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analysis. Most of the analyzed callus clumps was identified as diploid (64%), however haploid tissues (5%) were also detected.

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Protoplast culture and plant regeneration from open-pollinated and hybrid cultivars of parsnip (*Pastinaca sativa* L.)

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Plants have a remarkable regeneration potential, which ensures their adaptational success. Protoplasts serve as a unique tool for a range of procedures focused on overcoming naturally occurring sexual incompatibility barriers and efficient genetic transformation of plant cells. Several parameters such as genotype, physiological status of the source tissue, culture medium and environmental conditions play a crucial role in the ability of protoplast-derived cells to exhibit their totipotency and to develop into fertile plants. We compared the regeneration ability of protoplasts derived from four cultivars of parsnip (*Pastinaca sativa* L.) representing both open-pollinated and hybrid cultivars. Although this member of Apiaceae family has been grown for human consumption for centuries, it is still considered as an *orphan* crop. Today parsnip is gaining popularity, mostly due to the increased product variety on the European market.

The optimal conditions for seed germination along with the most suitable age of donor plants were determined for each cultivar. The protocol for the plant regeneration from carrot protoplast developed by Grzebelus et al. [1] was implemented as the gold standard. The composition of media used for parsnip protoplast culture were modified and supplemented with selected polyamines and peptides. The regeneration step was preceded with the calli culture on the B5 medium supplemented with phytosulfokine to obtain proembryogenic mass. Mature somatic embryos of 'Pódlugi biały' cultivar successfully developed into plantlets on the regeneration medium.

Considering the growing economic importance of cultivated parsnip, the developed protoplast-to-plant regeneration protocol might aid traditional breeding programs. It is also a crucial premise for further attempts at protoplast fusion. It could be exploited as a tool for the transfer of cytoplasmic male sterility into other fertile Apiaceae species.

References

- [1] E. Grzebelus, M. Szklarczyk, R. Barański. An improved protocol for plant regeneration from leaf- and hypocotyl-derived protoplasts of carrot. *Plant Cell Tiss Organ Cult*, 109:101–109, 2012.

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