

Olive Leaf and Seed Polyphenols: Preventing UV Damage and Skin Pigmentation

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(オリーブ葉および種子由来ポリフェノールによる紫外線損傷および皮膚色素沈着抑制効果)

Category : Kou

Thesis Summary

Olive leaves and seeds are by-products obtained during pruning and oil extraction processes, respectively. Cultivation of olive trees generates branch and leaf wastes, while olive oil production results in seed wastes. These by-products are rich in polyphenols—the main bioactive compounds found in olive leaves and seeds—making them promising candidates for the valorization of natural ingredients. However, the development of phytochemicals from olive leaves and seeds for cosmeceutical applications, such as sunscreen products, remains limited. Therefore, evaluating polyphenolic components from olive oil by-products for use as antioxidants, anti-melanogenesis agents, and skin protectants against UVB irradiation represents an innovative strategy for promoting natural (derived from nature or non-synthetic material) sunscreen products with fewer adverse effects.

Currently, skin cell damage caused by overexposure to UVB irradiation, which contains high-energy photons, is a major concern. Skin chromophores absorbed harmful UVB wavelengths in the dermal layer, leading to increased reactive oxygen species (ROS) production and structural rearrangement of nucleotides, which can contribute to skin cancer. Thus, antioxidants that scavenge free radicals and protect the skin from UVB damage are among the most effective treatments. Additionally, anti-melanogenesis activity is a desirable feature in commercial sunscreens, as UVB-stimulated melanocytes (particularly those producing pheomelanin) generated high levels of ROS (e.g., $O_2^{\bullet-}$, $\bullet OH$, and H_2O_2), which triggered melanin overproduction and increased the risk of pigmentation disorders.

In the present study (**Figure 1**), crude extract from olive leaves and seeds was partitioned using organic solvents with varying degrees of polarity (*n*-hexane, dichloromethane (DCM), and ethyl acetate (EtOAc)) to separate secondary metabolites based on their polarities. An antioxidant assay was carried out as part of this preliminary screening to isolate promising chemical compounds as skin protectants against UVB radiation in NHDF cells and anti-melanogenesis agents in B16 melanoma cells, sequentially. Furthermore, the isolation process was performed using various chromatographic techniques. The EtOAc-soluble fraction from olive seeds which showed potent antioxidant activity (IC_{50} 137.70 ± 3.5 $\mu g/mL$) led to the isolation of two phenolic lignans (pinoresinol (**1**) and olivil (**2**)), one phenolic aldehyde (3,4,5-trimethoxybenzaldehyde (**3**)), two phenolic secoiridoid glycosides (oleuropein (**4**) and oleuroside (**5**)) from EtOAc-soluble fraction of olive leaves (IC_{50} $= 63.50 \pm 1.70$ $\mu g/mL$) as well.

Of the isolated compounds, pinoresinol (**1**) derived from olive seeds and oleuropein (**4**) from olive leaves exhibited pronounced bioactivities. While pinoresinol is a known compound, this study marks its first isolation from olive seeds, highlighting olive seeds as a previously unrecognized source. Pinoresinol (**1**) exhibited a protective effect against UVB-induced fibroblast damage (300 mJ/cm²), without showing cytotoxicity at a concentration of 100 µg/mL (279.03 µM). Its UVB-protective effect was attributed to its free radical scavenging activity (IC₅₀ = 30.75 ± 1.6 µg/mL). Furthermore, at a low concentration of 1.25 µg/mL (3.50 µM), pinoresinol (**1**) significantly reduced intracellular melanin production in B16 melanoma cells, surpassing the efficacy of kojic acid (used as a positive control) without cytotoxicity. Oleuropein promoted fibroblast proliferation at 100–200 µg/mL (185.01–370.02 µM) and skin protection at 200 µg/mL (370.02 µM), after UVB exposure (250 mJ/cm²). Compared with gallic acid as the positive control for skin protectant, oleuropein (**4**) showed a higher ability to promote cell proliferation, indicating its potential for application in wound healing. The protective effect for oleuropein (**4**) was supported by antioxidant activity against DPPH free radicals (IC₅₀ = 35.48 ± 2.68 µg/mL). Additionally, oleuropein (**4**) exhibited the most potent anti-melanogenesis effect at 100 µg/mL (185.01 µM), without cytotoxicity.

In conclusion, this dissertation has successfully identified bioactive polyphenolic compounds from olive leaves and seeds, which are agricultural by-products generated during pruning and oil extraction. These compounds exhibited significant UV protective and anti-melanogenesis properties with minimal cytotoxicity. The findings support the valorization of olive residues as effective natural ingredients for cosmeceutical applications, particularly in the development of antioxidant-rich, skin-protective sunscreen formulations with reduced adverse effects.

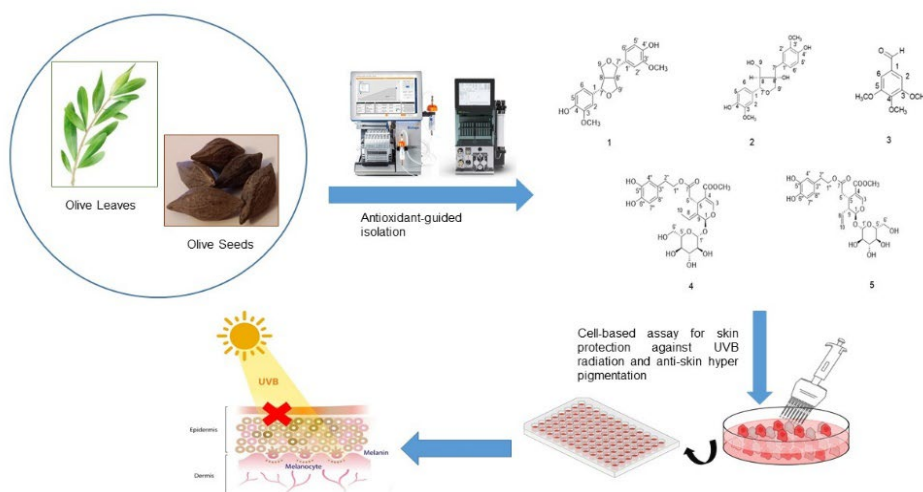


Figure 1. Research design for isolating secondary metabolites from olive leaves and seeds for UVB protection and anti-melanogenesis applications.