

An Investigation in to Micropropagation and the Effect of Colchicine on *In Vitro* Ploidy Induction in *Narcissus tazetta*

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Abstract

In this research, micropropagation of *Narcissus tazetta* was investigated by using bulb twin-scale explants using four different concentrations of BA (0, 0.5, 1 and 2 mg/l) and, therefore, four different concentrations of IAA (0, 0.5, 1 and 2 mg/l) in combination with 2 mg/l BA. The leaf growth and number of bulblet were measured 4 and 6 weeks after culture. *In vitro* grown plantlets were used for colchicines-mediate ploidy induction. Plantlets were treated by colchicine at four concentrations (0.00, 0.05, 0.10 and 0.20 %) for 24 and/or 48 h and the percentage of explants survival rate was measured. Ploidy levels of survived explants were analyzed by chromosome counting of root tip cells using acetoorceine squash method and by flow cytometry of leaf samples. Results indicated that colchicine concentrations had a significant effect on plantlet survival. However, treatment duration and their interaction were not significant. Application of colchicine reduced plantlet survival rate. There was no significant difference among the colchicine concentrations (0.05, 0.10 and 0.20%) on plantlet survival. Of 72 treated plantlets with colchicine, 32 plantlets were survived. Results of ploidy evaluation indicated the cells showing two different ploidy levels, diploid ones with $2x=20$ chromosomes and mixoploids ones with $2x=20$ and $4x=40$ chromosome tissues. Also, the obtained results from flow cytometry confirm chromosome counting of root tip cells. From the survived plantlets, 16 diploid plantlet and 16 mixoploid plantlets were evaluated. The highest mixoploid rate was obtained in 0.10% colchicine for 24 h. After several subcultures there was no tetraploid plant.

Keywords: *Narcissus*, *Micropropagation*, *Colchicines*, *Polyploidy*, *Chromosome counting*, *Flow cyto metry*