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# ISOLATION OF PROTOPLASTS FROM MESOPHYLL CELLS OF *DENDROBIUM*

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This study is part of the Project NOVAORCHID designed to explore the possibility to create intergeneric somatic hybrids of orchids by protoplasts culture technology. Our work is mainly focused on the genus *Dendrobium*, one of the largest genera in the orchid family Orchidaceae. It occupies a foremost position in ornamental orchid cut flower industry because of its high number of flower per inflorescence and recurrent flowering. The demand of the international market for *Dendrobium* cut flowers is, therefore, continuously increasing.

Somatic hybridization through protoplast fusion offers the opportunity to broaden the genetic variability among *Dendrobium* and generate cultivars showing new, fascinating and persisting flowers.

Plants of *Dendrobium spp* were obtained after *in vitro* cultivation of protocorm-like bodies (PLBs) and from vegetative micropropagation of selected genotype. PLB were subcultured on a modified MS medium. Young leaves of plantlets regenerated from PLBs were used as the explants for protoplast isolation. In order to obtain the optimal conditions for protoplast isolation several procedures were tested. Three different kinds of enzyme solutions together with three different incubation times were examined. After digestion a new and effective method of protoplast suspension has been set up. High number of purified protoplasts were collected and their vitality was tested.

In conclusion, an efficient procedure for *Dendrobium* protoplast isolation and culture conditions is described by adjustment of different steps which result in the enhancement of protoplast yield.

By utilizing the method developed in the present study we are now carrying a large programme of somatic hybridization involving *Dendrobium* and *Phalaenopsis* orchid species.