Optimization by Ultrasound Assisted Extraction of the phenolic compounds from Myrtle leaves using Natural Deep Eutectic Solvent

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Keywords: NADES, *Myrtus communis*, ultrasound, polyphenols, antioxidant

Myrtle (*Myrtus communis* L.) is an aromatic medicinal plant that is native to the Mediterranean coast. Its berries, leaves and seeds are natural sources of bioactive compounds. Myrtle is traditionally used to treat gastrointestinal, urinary disorders and type 2 diabetes, however it is also used in the food and cosmetics industries [1]. These properties are secondary metabolite such as polyphenols, which are known for their antioxidant, anti-inflammatory and hypoglycaemic effects. Nevertheless, conventional extraction methods involve long extraction times and the use of toxic solvents. The aim of this study was to develop a sustainable method for recovering polyphenols from dried myrtle leaves.
To this aim, NADES (Natural Deep Eutectic Solvents) were chosen for extraction, these are green and sustainable solvents obtained from the combination of a donor (HBD) and an acceptor (HBA) of hydrogen bonds, capable of forming a deep eutectic characterized by low toxicity and high extraction efficiency [2]. In addition, these solvents were combined with ultrasound, a sustainable extraction technique that improving extraction yield thanks to the cavitation [3].

First, the extraction efficiency of NADES, obtained from different combinations of hydrogen bond donors and acceptors, was evaluated by assessing the total phenolic content, antioxidant activity and hypoglycaemic activity in the myrtle extracts using spectrophotometric assays (DPPH, ABTS, Folin–Ciocalteu and α-glucosidase inhibition). Additionally, the polyphenolic fraction of the extracts was characterised using liquid chromatography coupled with high-resolution mass spectrometry to identify the primary metabolites in myrtle leaves. Next, the main factors influencing the extraction process (temperature, number of cycles, HBD/HBA ratio and percentage of water in the solvent) were optimised using a chemometric approach with a multivariate response surface design. This was done to identify operating conditions that would maximise antioxidant and hypoglycaemic activity while minimising energy consumption and environmental impact.

The extract obtained under optimal conditions was compared with a hydroalcoholic extract containing a comparable amount of ethanol to the NADES, as well as an extract obtained by traditional maceration in methanol. The results demonstrated the high extraction efficiency of the NADES-ultrasound system compare to conventional method, thereby confirming the effectiveness of this method as an environmentally friendly, high-performance alternative for extracting polyphenols from myrtle.

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