



Department of Biology and Ecology,
Faculty of Sciences and Mathematics
University of Niš
Institute for Nature Conservation of Serbia

ABSTRACTS APSTRAKTI

**14th Symposium
on the Flora of Southeastern Serbia
and Neighboring Regions**

Kladovo 26 to 29 June 2022

**14. Simpozijum
o flori jugoistočne Srbije
i susednih regiona**

Kladovo 26. do 29. jun 2022.

Niš-Belgrade, 2022

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An efficient *Agrobacterium tumefaciens* - mediated genetic transformation method for *Centaureum erythraea* via secondary somatic embryogenesis

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A genetic transformation method via secondary somatic embryogenesis is described for *Centaureum erythraea*. Cotyledonary somatic embryos (cse) induced on leaf explants were used for inoculation with *A. tumefaciens* strain GV3101 containing the binary vector pXK7S2D that provides kanamycin resistance. Inoculated embryos were cultured on MS medium enriched with 2,4-dichlorophenoxyacetic acid and *N*-(2-chloro-4-pyridyl)-*N'*-phenylurea that promote somatic embryogenesis, with addition of acetosyringone. Cse were transferred on induction medium supplemented with cefotaxime and 5 or 10 mg/L kanamycin for selection. Embryogenic tissue, induced on primary cse explants during the selection period in the dark, was transferred to hormone-free medium maintaining the same antibiotic combination. Morphologically normal secondary cse that survived kanamycin exposure were isolated and subcultured on hormone-free medium containing cefotaxime and IBA to enhance germination. Fully regenerated plants were analyzed by Phire PCR to determine the transgene presence. Transformation efficiency was higher on media with 10 mg/L kanamycin (17.64%) as compared to 6.67% efficiency obtained on 5 mg/L. To the best of our knowledge, this is the first report on centaury transformation via secondary somatic embryogenesis, which offers an alternative to leaf or root explant transformation and provides an additional tool for investigating *in vitro* developmental pathways in this plant species.

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