# Optimization and characterization of olive leaf (*olea europaea*) obtained ultrasonic stimulations and vacuum by response surface methodology

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## ntroduction:

Olive leaves, an agricultural byproduct, are rich in (poly)phenolic compounds specially oleuropein and hydroxytyrosol, which possess recognized antioxidant, anti-inflammatory, and antimicrobial properties. Conventional extraction methods for these bioactive compounds often rely on large volumes of solvents, and time, raising environmental and economic concerns. To address these challenges, an innovative extraction approach combining temperature, ultrasound, and vacuum has been developed to enhance efficiency while minimizing solvent usage and processing times. This study aims to optimize the extraction process of bioactive compounds from olive leaves, focusing on maximizing yield. It seeks to quantify and optimize phenolic compounds such as oleuropein and hydroxytyrosol in the extracted samples and to evaluate the effects of varying extraction parameters on compound composition and the biological potential measured by the antioxidant activity under different processing conditions.

Methods:



The extraction process was conducted under varying experimental conditions, including powder:water ratio, water:ethanol ratio, temperature, extraction time, vacuum pressure, and cycles of ultrasound (US) and vacuum stimulation. High-Performance Liquid Chromatography with Diode Array Detection (HPLC-DAD) was employed to quantify the bioactive compounds hydroxytyrosol and oleuropein, along with the total phenolic content. The antioxidant activity of the extracts was analyzed using the Ferric Reducing Antioxidant Power (FRAP) assay and the 2,2-Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay.



### **Results:**







Figure 1. Surface response of antioxidant activity (DPPH) of the samples.



Figure 3. Representation of the amounts of hydroxytyrosol and oleuropein and the sample.



### **Conclusions:**

The optimal combination of variables enhanced the extraction yield by up to 24% for bioactive compounds like oleuropein, compared to unprocessed samples. The biological activity improves with the number of cycles for DPPH and FRAP methods.