# BOOK OF ABSTRACTS

# 3rd International Conference on Plant Biology

## (22nd SPPS Meeting)

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**BELGRADE**

**Serbian Plant Physiology Society**

**Institute for Biological Research "Siniša Stanković", University of Belgrade**

**Faculty of Biology, University of Belgrade**

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## Transformation of tomato cultivar Moneymaker with *Agrobacterium tumefaciens*

PP1-3

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Tomato (*Solanum lycopersicum*) is not only one of the most important vegetable crops, but a widely used model for genetic improvement of dicotyledonous crops. Even though the first report on tomato transformation using *Agrobacterium tumefaciens* has been published over 30 years ago, numerous protocols published since then demonstrate that tomato transformation is neither standardized nor routine. Tomato cultivars and genotypes greatly differ in their susceptibility to transformation and response to growth regulator and antibiotics combinations used during regeneration and selection. Hereby we report a successful protocol for "Moneymaker" transformation with *A. tumefaciens* GV3101:pSM90 bearing pAGT174 with Kanamycin resistance cassette. Whole larger leaves (from ~10 cm high plantlets), rather than leaf segments were selected for inoculation, as the explant size significantly affected regeneration rate. Combination of zeatin (1 mg $L^{-1}$) and IAA (0.1 mg $L^{-1}$) was used for preculture and throughout the selection process, with addition of Timentin (200 mg $L^{-1}$) and increasing concentrations of Kanamycin (from 20 to 50 mg L 1) during the selection and regeneration. The regeneration on Kan occurred almost exclusively in transformed explants and proceeded via organogenesis with the callusing stage. Using Timentin instead of commonly used Cefotaxime to kill the bacteria proved beneficial for Moneymaker regeneration. In this system the regeneration frequency was 7%. Plantlets that survived Kan selection were PCR-tested, and 89.6% of them were transformed. The protocol is convenient and robust in terms of very low false-positive rate (10.3%).

*Keywords:* Moneymaker, transformation, Timentin, Zeatin

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## Distribution of some arabinogalactan protein epitopes during somatic embryogenesis and organogenesis on leaf explants of centaury (*Centaurium erythraea* Rafn)

PP1-4

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Arabinogalactan proteins (AGPs) are a family of ubiquitous hydroxyproline-rich glycoproteins present in plasma membranes, cell walls and secretions of plants. Since AGPs are highly glycosylated, more than 90% of their total molecular mass comes from carbohydrate moieties consisting of various arabinogalactosyl chains (AG sugar chains), which are thought to be important for the

diverse functions of AGPs. AGPs are implicated in many aspects of plant growth and development, including cell differentiation, organogenesis and somatic embryogenesis (SE). The localization of AG sugar chains in plant tissues can be visualised using monoclonal antibodies (mAbs) that can detect different AGP epitopes. The aim of this study was to investigate changes in the localization of AGPs during induction of indirect SE and shoot organogenesis on leaf explants of centaury. Immunofluorescence labelling of leaf sections was performed with a set of mAbs (MAC207, JIM4, JIM8, JIM13, JIM15, LM2 and LM14). The results revealed that AGPs recognized by all mAbs tested were expressed in numerous meristematic cells from which somatic embryos develop. The AGP epitope recognised by the JIM4 antibody showed stronger intensity of immunofluorescence in the cell walls of protodermal cells of globular somatic embryos, whilst MAC207 and JIM13 epitopes were detected in cotyledonary somatic embryos. Strong immunofluorescence of the AGPs epitopes recognized with JIM4 and MAC207 mAbs were observed in the cells of apical meristem and leaf primordia of adventitious buds. These results suggest that AGPs play an important developmental role during formation of somatic embryos and adventitious buds from leaf explants of centaury.

***Keywords:*** somatic embryo, shoot organogenesis, JIM4, JIM13, MAC207

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## Glandular trichomes on the leaves and flowers of *in vitro* cultured *Micromeria thymifolia* (Scop.) Fritsch: morphology, distribution and histochemistry

PP1-5

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The genus *Micromeria* Bentham (Lamiaceae) comprises more than 70 aromatic species distributed throughout the temperate belt. Micromeria species produce considerable quantities of the essential oils, the biosynthesis, accumulation and secretion of which are generally restricted to glandular trichomes. *Micromeria thymifolia* (Scop.) Fritsch is a Balkan endemic species, traditionally used as a condiment and medicinal plant in the Mediterranean area. In view of the potential pharmacological and commercial value of this species, the glandular trichomes of *in vitro* cultured *M. thymifolia* on both vegetative and reproductive organs were examined, in relation to their morphology, distribution and histochemistry of the main secretion compounds, using light and scanning electron microscopy. Leaf indumentum of *in vitro* grown *M. thymifolia* comprised