Building dual-purpose ingredient by covalently conjugating quercetin to WPI

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The development of dual-functional ingredients has attracted expressive attention food, nutraceutical and pharmaceutical industries. To meet this demand, quercetin (QUE)-whey protein isolate (WPI) conjugates by free-radical grafting technique (pH4) and alkaline reaction method at different pH (pH 9, 10, and 11) were produced aiming to produce antioxidant-emulsifier compounds. All treatments utilized the same content of WPI (5 g/100 g of water) and QUE (0.03 mmol). WPI-QUE conjugates and native WPI were characterized by Fourier transformed infrared spectroscopy (FTIR), differential scanning calorimetry (DSC), Folin-Ciocalteu reducing capacity (FCRC), and antioxidant capacity (DPPH and FRAP). WPI-QUE conjugates produced at pH 4, 9, 10 and 11 were named QpH4, QpH9, QpH10 and QpH11, respectively. To test their emulsifying capacity, oil-in-water (O/W) nanoemulsions were prepared by incorporating pomegranate seed oil (10% w/w) into an aqueous solution of conjugates, or native WPI (1% w/w) using a rotor-stator (T18, IKA) for 5 min at 14,000 rpm followed by homogenization in a microfluidizer (M-110Y, Microfluidics Co.) at 100 MPa, for three cycles. The O/W nanoemulsions were evaluated by droplet size and instability Index (iI). All conjugation conditions produced compounds with improved antioxidant activity; however, the method and reaction pH affected the conjugate characteristics. QpH4 and QpH9 showed lower solubility, larger particle size and higher antioxidant activity probably because there was a higher attachment of QUE to WPI promoting the dimerization of WPI. The conjugation with QUE altered the secondary and tertiary structure of native WPI, according to the results from FTIR and DSC analysis. These conformational changes facilitated the anchorage of the conjugates onto the interface. WPI-QUE conjugate-stabilized O/W nanoemulsions, except QpH4, showed smaller droplet sizes than that produced with native WPI. QpH4-nanoemulsions had larger diameter (~226 nm) because its pH was close to isoelectric point of WPI. Even though they had different droplet sizes, nanoemulsions stabilized by conjugates, except QpH4, and native WPI had similar instability Index (~0.35). In summary, the results showed that the conjugation by alkaline method at pH 9 was an efficient technique to produce dual-functional compounds, with improved antioxidant and emulsifying potential.