Ultrasound and salt-assisted liquid–liquid extraction as an efficient method for natural product extraction

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A simple, rapid and efficient ultrasound and salt-assisted liquid–liquid extraction (USALLE) method coupled with high-performance liquid chromatography (HPLC) has been introduced for extraction, clean-up and pre-concentration of oleuropein from olive leaves as a model analyte. In this technique, the plant sample was transferred into the extraction solvent (consisting of phosphate buffer and miscible organic solvents) and the mixture was exposed to ultrasonic waves. After ultrasonic-assisted extraction (UAE), phase separation was performed by addition of salt to the liquid phase. During salt-assisted liquid–liquid extraction (SALLE) the analyte was transferred into the supernatant organic phase. Various parameters that affect the extraction efficiency such as ultrasonic time and temperature, sample amount, type and volume of miscible organic solvent, type and concentration of salt and pH were evaluated and optimized. The calibration curve shows good linearity ($r^2 = 0.9934$) and precision (RSD < 5.5%) in the range of 2.5–50 μg mL$^{-1}$. The limit of detection (LOD) and limit of quantitation were 0.5 and 2.5 μg mL$^{-1}$, respectively. The recoveries were in the range of 90.0–97.0% with RSD values ranging from 4.0 to 6.5%. Unlike the conventional extraction methods for plant extracts no evaporation and re-solubilization operations were needed in the proposed technique.

1. Introduction

Sample preparation techniques are used to improve the performance of an analysis. The most important aims of sample preparation are: to eliminate or reduce interference, to enhance the sensitivity of the analysis by increasing the enrichment factor (EF) of the analyte and sometimes to convert the analytes into a more appropriate form that can be easily separated, detected, and quantified.1–4 The obtained sample in this step should have a high concentration of target analytes free of interfering compounds from the matrix. Therefore, extraction of target analytes is one of the most important steps in sample preparation.5

Several solid–liquid extraction (SLE) techniques such as soaking (maceration) extraction, soxhlet extraction (SE), supercritical fluid extraction (SFE) and distillation are available for natural product extraction.6–9 Choosing the appropriate extraction technique for extracting natural compounds depends on various process conditions such as temperature, pressure, shaking, and solvent type. Although applying heat, pressure and agitation usually leads to acceleration of the extraction process, its destructive effects on natural compounds must also be considered.

Conventional extraction of natural compounds by using maceration, SE and distillation techniques requires a large volume of organic solvent and a longer extraction time. Also, SE and distillation techniques have destructive effects on natural compounds due to the high processing temperature. In conventional solvent extraction (CSE) methods, due to the use of a large volume of extraction solvent and its incompatibility with analytical instruments, evaporation to dryness and reconstitution of the extract in a very small volume of appropriate solvent are essential.10–12 As a result, there is an increasing demand for extraction of natural compounds by using an appropriate extraction method with safe solvents at low temperatures. Ultrasonic-assisted extraction (UAE) and microwave-assisted extraction (MAE) methods present several advantages such as: increasing the extraction efficiency of target analytes, and decreasing the volume of the solvent and extraction time in comparison with conventional methods.13–17

Salt-assisted liquid–liquid extraction (SALLE) is based on the phase separation of water-miscible organic solvents from aqueous solutions by addition of salt.18–21 In this technique water-miscible organic solvents with low toxicity were used as extraction solvents. Compared to conventional liquid–liquid extraction (LLE) methods, in the SALLE technique large volumes of immiscible organic solvents and vigorous