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Exploring Natural Deep Eutectic Solvents (NADESs) for Extraction of Antioxidant Compounds from Aromatic Herbs used in Mediterranean Diet

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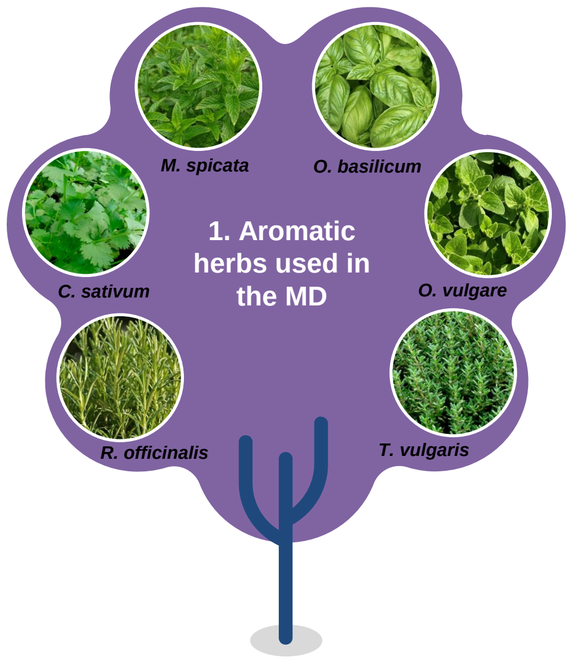
INTRODUCTION & AIM

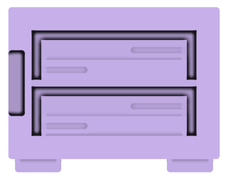
Aromatic herbs are a fundamental component of the Mediterranean Diet (MD), enhancing the flavor and aroma of dishes and serving as a healthier substitute for salt [1]. This not only improves the overall appeal of the diet but also contributes to its health benefits due to the richness of these herbs in bioactive molecules, such as phenolics with antioxidant properties [2].
The extraction of phenolics is a crucial phase in their application across various industries. However, the use of conventional solvents poses risks to both the environment and human health. Green solvents, such as Natural Deep Eutectic Solvents (NADESs), have been identified as a promising alternative due to their non-toxic, eco-friendly, and energy-efficient nature [3].

The **aim of this study** was to assess the efficacy of two NADESs – namely glycerol:urea (Gly:U) (1:1) and choline chloride:lactic acid (CC:LA) (1:3) – in comparison to the conventional solvent methanol for the extraction of phenolic compounds from six aromatic herbs commonly used in the MD: *Mentha spicata* L., *Coriandrum sativum* L., *Rosmarinus officinalis* L., *Thymus vulgaris* L., *Ocimum basilicum* L., and *Origanum vulgare* L..

METHODS



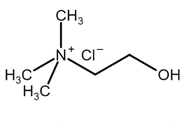
2. Oven-dried and grinding



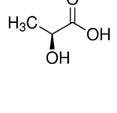
2 days, 40 °C

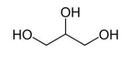
Plant powder

3. Extraction of phenolic compounds

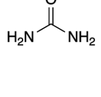


CC:LA (1:3)

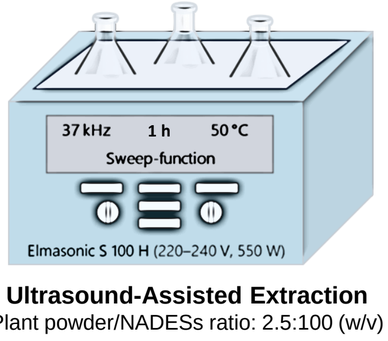




Gly:U (1:1)

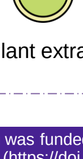


NADESs preparation
Agitation, 40 – 60 °C, 30-60 min



Ultrasound-Assisted Extraction
Plant powder/NADESs ratio: 2.5:100 (w/v)

4. Phytochemical and antioxidant activity analysis



Plant extract

- **Total phenolic content, TPC** (Folin-Ciocalteu method)
- **Total flavonoid content, TFC** (Aluminium chloride method)
- **Antioxidant activity:**
  - ORAC (Oxygen radical absorbance capacity)
  - DPPH (2,2-Diphenyl-1-picrylhydrazyl)

