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Somatic embryogenesis and plantlet regeneration in chilli (*Capsicum annuum* L)

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Plant regeneration protocol through somatic embryogenesis, was developed from whole cotyledon explants of two local cultivars of chilli, Byadagi Dabbi and Sankeshwar Local. Explants cultured on Murashige and Skoog (1962) medium supplemented with different levels of thidiazuron (TDZ) (0.0, 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 3.5, and 4.0mg/l) induced somatic embryos and the best response was at 3mg/l TDZ. On an average, 51.3 percent explants responded in both the genotypes with over nine embryos per explant. Complete plantlet formation occurred on MS with 3mg/ABA or 2mg/l IBA. Among the different levels of ABA, IBA and higher levels of agar tried for maturation of somatic embryos and germination, ABA 3mg/l was found to be better.

Key words: Capscicum annuum, Chilli, Cotyledon, Somatic embryogenesis,

INTRODUCTION

Thilli (Capsicum annuum L.) also known as hot pepper is one of the most widely cultivated vegetable and spice crops in the world. It has become an essential component of diet of the rich and poor. Chilli is grown over an area of 1.7 million hectares in the world, with a production of 19.4 million tones. India is the largest producer of chillies with an area of 0.91 m.ha and production of about 0.97 metric tones. In India it is one of the important spices, earning foreign exchange. The susceptibility to many pathogens and insect pests limit the productivity of chilli crop. Selection of varieties tolerant to diseases and pests is a priority, but the progress is limited due to lack of resistance to many pests and diseases in the available germplasm. Transformation of crop plants with the desired genes from a different source to control pests and diseases is the focus of many plant genetic engineering research programmes, which require efficient and reliable regeneration protocol. Although chilli belongs to the Solanaceae family, whose members are easily amenable to tissue culture and transformation, it is recalcitrant to regeneration especially at the shoot elongation stage (Steinitz, et al., 1999; Ochoa-Alejo and Ramirez-Malagon, 2001; Pozueta, 2001) and the responses are genotype specific (Christopher, T. and Rajam, 1996; Ramirez-Malagon and Ochoa-Alejo, 1996). Though many crops are known to produce somatic embryos in vitro, only few reports are available on pepper somatic embryogenesis from zygotic embryos (Harini and Lakshmi Sita, 1993; Binzel, 1996; Bodhipadma, and Leungd, 2003),

leaf (Kintzios *et al.*, 2001) and seed (Kaparakis, and Alderson, 2002) explants.

MATERIALS AND METHODS

Explant preparation:

Two popular and local cultivars of this region –Byadagi dabbi and Sankeshwar local were used in this study. Seeds were surface sterilized with 5% (v/v) sodium hypo chlorite for 20 minutes, rinsed 4-5 times with sterile distilled water and germinated on half strength MS medium (Murashige and Skoog,1962) under dark incubation. Whole cotyledon explants from 15-day-old in vitro grown seedlings were used as explants.

Induction, maturation and germination of somatic embryos:

Explants were cultured on MS medium with different concentrations of (0.0, 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 3.5, and 4.0mg/l) thidiazuron and incubated in 16/8hrs light and dark cycles for the induction of somatic embryos. Response of individual cultures to somatic embryogenesis and the number of somatic embryos produced in each explants was noted 30 days after inoculation. Clusters of somatic embryos obtained were transferred to MS supplemented with various levels (1,2,3,4mg/l) of ABA or Agar (8,10,12,14,16mg/l) or IBA (0.1,0.5,1.0,1.5,2mg/l) for maturation and development. Response in each cluster was noted 30-40days after culture for germination and plant formation.

Data collected on somatic embryo induction and their