

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

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**Programme and Abstracts** 





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nainly distributed in Asia, contain various biologiclassified into three catse plants are used as asantihepatotoxic, antifuning, wound healing, ton-

cordifolia, as well as the n the extract was detervonol contents were deof methanol extract of B. nd reactive oxygen speation scavenging ability

r amounts of flavonoids el of total antioxidant ac-DPPH assay ( $IC_{50}$  = 12.12 radical scavenging assay cid, quercetin and rutin. mages to biomolecules. This research was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (grant No. III 43004).

## Radical scavenger activity and chelating ability of *Filipendula hexapetala* Gilib. root extract

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Filipendula hexapetala Gilib. belongs to the genus Filipendula (fam. Rosaceae). F. hexapetala (dropwort) has a long history of use in folk medicine and phytotherapy in Serbia. Powdered roots of F. hexapetala (tuberous roots containing starch are edible but bitter), or used in decoction, were utilized to treat kidney problems, breathlessness, wheezing, sore throats and congestion. Also, it is frequently used to treat stomachache and diarrhea. The aim of this study was to evaluate the antioxidant and radical-scavening activities of methanol root extract of *F. hexapetala*. Amount of total phenols, monomeric anthocyanins and total anthocyanins were also determined. The content of total phenolics in the extract was determined according to the Folin-Ciocalteu procedure, while the pH-differential and single pH methods were used to determine anthocyanin contents. The antioxidant activity of the methanol extract of F. hexapetala roots was assessed in vitro by the assay for DPPH free radical scavenging ability (DPPH), the superoxide radical scavenging assay and the assay for ferrous ion chelating ability. The methanolic extract showed high total phenolic content (236.47 mg GA/g). The amounts of monomeric anthocyanins and total anthocyanins recovered were 0.56 and 1.36 mg/g dry extract, respectively (expressed as cyanidin-3-glucoside equivalent). The investigated extract expressed a strong antiradical activity in the DPPH assay ( $IC_{50} = 26.3 \,\mu g/mL$ ) and in the superoxide radical scavenging assay ( $IC_{50}$  = 432.27 µg/mL). Therefore, the extract has been proven able to prevent the initiation of free radicalmediated chain reactions. Finally, tested extract have not shown ferrous ion chelating capacity in used concentration (IC $_{50}$  > 8000  $\mu$ g/mL). Investigated root extract showed high level of radical scavenger activity compared to natural antioxidants such as gallic acid, ellagic acid, quercetin and rutin.

This research was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (grant No. III 43004).

### Sesquiterpene lactone production in transformed chicory

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Chicory (Cichorium intybus L.) is a plant valued for its medicinal and culinary properties. C. intybus owes its healing effect, at least in part, to a group of secondary metabolites known as sesquiterpene lactones. These compounds are shown to have anti-feedant, anti-fungal, anti-bacterial, anti-protozoan, schistomicidal, molluscicidal and anti-cancer effects. Since they are present in relatively small amounts in the plant, there has been ongoing interest in ways to improve the production and yield of these compounds. One

way of achieving this goal is Agrobacterium-mediated transformation, which is known to boost secondary metabolism. Chicory is readily transformed with A. rhizogenes to produce hairy-roots, from which whole plants can be regenerated. Several clones previously obtained through A. rhizogenes A4 transformation were analyzed by targeted LC-MS for the content of 8-deoxylactucin, lactucin, dihydrolactucin and lactupicrin. Three clones and one untransformed plant were chosen for the analysis, based on growth, biomass production and initial screening for these metabolites. Both leaves and roots of in vitro grown plants were analysed, in two growth phases: non-flowering rosette and flowering elongated plant. It was found that the selected clones indeed produce more sesquiterpene lactones than the control plants, and that this was dependent on the plant organ and growth phase. Roots in general contain more of these compounds than leaves, and roots of flowering clones have higher content of all target sesquiterpene lactones than control plants. Since these plants were transformed with the native Ri plasmid (pRiA4-GUS), expression of bacterial genes - rolA, rolB and rolC, which are known to affect secondary metabolism, was quantified, in an attempt to correlate elevated compound content with the rol genes' transcript levels. Expression was highest for the rolC, and barely measurable for rolB. Higher transcript level of these genes was detected in roots, and all three clones had high expression of rolC in the roots of flowering plants, which coincides with sesquiterpene lactones presence.

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#### Profiling antioxidant activity in salt-stressed Carpobrotus rossii

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Carpobrotus rossii, commonly known as Pig Face, is a prostrate succulent with dark green, triangular shaped trailing foliage. Tolerant of most soil types and conditions, however, prefers an open, sunny position with welldrained soil. It is important as a medicinal plant used for the preparation of extracts that are applied in the treatment of cardiovascular and degenerative disease (diabetes, cancer, Alzheimer's, etc.). Considering that its leaves and fruits are edible, this plant is important for human nutrition. In this work, we precisely separated the metabolically active palisade mesophyll tissue from spongy parenchyma. Subsequently prepared methanol extracts from this succulent halophyte has been shown to have potent in vitro total antioxidant activity (TAC). Current evidence suggests that these activities are due to the presence of various antioxidant components that include a number of enzymes and low molecular weight compounds capable of quenching reactive oxygen species (ROS). The importance of photosynthetically active tissues and their involvement in ROS production is illustrated when one compares the difference in TAC, anthocyanidin (TACY) and total phenolic (TPC-gallic acid equivalents) production between the spongy parenchyma and the photosynthetically active mesophyll of the salt treated plants. Both, TPC and TACY content were much higher in mesophyll compared to parenchyma which is strongly correlated with expressed TAC. Despite very poor literature data of Carpobrotus phenolic composition, we acquired different phenolic profiles of parenchyma and mesophyll from salt treated plants. HPLC analysis of phenolic compounds shows the presence of phenolic acids, flavanols and flavonoids: catechine, epicatechine, chlorogenic acid and umbeliferone-like compound. The main lines of defense that protect cells against salt stress are various antioxidant enzymes: ascorbat peroxidase (APX), catalase (CAT), glutathione reductase (GR) and superoxide dismutase (SOD). These four antioxidant enzymes showed identical spatial patterns of expression among plant tissues. In control, as well as in treated plants, the enzymes activity was higher in the mesophyll, compared to parenchyma. However, antioxidant enzymes showed distinct was characteristic of the treated plants sults may be indicative of differential reent salt-treated tissues. The present sturn of various antioxidants and additional Our hypothesis that salt stress increase our observations in the salinity and cor Project 173040 of MESTD.

# Anti-hydroxyl radical activifluorescent spectroscopy

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The essential oil extracted from Mility in vitro using fluorescent spectrosco oxygen species. Indeed, fluorescence edropped to 5% of the initial intensity, immore, after 1 min the radical production by after 3 min it extincted. As it has received an analysed oil sample are piperitenor and menthone (5.30%). High quantities observed anti-hydroxyl radical activity, showed a promising antimicrobial pote ther investigations will focus on in vivo oil in food industry.

This work was supported by the Minis public of Serbia (grants No. 173029 and

### Secoiridoids and xanthone Centaurium erythraea plant

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The qualitative and quantitative cor of *Centaurium erythraea* plants expressir and roots used as control. The main acti (gentiopicrin and swertiamarin) and xa