# Response surface methodology for the optimization of biophenols recovery from "alperujo" using supercritical fluid extraction

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"Alperujo" or "alpeorujo" is a humid semi-solid by-product generated during two-phase centrifugation system for olive oil extraction. Several studies have reported the phytotoxic effects of olive mill wastes mainly due to its high phenolic content, which leads to a serious disposal problem since 800 kg of alperujo result from each ton of processed olives. Bioactive compounds present in alperujo can have health and well-being applications. The study of technologies that maximize the extraction yield of such bioactive compounds, at low cost and using enviromentaly friendly solvents is needed to make a profitable use of this by-product, simultaneously reducing the environmental burden of wastes contributing to a sustainable agriculture system. In this work, supercritical fluid technology was used to obtain natural extracts from alperujo aiming at maximizing the total phenol content and antioxidant activity obtained.

Fresh alperujo from Arbequina cultivar was collected directly from the almazara and dried in conventional oven at 40ºC, 22 hs. A total of 15 extractions were carried out following a Box-Behnken Design in which the three independent variables considered were: extraction temperature (40-60ºC), extraction pressure (200-400 bar) and % ethanol as modifier (0-10%). The central point of the design was 50ºC, 300 bar and 5% ethanol. Supercritical fluid extractions were carried out using a laboratory-built system equipped with a 25 mL stainless steel vessel. For each extraction, 13g of dried alperujo were used. A constant flow rate of 0,5 L/min $CO_2$ was set and each run was finished when 100L of $CO_2$ were measured in the flow totalizer (approximately 3.5 hs). Total phenolic content and antioxidant activity (TEAC) of the extracts were determined using Folin-Ciocalteau method and radical ABTS assay respectively.

Extraction yield ranged from 3.3 to 12.6 % (w/w) and it was positively correlated with Pressure and % ethanol as co-solvent. Phenolic content of the extracts varied from 218.5 to 702.1 ppm. These data fitted a multiple linear regression model with 93.78% adjusted R-squared, being the values obtained positively correlated with temperature and % co-solvent. Antioxidant activity showed similar behavior as total phenolic content. According to these results, optimal conditions for the recovery of biophenols from alperujo can be achieved at 60ºC, 200 bar and using 10% ethanol as co-solvent. Experimental data resulting from these extraction conditions were very similar from those predicted by the model.