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sciforum-134552: Polyphenol Bioaccessibility, Caco-2 Cell Uptake and Prebiotic Potential of Walnut Oil Industry By-products Following Simulated Digestion

Rafael Mascoloti Spréa [1,2,*], Daniele Bobrowski Rodrigues [1], Ricardo Calhelha [1], Tânia Pires [1], María Brassesco [3], Manuela Pintado [3], Susana Santoyo [4], Elena Arranz [4], Miguel Prieto [2], Lillian Barros [1] and Joana Amaral [1]

[1] CIMO, LA SusTEC, Instituto Politécnico de Bragança, Campus de Santa Apolónia, 5300-253 Bragança, Portugal
[2] Nutrition and Bromatology Group, Department of Analytical and Food Chemistry, Faculty of Food Science and Technology, University of Vigo-Ourense Campus, E-32004 Ourense, Spain
[3] Universidade Católica Portuguesa, CBQF - Centro de Biotecnologia e Química Fina – Laboratório Associado, Escola Superior de Biotecnologia, 4169-005, Porto, Portugal
[4] Institute of Food Science and Research (CIAL), Universidad Autónoma de Madrid (CEI UAM + CSIC), 28049 Madrid, Spain

The valorisation of agri-food by-products represents a sustainable strategy for the development of functional ingredients. Walnut oilcake (WOC) and walnut oil dregs (WOD), respectively obtained after cold-press extraction and from the decantation residues of the oil following cold pressing, were evaluated for their nutritional composition, phenolic profile, and in vitro bioaccessibility and bioavailability. The standardized INFOGEST 2.0 protocol was applied to simulate human gastrointestinal digestion, enabling the separation of bioaccessible and non-bioaccessible fractions. Phenolic compounds were quantified by HPLC-DAD-ESI/MS, revealing glansreginin A and ellagic acid as major constituents. After digestion, WOD displayed significantly higher total polyphenol bioaccessibility (78%) compared to WOC (15%). Ellagic acid presented the highest bioaccessibility in both matrices (>100%), while dicarboxylic acid derivative showed limited release. Antioxidant assays demonstrated a marked improvement in activity after in vitro digestion, with EC_{50} values decreasing from 4.2 ± 0.6 to 2.6 ± 0.8 mg/mL (TBARS) and from 2.5 ± 0.2 to 0.4 ± 0.1 mg/mL (DPPH) for WOD, and from 2.6 ± 0.2 to 0.4 ± 0.1 mg/mL (TBARS) and from 0.9 ± 0.1 to 0.4 ± 0.1 mg/mL (DPPH) for WOC, indicating enhanced compound release during in vitro digestion.

Caco-2 cell transport assays were used to evaluate intestinal absorption. Glansreginin A, a dicarboxylic acid derivative, was detected only in the apical compartment, with no phenolics found in the basolateral fraction, indicating limited trans-epithelial transport and suggesting that the health benefits of these compounds may be mediated by colonic metabolism rather than direct absorption. The non-bioaccessible fractions showed strong prebiotic activity, stimulating the growth of three *Lactobacillus* and one *Bifidobacterium* strains, surpassing, in some cases, the positive control fructooligosaccharides.

These findings highlight the relevance of combining in vitro digestion and cell culture models to assess the fate of polyphenols from food by-products. This study supports the potential of WOC and WOD as sources of bioactive and prebiotic compounds, reinforcing their application in functional foods and sustainable food systems.