Investigation on Regeneration From Seedling Tissues of *Campanula carpatica*©

Eline Kirk Mørk, Sridevy Sriskandarajah, and Margrethe Serek
Department of Agricultural Sciences, Laboratory for Plant and Soil Science, Floriculture, The Royal Veterinary and Agricultural University, 40 Thorvaldsensvej, 1871 Frederiksberg C., Copenhagen, Denmark

Margrethe Serek
Department of Natural Sciences, Floriculture, University of Hannover, Germany

Genetic transformation of several plant species by using useful genes is becoming more and more popular. All in vitro transformation protocols require efficient regeneration systems for the chosen plant. Efficient regeneration methods would make a large number of plant cells regenerative and make them more receptive to genetic modification. The efficient methods will also help to speed up the recovery of the whole plants from the transformed cells or tissues.

Totipotency is one of the main properties of plant cells, which ensures the possibility of survival of the plants under stressed conditions. In nature, this characteristic proves itself most distinctly in the ability of plants to use various pathways of vegetative reproduction and in the possibility of fast restoration of the lost or stress-damaged parts of shoots and roots. In in vitro conditions, practically any living cell with a nucleus can begin cell division and form callus or suspension culture, if it is placed on a medium containing the right nutrients. Thereby the formation of shoots, roots, or somatic embryogenesis can begin.

Generally, there are three methods available for regeneration. These are direct somatic embryogenesis, indirect somatic embryogenesis, and organogenesis. Both types of somatic embryogenesis occur by the formation of embryo-like structures, which may develop into whole plants. In the direct somatic embryogenesis, adventitious shoots are formed either from a single cell or from a group of cells. In the indirect somatic embryogenesis, an intervening callus phase has taken place, from which the embryos can be produced. Callus is a mass of undifferentiated paranchymatic cells, which grows and divides. In organogenesis, adventitious shoots are formed directly on explants or through callus phase.

It is common to use explants such as leaves, stems, roots, and pollen for regeneration. Several factors could influence the regenerative ability of explants. The genetic make up of the plant itself can have a significant influence on regeneration. It has been reported that even cultivars within a species can differ in their regenerative ability (Rivera-Domìnguez et al., 2004; Sriskandarajah et al., 2001; Chang et al., 1996; Arroyo and Revilla, 1991). In addition, several factors such as explant type and explant age can have an influence on regeneration. Immature tissues such as hypocotyls or young cotyledons are often the only parts of plants capable of regenerating.

As mentioned earlier, many factors can influence the regenerative ability of the plant. The most important exogenous factors, which could influence regeneration are temperature, light, and the composition of the growth medium. Work with *Campanula carpatica* has shown that explants incubated in the dark produced sig-

nificantly more shoots compared with those regenerating in light (Sriskandarajah et al., 2001). The amounts of nutrients and hormones in the growth medium were shown to have a significant influence on the regenerative ability in almost any species. Therefore, it is necessary to find the optimal medium composition for the chosen species. (Franklin et al., 2004; Lee et al., 2001; Sriskandarajah et al., 2001).

In the present study, we investigated the effects of germination conditions on the regenerative ability of the explants obtained from *C. carpatica* seedlings. The examined factors were the content of the seed germination medium and the light intensity. In addition, different types of explants were investigated.

The results of the present study indicated that the amounts of nutrients in the seed germination medium had a significant influence on subsequent regeneration of the hypocotyl explants. The results have also shown that the number of shoots formed on explants from seedlings grown under high light intensity decreased with increasing amount of salts in the medium. Low levels of salts appear to have encouraged vigorous root formation and the development of secondary leaves. When the different types of explants were compared, the results indicated that hypocotyls were significantly better than the cotyledons in producing adventitious shoots. The present study has also shown that the maximum number of shoots was produced by the hypocotyls taken from the seedlings grown in high light. Hypocotyls produced adventitious shoots faster than the cotyledons. Detailed information from this study can be found in a forthcoming publication under the title "Influence of seed germination conditions on regenerative ability in *C. carpatica*".

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