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IN-DEPTH LC-ESI/HRMS-GUIDED PHYTOCHEMICAL ANALYSIS AND ANTIOXIDANT ACTIVITY OF ECO-SUSTAINABLE EXTRACTS OF *CYNARA CARDUNCULUS* (CARCIOFO DI PAESTUM PGI) LEAVES

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SPOKE, WP AND TASK

SPOKE 8
WP8.1 Producing new products to upgrade waste value
Task 8.1.1 Valorisation of the waste by green chemistry to obtain high value molecules or new products

BACKGROUND AND AIM

The globe artichoke, known as *Cynara cardunculus* var. *scolymus* (Asteraceae), is a perennial plant highly valued for its edible immature flower buds which are harvested before they fully bloom [1]. Flower buds are commonly referred to as "capitula" or "heads" and represent the edible part; they are consumed fresh, canned, or frozen [2]. Italy is currently the leader in the production of globe artichoke, with 390 Kt, corresponding to 26% of the global production [3]. One of the most well known cultivars of artichokes in Italy is "carciofo di Paestum" PGI, which is prized by customers and chefs for its unique flavor, delicacy, and size. The main growing area for "carciofo di Paestum" is Paestum, the well-known archeological site in Campania region, also known for its bountiful plains and rich agricultural heritage. The outstanding flavor and quality of this artichoke variety are a result of the particular soil and environment [4]. During the industrial process, a large amount of byproducts, corresponding to external bracts and leaves, as well as stems, stalks, roots, and, to a lesser extent, seeds, is generated [5].

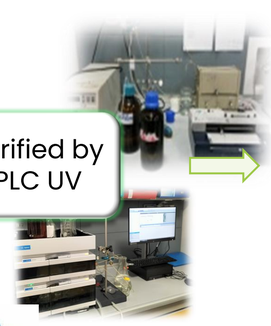
Supported by these premises, for the first time, eco-sustainable extracts of "carciofo di Paestum" PGI leaves were investigated in order to add value to this byproduct as a source of bioactive phytochemicals.

WORKFLOW

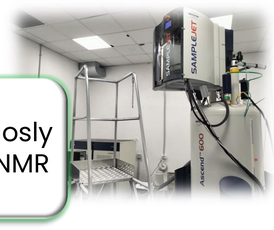
eco-sustainable EtOH: H_2O (50:50 and 75:25) extracts of "carciofo di Paestum" PGI leaves

LC-ESI/HRMS analysis of eco-sustainable extracts

Extracts were purified by HPLC-RI and HPLC UV

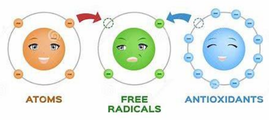


Structures of isolated compounds were unambiguously characterized by 1D and 2D NMR experiments



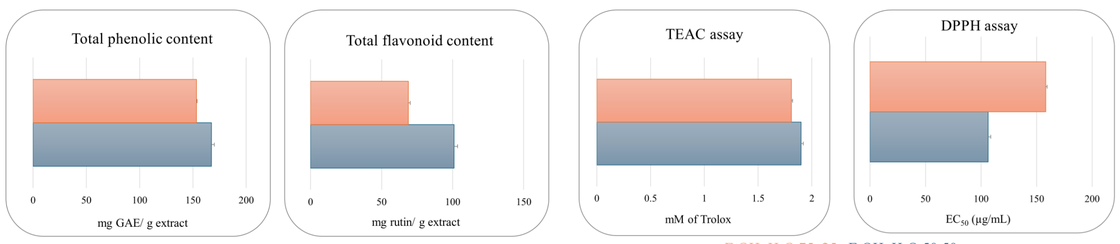
Evaluation of the total phenolic , flavonoid content and radical scavenging activity by DPPH and TEAC assays.

a cell-based antioxidant *in vitro* test was conducted on Caco-2 cells.



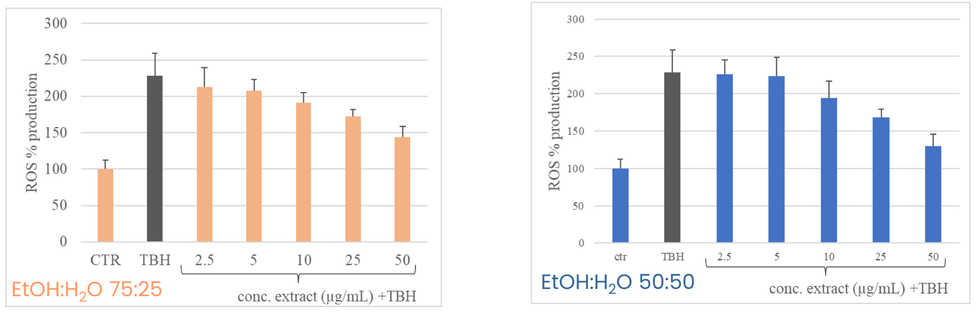
RESULTS

Evaluation of phenolic content, flavonoid content, and radical scavenging activity of extracts obtained from "carciofo di Paestum" PGI leaves



Total phenolic and flavonoid content, TEAC and DPPH assay of EtOH: H_2O (50:50) (blue) and EtOH: H_2O (75:25) (orange) extracts of "carciofo di Paestum" PGI leaves.