RESULTS & DISCUSSION

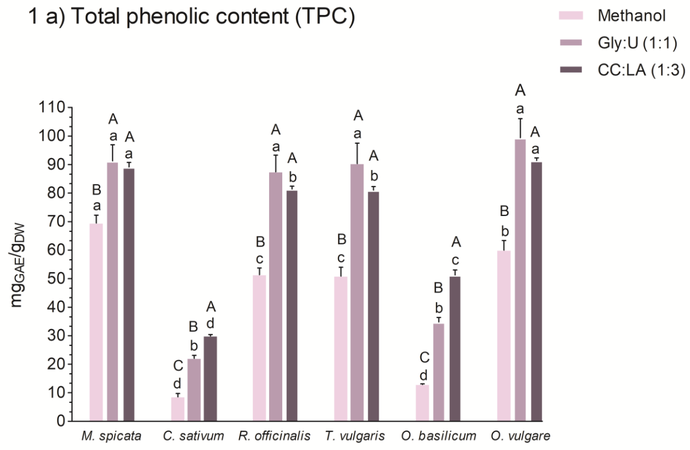


Figure 1. **a)** TPC and **b)** TFC of extracts from six aromatic species commonly used in the MD, using two NADESs and a conventional organic solvent (methanol).
Values are expressed as mean ± SE (n = 3). Different letters in each series indicate significant differences at $p < 0.05$ (Duncan's New Multiple Range Test). Capital letters indicate significant differences between the three solvents while lowercase letters evidence significant differences between the species. Notes: GAE, Gallic Acid Equivalents; QE, Quercetin Equivalents; DW, Dry Weight.

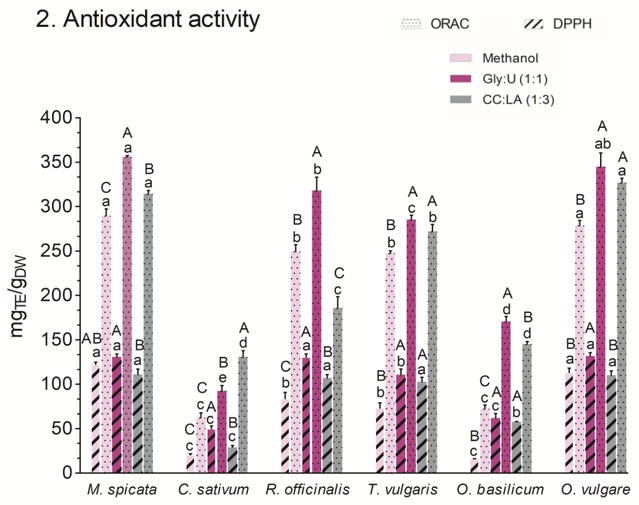


Figure 2. Antioxidant activity evaluated by DPPH and ORAC of extracts from six aromatic species commonly used in the MD, using two NADESs and a conventional organic solvent (methanol).
Values are expressed as mean ± SE (n = 3). Different letters in each method indicate significant differences at $p < 0.05$ (Duncan's New Multiple Range Test). Capital letters indicate significant differences between the three solvents (in each method) while lowercase letters evidence significant differences between the species (in each method). Notes: TE, Trolox Equivalents; DW, Dry Weight.

Both NADESs exhibited a higher capacity than methanol to extract phenolic compounds from all studied species.
Extracts from *O. vulgare, M. spicata, R. officinalis* and *T. vulgaris* obtained with Gly:U showed the highest TPCs.

Gly:U mixture also proved to be the best solvent for extracting flavonoids from the six studied aromatic herbs. Among these species, *M. spicata* extract showed the highest TFC (39.95 ± 0.55 mg_{QE}/g_{DW}), followed by *R. officinalis* and *O. vulgare*.

Both antioxidant activity assays demonstrated that Gly:U was the best solvent for extracting antioxidants from the six aromatic species studied, consistent with the results obtained for TPC and TFC.

M. spicata, O. vulgare, and *R. officinalis* extracts displayed the highest antioxidant capacities.

CONCLUSIONS

The findings suggest that Mediterranean herbs are an excellent source of antioxidant compounds and that extractions using NADESs are more effective than conventional solvents for extracting phenolics and promoting bioactivity. These green solvents align with sustainable practices for use in various industries, thereby enhancing the valorisation of Mediterranean herbs.

These plant extracts are being analysed by HPLC to assess their phytochemical profiles, and new biological activities will also be tested.

References: [1] Delgado, Gonçalves & Romano, 2023, Foods, 12, 840; [2] Gonçalves et al. 2017, J Food Sci Technol, 54(1):219–227; [3] Mansinhos et al. 2021, Antioxidants, 10, 582.

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