

ABIOTIC STRESS INDUCED OVER-EXPRESSION OF SUPEROXIDE DISMUTASE ENZYME IN TRANSGENIC *HEVEA BRASILIENSIS*

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Abiotic stress induced over-expression of superoxide dismutase (SOD), catalase and peroxidase enzymes were studied in the transgenic embryogenic calli of *Hevea brasiliensis*, clone RR1 105. Two month old calli were transformed using *Agrobacterium tumefaciens* harbouring the binary vector containing neomycin phosphotransferase (*npt-II*) for kanamycin resistance as the marker gene for selection, β -glucuronidase (GUS) as the reporter gene and the sequence coding for SOD enzyme under the control of FMV 34S promoter. The transformed calli were proliferated in modified Murashige and Skoog medium fortified with hormones 2,4-D (4.5 μ M), BA (2.2 μ M) and NAA (1.1 μ M). Over-expression of SOD, peroxidase and catalase enzymes in response to abiotic stresses like water stress, osmotic stress and different light regimes in transgenic embryogenic callus cultures were determined. Water stress was induced by the addition of different concentrations (0.2-1.0%) of phytagel and osmotic stress using polyethylene glycol (PEG), mannitol and sorbitol (2-10%) in the culture medium. More than 50% over-expression of SOD was observed when 0.4% phytagel was added to the medium and 40% over-expression was obtained when the culture medium was supplemented with 4.0% PEG. Catalase and peroxidase were also over-expressed correspondingly.

Key words: Genetic transformation, *Hevea brasiliensis*, Superoxide dismutase, Transgenic callus, S. Sobha (for correspondence), S. Sushamakumari, I. Thanseem, K. Rekha, R. Jayashree, R.G. Kala, P. Kumari Jayasree, M.P. Asokan, M.R. Sethuraj, and A. Thulaseedharan, Rubber Research Institute of India, Kottayam-686 009 India; A.M. Dandekar, University of California, Davis, CA, USA.

INTRODUCTION

The recent developments in genetic modification techniques have opened new avenues for the production of fertile transgenic plants with increased yield, stress tolerance and resistance to pathogens. Although, genetic transformation and stable integration of foreign genes have been successful in many cereal crops and herbaceous annual plants, the success in perennial tree crops is rather limited (Dandekar *et al.*, 1988; James *et al.*, 1989; Ueno *et al.*, 1996). In *Hevea brasiliensis*, *Agrobacterium tumefaciens* as well as particle bombardment mediated genetic transformation and subsequent plant regeneration have been reported earlier (Arokiaraj *et al.*, 1994 and 1998; Jayashree *et al.*, 2000).

Active oxygen species such as superoxide (O_2^-), hydrogen peroxide (H_2O_2) and hydroxyl radicals (OH) are produced in aerobic organisms as a consequence of oxygen metabolism (Asada and Takahashi, 1987). Green plant tissues may produce more active oxygen species than animal tissues, because plants generate oxygen during photosynthesis and consume it during respiration. Further, because of their growth under high light intensities and a high cellular concentration of dioxygen, plants are subjected to oxidative stress. Increased antioxidant enzyme activities have been reported in response to heat and light conditions that cause sunscald in vegetables, fruits and flowers (Rabinowitch and Sklan, 1980).