

## ***In vitro* studies on regeneration of *Chrysanthemum* cv. SNOW CEM**

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### **SUMMARY**

A successful procedure was established for *in vitro* regeneration from callus derived from shoot tip, leaf, axillary bud and internodal segment explants of *Chrysanthemum* c.v. SNOW CEM cultured on MS (Murashige and Skoog, 1962) medium supplemented with various combination of auxins (IAA and NAA) and cytokinins (BAP and kinetin). The biotechnological interventions were required to modify the plant traits as desired by consumer. The present investigation dealt with micropropagation of *Chrysanthemum* c.v. SNOW CEM and the highest regeneration frequency (45.3%) was recorded in SNOW CEM with axillary bud explant culture when MS medium supplemented with IAA (1.5 mg/l) and BAP (2.5 mg/l).

**Key words :** *In vitro*, Regeneration, *Chrysanthemum*, IAA, NAA, BAP.

*Chrysanthemum* (*Dendranthema grandiflora* Tzvelev) is one of the world's most leading and popular flower crop of commercial importance. The commercial cultivars are usually propagated vegetatively through cuttings and suckers. Breeding programmes have focused on improving various characteristics to enhance the ornamental value, including the colour, size and form of the flower, production quality and reaction to the environment (Broertjes *et al.*, 1980).

Biotechnology involving modern tissue culture, cell biology and molecular biology offers the opportunity to develop new germplasm that are better adapted to changing demands. In this way, extensive studies have been carried out with *Chrysanthemum* on various aspects of its biotechnology, such as micropropagation, adventitious shoot bud regeneration from various explants and somatic embryogenesis. *Dendranthema grandiflora* has been micropropagated to meet domestic and external market. The advantages are many including propagating large number of plants in a short period with genetically true to type nature. In addition, the rapid protocols also help in fundamental studies such as understanding of plant biology and applied areas like genetic engineering and development of pathotoxin resistant cultivars and stable mutants *in vitro*.

### **MATERIALS AND METHODS**

The present study was carried out at Tissue Culture Laboratory, Department of Genetics and Plant Breeding, College of Agriculture, Rajendranagar, Hyderabad during December, 2004 to August, 2005. From the Agricultural Research Institute, Rajendranagar, Hyderabad, the explants *viz.*, shoot tip, leaf, axillary bud and internodal

segment of *Chrysanthemum* cv. SNOW CEM were collected from their mother plants. Shoot tips of one cm length, leaf bits of one cm<sup>2</sup> size, axillary bud of 0.5 cm size and internodal segment of 0.5 cm length were excised and thoroughly washed with tap water followed by rinsing with distilled water and subsequently treated with 0.1 per cent mercuric chloride (HgCl<sub>2</sub>) for four minutes and rinsed with sterile double distilled water to remove any traces of mercuric chloride. They were placed on sterile filter paper to remove any excess moisture. The explants were inoculated on basal MS (Murashige and Skoog's, 1962) medium supplemented with different growth regulators like IAA, NAA, BAP and kinetin at different concentrations needed to achieve the target response. The number of regenerants produced and mean number of regenerants produced from different explants from the cultivar were studied periodically.

### **RESULTS AND DISCUSSION**

Four explants of SNOW CEM cultivar *viz.*, shoot tip, leaf, axillary bud and internodal segment have been studied for regeneration frequency on MS medium supplemented with growth regulators in varied concentrations of IAA (1.0, 1.5 and 2.0 mg/l) and BAP (2.0, 2.5 and 3.0 mg/l). IAA (1.5 mg/l) and BAP (2.5 mg/l) recorded high frequency of regeneration (45.3%) with axillary bud followed by IAA (1.0 mg/l) and BAP (2.0 mg/l) with 41.9 per cent when shoot tip was used as explant (Table 1). The frequency was relatively reduced.

MS medium supplemented with growth regulators of IAA (1.0, 1.5 and 3.0 mg/l) and kinetin (2.0, 2.5 and 3.0 mg/l) was used for regeneration studies in SNOW CEM cultivar explants *viz.*, shoot tip, leaf, axillary bud and

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