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Natural deep eutectic solvents to the extraction of bioactive compounds from orange peel and antioxidant capacity of extracts. Replacing organic conventional solvents?

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PURPOSE OF THE ABSTRACT

INTRODUCTION: The residue generated after the technological treatment of plant raw materials is a source of bioactive compounds available for obtaining products with high added value. Natural Deep eutectic Solvents (NADES), as alternative solvents, can present physicochemical properties of interest in the process of extraction and conservation.

OBJECTIVE: This work aims to evaluate the antioxidant capacity of extracts obtained by heating and stirring (HS) and ultrasound treatment (US) using four NADES as solvents, shown in Table 1, and to determine the concentration of ascorbic acid (AA) and total soluble phenols (TSP) on these extracts.

METHODS: 45 mL of each NADES or 50% ethanol were employed for the extraction of 3 grams of orange peel. Each extract was tested in triplicate. The treatment conditions for HS were 20 minutes at 40 ± 5 ?C in agitation and for US, cycles of 1 minute were performed at 40% amplitude, 200 watts, and 40 ± 5 ?C, with 2 minutes of cooling between cycles. AA was determined by the volumetric method of 2,6-dichlorophenolindophenol [1] and TSP by spectrophotometry at 745 nm using the Folin?Ciocalteu method [2] with Lambda 365 UV-vis (Perkin-Elmer, USA). The antioxidant capacity of the extracts was evaluated by three methods: ferric reducing antioxidant power (FRAP) [3] by UV-vis spectrophotometry at 593 nm, oxygen radical absorbance capacity (ORAC) [4] by fluorimetry with an excitation/emission maximum wavelength of 485 and 535 nm respectively, and 2,2-diphenyl-1-picrylhydrazyl radical scavenging activity assay (DPPH) [5] by spectrophotometry at 512 nm. ORAC and DPPH were carried out on the Wallac 1420 VICTOR2 multilabel counter (Perkin-Elmer, USA). Subsequently, the statistical treatment of the results was carried out by means of two factors ANOVA and Pearson's correlation test (Statgraphics®).

RESULTS: The extract obtained with ChChl:Pro:Ma presents the highest antioxidant capacity measured by FRAP, being HS (3.2 ± 0.2 mM TE) and US (5.8 ± 0.9 mM TE)followed by the ChChl:La extract. Furthermore, in the case of ChChl:Pro:Ma, there is a positive correlation between FRAP and TSP values (R=0.9664, p=0.0017). The extract obtained using ChChl:Pro:Ma measured by ORAC presents the highest antioxidant capacity for HS (16.5 ± 3.7 mM TE) and US (17.7 ± 1.4 mM TE) followed by the ChChl:Fru extract, in which the values obtained correlate with the TSP content (R=0.8516, p=0.0314). The antioxidant capacity, measured by DPPH of the ChChl:La extract presents the highest antiradical levels being 812.8 ± 7.6 and 840.5 ± 14.4 µM TE, for HS and US, respectively. In the case of ChChl:Fru, the concentration of ascorbic acid in the extract correlates with the antioxidant capacity by DPPH (R=0.9455, p=0.0044). The highest concentration of TSP is obtained using ChChl:Fru in both HS (79.0 ± 4.1 mg/100 mL) and US (91.0 ± 2.4 mg/100 mL), while with ChChl:La, the highest AA concentration is obtained 28.8 ± 2.4 and 40.6 ± 4.4 mg/100 mL for HS and US respectively, reaching similar levels to 50% ethanol extraction by US.

CONCLUSION: The cavitation phenomenon increases the extraction of bioactive compounds, in addition, if the choice or design of the solvent used is adequate, the interaction with the target compound can be increased. A higher antioxidant capacity, high redox potential, and polarity together with the acidic pH of the NADES, can contribute to the stability and shelf life of the extract, as well as, of its components separately. However, the antioxidant capacity can be overestimated due to the nature of the components that constitute the eutectic mixture and the method used for its evaluation.

FIGURES

NADES 75% (v/v)	Molar ratio	pH	Polarity (kcal mol^{-1})
Choline chloride : Fructose (ChChl:Fru)	1,9 : 1	4,02 ± 0,01	53,15 ± 0,87
Choline chloride : Glycerol (ChChl:Gly)	1 : 2	5,00 ± 0,00	48,85 ± 0,05
Choline chloride : Lactic acid (ChChl:La)	1 : 3	1,03 ± 0,02	47,97 ± 0,00
Choline chloride : L-Proline : Malic acid (ChChl:Pro:Ma)	1 : 1 : 1	2,06 ± 0,00	48,87 ± 0,19

FIGURE 1

Table 1. Acronym, molar ratio, pH and polarity of used Nades based on choline chloride as hydrogen bond donor.

v/v: volumen:volumen

FIGURE 2

KEYWORDS

Natural deep eutectic solvents | Bioactive compounds | Ultrasound treatment | Antioxidant capacity

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