Effects of the Antibiotics Kanamycin, Cefotaxime and Carbenicillin on the Differentiation of Flax Hypocotyls

(Received: 01/09/1998)

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ABSTRACT

Preliminary experiments showed that untransformed flax tissues did not contain genes conferring NPTII activity, which gave resistance to kanamycin. Thus, kanamycin seems to work well as an in vitro selective agent in fiber and dual use Egyptian flax cultivars as well as Canadian oilseed cultivars at a concentration of 200 ppm. This made it possible to use the kanamycin selection scheme to separate transformed from untransformed cells. Also, 250 ppm cefotaxime and 500 ppm carbencillin used in flax transformation system to suppress Agrobacterium growth, were found to be effective and safe for using, since their effects on regeneration were found negligible.

Key words: Biotechnology, Flax, Linum usitatissimum, Kanamycin, Cefotaxime, Carbencillin.

INTRODUCTION

hen gene transfer is attempted using explants, it is convenient to have a selection scheme whereby transformed cells can separated from untransformed cells. Drug resistance is usually the marker of first choice in plant transformation experiments (Fraley et al., 1986). The nptII gene provides resistance to kanamycin or genticin (G418) and is useful as a selectable marker in many plant species (Fraley, et al., 1986) and was also effective in Linum (Jordan and McHughen, 1988; Zhan et al., 1988; McHughen, 1989; McHughen and Jordan, 1989; Dong and McHughen 1991; Dong and McHughen, 1993; Koronfel, 1994). Kanamycin is a milder selection agent than G418 (Dong and McHughen

Kanamycin resistance does not spontaneously appear in flax (McHughen, 1989; McHughen et al., 1989). The results of Gao et al., (1991) suggested that the presence of kanamycin in the medium did not significantly affect the stability of foreign protein production in genetically engineered plant cells. Generally, kanamycin is added to the growth medium in a concentration previously shown to be inhibitory for regeneration of untransformed flax cells.

However, the sensitivity of plant cells to the selection agent depends upon the genotype, the explant type, the developmental stage, and the tissue culture conditions and should, therefore, be determined under the actual conditions of the transformation and regeneration process. The objective of this study was to investigate the effect of the