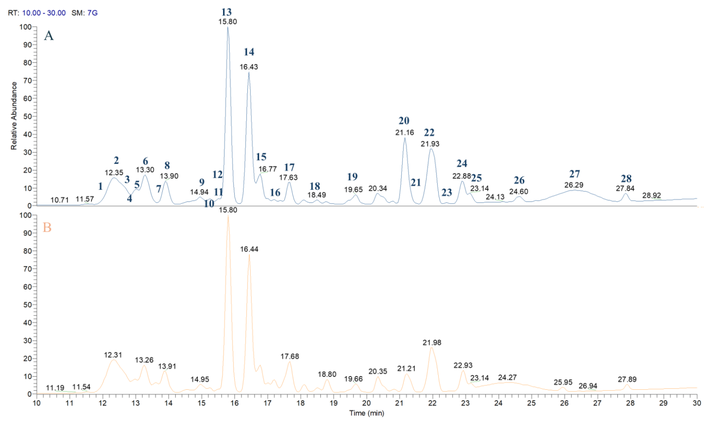
Evaluation of ROS production



Determination of intracellular ROS production. ROS level, visualized as H2-DCF-DA fluorescence and expressed as % of the control samples (non-oxidized nor pretreated samples), in Caco-2 after 120 min with TBH 2.5 mM and pretreated with the two extracts (2.5–50 µg/mL). $p < 0.001$ vs. control.

LCESI/LTQOrbitrap/MS analysis of "carciofo di Paestum" leaves extracts

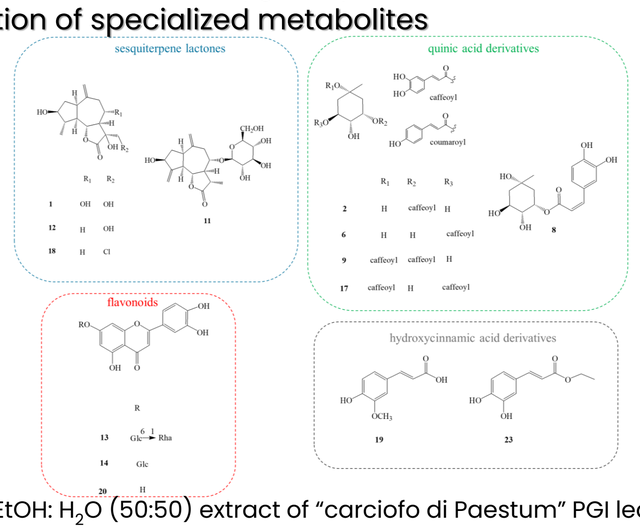
With the aim of identifying bioactive metabolites in eco-sustainable extracts of "carciofo di Paestum" PGI leaves, LC-ESI/LTQOrbitrap/MS was performed. Careful analysis of the LC-ESI/HRMS spectra allowed us to identify 28 compounds belonging to the class of sesquiterpenoids (**1**, **3**, **7**, **11**, **12**, **18**), megastigmanes (**4**, **15**), quinic acid derivatives (**2**, **6**, **8**, **9**, **10**, **17**), flavonoids (**13**, **14**, **20**), hydroxycinnamic acid derivatives (**5**, **19**, **23**), lignan (**16**), triterpenoid saponins (**21**, **25**), and polar fatty acids (**22**, **24**, **26**-**28**).



Eco-sustainable EtOH: H_2O (50:50 and 75:25) extracts of "carciofo di Paestum" PGI leaves

Isolation and Identification of specialized metabolites

In order to characterize unambiguously the main compounds in the eco-sustainable extracts of "carciofo di Paestum" PGI leaves, EtOH: H_2O (50:50) extract was purified by HPLC-UV and HPLC-RI. Compounds **1**, **2**, **6**, **8**, **9**, **11**-**14**, **17**-**20**, and **23** were isolated and unambiguously characterized by 1D and 2D NMR experiments.



Specialized metabolites isolated from EtOH: H_2O (50:50) extract of "carciofo di Paestum" PGI leaves.

CONCLUSIONS. In this study, the chemical and biological investigation of the eco-sustainable extracts of the Carciofo di Paestum PGI leaves was carried out. The ability of the extracts of reducing the production of free radicals in Caco-2 cell-line along with the identification of the specialized metabolites, mainly quinic acid derivatives, flavonoids and hydroxycinnamic acid derivatives, pointed to suggest recycling of artichoke byproducts, favoring the circular economy, and to support their use in the formulations of food supplements.

REFERENCES

1. Pedrali, D.; Zuccolo, M.; Giupponi, L.; Sala, S.; Giorgi, A. Plants 2024, 13. **2.** Borsini, A.A.; Llavata, B.; Umaña, M.; Cárcel, J.A. Foods, 2021. 10. **3.**Food and Agriculture Organization of the United Nations. FAOSTAT. Crops and Livestock Products. 2022. (accessed on March 2024). **4.** Cerulli, A.; Masullo, M.; Pizza, C.; Piacente, S. Molecules 2022, 27. **5.** Bonasia, A.; Conversa, G.; Lazzizera, C.; Elia, A. Plants 2023, 12.

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