A Polyphenol-Rich Grape Pomace Extract Obtained by Subcritical Water Extraction Modulates Oxidative Stress, Nrf2 Signaling, and Stress Granule Formation in Human Bronchial Epithelial Cells

Federica Affranchi[1], Chiara Zichittella[2], Raffaele Raimondo[3], Pavel V. Kieslev[3], Giuseppe Calvaruso[1], Michela Giuliano[1], Antonietta Notaro[1]

[1]Dipartimento di Scienze e Tecnologie Biologiche, Chimiche e Farmaceutiche (STeBiCEF), Plesso di Biochimica, Policlinico Universitario, Università degli Studi di Palermo,, Palermo, Italy. [2]Dipartimento di Biomedicina, Neuroscienze e Diagnostica avanzata (BiND), Sezione di Biochimica, Università degli Studi di Palermo,, Palermo, Italy. [3]Azienda Mater Soc. Cons a R.L., Napoli, Italy

Abstract

This study investigates the biological properties of an extract (SWE) from Sicilian grape pomace obtained through Subcritical Water Extraction, an innovative and eco-friendly method for isolating bioactive compounds. Natural polyphenol-enriched extracts can mitigate oxidative stress by neutralizing ROS and free radicals, linked to cellular damage and disease.

Chemical characterization of SWE revealed high levels of phenolic acids (gallic, syringic, and protocatechuic acids). Cell-based assays using H_2DCFDA in human bronchial epithelium (HBE) cells showed that the extract reduces both basal ROS levels and those induced by acute sodium arsenite treatment.

Natural antioxidants exert their protective effects through a direct scavenging activity or by activation of antioxidant signaling pathways, like the Nrf2 pathway. In our model, SWE treatment induced no significant changes in total Nrf2 levels. However, following subcellular fractionation, we detected a dose-dependent increase in the phosphorylated, active form of Nrf2 within the nuclear compartment. This activation led to the upregulation of key antioxidant enzymes, including heme oxygenase-1 and SOD-2, thereby enhancing the cellular defense against oxidative stress. Furthermore, in response to sodium arsenite-induced stress, HBE cells assemble stress granules (SGs), membrane-less structures that form as one of the first responses to cellular stress. We observed that SWE prevents sodium arsenite-induced SG formation, helping to reduce the risk of stress granule–related cellular dysfunction.