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Unravelling Nanoemulsions *vs* Excipient Nanoemulsions: formulation optimization, particle characterization and behavior as α-tocopherol delivery systems

FERNANDES[1], J.M.; ARAÚJO, J.F.; GONÇALVES, R.F.S.; VICENTE, A.A.; PINHEIRO, A.C.

[1]Centre of Biological Engineering, University of Minho – Braga, Portugal, jmichel@ceb.uminho.pt

The application of bio-based nanosystems for development and fortification of functional food products has been regarded as an excellent approach to improve foods' functional and sensorial characteristics, whilst enhancing the stability, bioaccessibility and bioavailability of bioactive compounds. Oil-in-water nanoemulsions (NE) have been extensively used for the encapsulation of lipophilic bioactive compounds and posterior incorporation into food matrices to obtain functional foods. Conversely, novel excipient oil-in-water nanoemulsions (Exc-NE) aim at improving the bioavailability of foods' natural bioactive compounds upon co-ingestion with nutrient rich foods. In this sense, the aim of this work was to produce NE and Exc-NE and compare their stability and functionality as delivery systems for α-tocopherol. Nanoemulsions' formulations were produced with the same ingredients (i.e., corn oil, lecithin and water), and an experimental design (i.e., 23 plus 3 central points) was established to optimize ingredients' composition (i.e., concentration of surfactant and oil-water ratio) and compare high-energy methodologies (i.e., Ultra-Turrax (UT) and homogenization (HPH)). For NE production, α-tocopherol was dissolved in the organic phase prior processing. Formulations composed of 3 % lecithin and 5 % oil produced smaller particles with lower polydispersity (PDI) regardless of nanoemulsion type and processing methodology (i.e., NE exhibited sizes of 253.5 nm and 242.5 nm when processed in HPH and UT, respectively; Exc-NE exhibited sizes of 260.5 nm and 241 nm when processed in HPH and UT, respectively). Longer processing times originated better particle dispersity in either methodology (i.e., nanoparticles presented PDI < 0.23 and PDI < 0.29, when processed in HPH and UT, respectively). Moreover, nanoparticles remained stable up to 60 days presenting modest size and PDI variations (i.e., NE size changed < 6 % and PDI changed < 17 %, whereas Exc-NE size changed < 10 % and PDI changed <

13 %). Optimized nanoemulsions' functionality as delivery system for α-tocopherol was evaluated using an *in vitro* gastrointestinal digestion protocol (INFOGEST 2.0). NE were submitted to digestion as produced, whereas Exc-NE were mixed with α-tocopherol prior to digestion, to assess their influence on α-tocopherol stability throughout the gastrointestinal tract, and its bioaccessibility and bioavailability. α-Tocopherol stability during simulated digestion was superior in NE regardless the processing methodology (i.e., NE stability < 16 %, Exc-NE stability < 9 %), indicating that NE offer greater protection against the digestive environment. On the other hand, nanoemulsions processed in HPH attained higher α-tocopherol bioaccessibility (i.e., bioaccessibility when processed in HPH was < 94 % compared to < 87 % when processed in UT), suggesting that nanoemulsions formed in the HPH were better adapted to form mixed micelles. Overall, the calculated bioavailability (i.e., estimation of α-tocopherol absorption) was superior in NE (i.e., ca. 13 % and ca. 7 % for NE and Exc-NE, respectively). In conclusion, NE were more efficient vehicles for the selected bioactive compound, however, the good results obtained with Exc-NE imply that excipient nanoemulsions have a great potential for applications on foods to improve their natural bioactive compounds' bioavailability without the need of further processing.

Keywords: Nanoemulsions; Excipient nanoemulsion; Delivery systems

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