. Most of the algal forms were identified at least to genus and the frequency of their occurrence was also recorded. No attempt was made to determine the actual number of filaments/cells of each alga which appeared in the MPN tubes.

Application of carbofuran, at 0.5 and 1 kg ha^{-1} levels, to soil under non-flooded conditions either did not affect or slightly enhanced the algal population (table 1). There was also a slight increase in the population size at 2 kg ha^{-1} level in 10 days after treatment but this concentration exhibited toxic effect towards the end of 20 days. Highest level (5 kg ha^{-1}) was significantly toxic to the algae at the start. Thus, only 8 out of 30 MPN tubes, maintained for this concentration, were scored positive for algae after 10 days. Carbofuran application up to 2 kg ha^{-1} level under flooded conditions of the soil significantly increased the algal populations and this trend continued for 20 days. On the contrary, application of carbaryl, a closely related methylcarbamate, to soil under flooded conditions was found to have an inhibitory effect on the population size of algae while a significant enhancement in algal population was observed with its addition up to 25 ppm to non-flooded soil[6]. Although carbofuran is known to yield significant amounts of carbofuran phenol upon its hydrolysis in predominantly anaerobic (flooded) conditions of soil[7], it would seem that the hydrolytic product of carbofuran had no toxic effect on soil algae. However, slight algal toxicity was observed with 5 kg ha^{-1} level. It is evident from these results that application of carbofuran, close to field doses, in rice cultivation would result in the enhancement of algal population in soil.

The soil algal populations belonged to four species of Chlorophyta and 9 of Cyanophyta (table