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ABSTRACT BOOK

PP209. Assessment of sun protecting factor and in vitro cytotoxicity of methanolic pulp extracts from Serbian Cucurbita maxima in human keratinocytes

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Ultraviolet radiation from sun exposure causes harmful skin effects, such as dermal photoaging and DNA damage, mediated by oxidative stress. Pumpkin (Cucurbita maxima) is abundant in carotenoids, polyphenols, and tocopherols which have antioxidant ability and protect cells from damage making them attractive as potential natural photo-protectants for skin applications. Pumpkin pulp, considered as by-product of the food industry, is the least explored in terms of its biological activities, although it contains considerable amounts of bioactive compounds. The aim of this study was to evaluate in vitro sun protecting factor (SPF) and cytotoxicity of methanolic pumpkin pulp extracts (MPE). They were prepared from the material of 4 accessions MAX 113, MAX 118-1, MAX 117, and MAX 1 from the breeding collection of the Institute of Field and Vegetable Crops, Novi Sad, Serbia. These accessions were selected according to the highest carotenoid content and antioxidant capacity. The effect of MPE on human keratinocytes (HaCaT) viability was determined by crystal violet assay. In vitro SPF based on the Mansur equation and the absorbance measurements was chosen to screen the photoprotective potential of MPE. The results show that SPF values for extracts MAX 113, MAX 118-1, MAX 117, and MAX 1 were 2.351, 1.875, 4.573, and 3.812, respectively. The most pronounced in vitro SPF values were in extracts MAX 117 and MAX 1 which were previously shown to contain the high amounts of carotenoids zeaxanthin and β-carotene. In vitro study of cell viability in keratinocytes showed that MAX 118-1, MAX 117, and MAX 1 did not reduce the number of viable cells up to the concentration of 1000 μg/mL and thus might be considered as non-toxic. Among the analyzed extracts, extract MAX 113 reduced HaCaT cells viability after 24 h incubation in a concentration-dependent manner, where the highest concentration of 1000 μg/mL significantly reduced number of viable cells compared to the non-treated control. On the other hand, the treatment of keratinocytes using extract MAX 117 led to a significant increase in the number of viable cells at 1000 μg/mL concentration. That was the same extract that exhibited the highest SPF value. These data demonstrate that bioactive compounds from MPE could have potential as anti-photoaging agents and can be considered as non-toxic for the skin cells. Additionally, findings suggest MPE of C. maxima Duchesne cultivar MAX 117 from Serbian accession has the best photoprotective potential and could be useful as component in natural cosmetic products.

PP210. Oleuropein treatment attenuates ROS generation, inflammation, and H2O2-induced damage in human trophoblast cells

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Higher energy demands during pregnancy lead to enhanced oxidative phosphorylation and increased production of reactive oxygen species (ROS) which have been shown to act as important regulators of angiogenesis, proliferation, differentiation and invasion of placental cells - trophoblast, autophagy and other important physiological processes and tissue adaptations. This controlled placental oxidative stress and inflammatory response are essential for successful early pregnancy. However, unbalanced, excessive ROS production has adverse effects on pregnancy outcome, causing trophoblast cell damage and dysfunction. Antioxidant supplements have been proposed as a possible approach in the prevention and treatment of such disorders. Secoiridoid oleuropein (OLE) is the most abundant phenolic compound found in olive leaves and drupes and has been shown to display profound antioxidant and anti-inflammatory activities. Its effects on trophoblast cells remain unexplored. The aim of our study was to investigate the impact of OLE on HTR-8/SVneo extravillous trophoblast cell line against hydrogen peroxide (H2O2)-induced oxidative damage. Results have shown that treatment with OLE at selected concentrations (10 and 100 μM) for 24h prevented a decrease in cell viability compromised by H2O2. Levels of lipid peroxidation indicators (MDA and LDH) were markedly reduced, and protein carbonylation and nitrosilation were significantly attenuated in trophoblast cells treated with OLE compared to untreated control cells. Further, OLE treatment decreased intracellular ROS production at both concentrations and significantly reduced the activity of antioxidant enzymes catalase, glutathione peroxidase and superoxide dismutase as well as restored glutathione levels in H2O2-exposed HTR-8/SVneo cells. OLE reduced the protein expression of inflammatory factor iNOS and decreased mRNA expression of pro-inflammatory cytokines IL-6 and TNFα in H2O2-treated cells. Our results indicate that OLE may ameliorate cellular oxidative damage, reduce inflammation and increase cellular antioxidant capacity in human trophoblast cells. It should also be emphasized that this olive-derived bioactive compound per se did not lead to any adverse effects in HTR-8/SVneo trophoblast cells under the described conditions, confirming its safety in vitro.

PP211. Fumaria officinalis L. as a novel source of biologically active compounds

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Fumaria officinalis L. (Fumariaceae) is a scrambling annual plant, distributed and cultivated throughout Europe, and represents a component of various phytotherapeutic formulations in the European ethnobotany used in hepatobiliary dysfunction, illnesses of gastrointestinal and urogenital tracts, cancer, rheumatism, high blood pressure, and skin disorders.
The study aimed to determine the total polyphenol and flavonoid contents (TPC and TFC, respectively) in F. officinalis extracts prepared using maceration and heat-assisted extraction (HAE), as well as to investigate their cytotoxicity on human keratinocyte cells (HaCaT), antioxidant potential, and spectrophotometric sun protecting factor (SPFs).
The influence of extracts on the viability of keratinocytes was investigated in the MTT assay, while antioxidant capacity was tested using ABTS and DPPH assays. SPFs value was calculated using the Mansur equation and the absorbance measurements (290-320 nm) with the aim of expressing the photoprotective activity of the extracts.
The polyphenol concentration in lyophilized extracts was 165.5±1.1 mg/g (maceration) and 171.8±1.4 mg/g (HAE), whereas the flavonoid content amounted to 71.6±1.9 mg/g (maceration) and 80.4±1.6 mg/g (HAE). The results show that ABTS and DPPH radical scavenging capacity of lyophilized extracts was 11.5±1.0 mmol Trolox/g and IC50DPPH 0.36±0.03 mg/mL (maceration) and 12.9±0.8 mmol Trolox/g and IC50DPPH 0.28±0.02 mg/mL (HAE). SPFs values for the extracts at the concentration of 100 μg/mL were 1.162±0.005 (maceration) and 1.255±0.015 (HAE). The higher antioxidant potential and SPFs of HAE extracts can be explained by the higher polyphenol and flavonoid contents. In vitro cytotoxicity assay in HaCaT cells showed that both types of extracts at all tested concentrations (25-100 μg/mL) did not reduce the number of viable cells. Additionally, HAE extract significantly increased the number of viable keratinocytes at the concentration of 100 μg/mL.
The presented study provides evidence of the polyphenol and flavonoid yields in F. officinalis extracts and their biological/pharmacological potential. Due to its proven antioxidant potential and positive impact on human keratinocytes, HAE extract can add value and/or improve the quality of the existing pharmaceutical and cosmetic products for the skin.