Title: Unravelling Carotenoid Digestion: Insights into Bioaccessibility and Functional Properties

Authors: Helena R. Rocha, Rui Morais, Manuela E. Pintado; Ana M. Gomes, Marta C. Coelho

Keywords: natural pigments; gastrointestinal digestion; bioaccessibility; metabolites.

Abstract: Carotenoids, vital lipid-soluble compounds essential for human health, face bioaccessibility challenges during digestion due to resistance and degradation within protein complexes and plant cell walls. This complexity is influenced by factors like dietary sources, seasonal variations, food composition, matrix structure, lipid presence, dosage, and absorption rate. Understanding the delicate interplay of these elements is crucial for unlocking the full potential of carotenoids in promoting human health.

To explore the impact of digestion on carotenoid stability and functionality, beta(β)-carotene, lutein, lycopene, a mixture of these three carotenoids, and *Osmundea pinnatifida* underwent simulated gastrointestinal digestion, using the INFOGEST methodology, and absorption (dialysis membrane with 3 kDa) and were evaluated in terms of functional properties. The results revealed distinct transformations during *in vitro* gastrointestinal simulation, with the generation of carotenoids different from the initial sample (e.g., β-cryptoxanthin), emphasizing the intricate changes carotenoids undergo. Recovery indexes highlighted the challenge of retrieving carotenoids during digestion ($< 0.5\%$), emphasizing the complexity of their fate in the digestive process. The absence of detected or identified carotenoids in *O. pinnatifida* by HPLC analysis suggests that, within a complex matrix like algae, the bioaccessibility of carotenoids may be significantly compromised, requiring extraction methods to release these pigments and isolate them effectively. UPLC-qTOF MS analysis provided detailed fragment patterns, revealing variable relationships among fragments across different gastrointestinal phases.
Functional property assessment showcased notable antioxidant (17.5 μM TE) and antidiabetic (7.6 – 97 % inhibition) activities in the tested carotenoid solutions. The Alga and the β-carotene groups displayed the highest values in absorbed fractions, revealing their effectiveness. Additionally, all carotenoid samples exhibited antimutagenic effects regardless of concentration, with no observed cytotoxicity except at higher concentrations. This study provides valuable insights for optimizing carotenoid utilization and realizing their multifaceted health benefits. Despite bioaccessibility challenges, understanding these complexities contributes to unlocking the full potential of carotenoids for human health.