FTIR analysis in assessment of complex coacervation of pumpkin leaf proteins and natural polysaccharides

Abstract

Complex coacervation using plant proteins and polysaccharides is atractive field of science nowadays. The application of plant proteins instead of animal proteins in complex coacervation enabled overcoming of some drawbacks linked with the production of animal proteins and its environmental impact, the spread of diseases, pathogens etc. The sustainable research based on extraction of waste leaf biomass as a rich source of plant proteins can contribute to wider usage of plant proteins in coacervation. The aim of this work protein extraction from pumpkin leaves, using different procedures: thermal-acidic (TA), alkaline-acidic (AA) and ammonium-sulfate (AS) precipitation. "Green fraction" was precipitated by heating pumpkin leaf juice to 50 o C for 30 min (TA and AS), or by adjusting pH to 10 (AA). "White protein fraction" was then precipitated from the supernatant by adjusting pH to 4 (TA and AA) or by addition of ammonium-sulfate (50% w/v)(AS) and freeze-dried. The samples were subjected to electrophoresis to determine the presence of RuBisCO. Further, the isolates were used for preparation of complex coacervates in interaction with alginate. The formation of complex coacetvates (TA-A, AA-A and AS-A) in colloid solution was detected by changes in optical density in the pH range 7.00-1.50, at 600 nm. Prepared samples were then freeze-dried and examined using FTIR. Electrophoresis confirmed the presence of RuBisCO in TA and AS, but all three protein-enriched fractions had very similar FTIR spectra with most dominant bands found in the peptide carbonyl region (1635 and 1515 cm-1); these bands were of the highest relative intensity in AS, indicating the highest protein content. FTIR spectrum of the alginate exhibited characteristic bands at 1725 and 1601 cm-1 , corresponding to carboxylic acid/carboxylate groups, respectively. Bands associated with sugar structures (C-O bonds), in the range between 1200 and 800 cm-1 were well developed, with a band peaking at 1030 cm-1 being the most dominant. The spectra of the coacervates resembled mostly those of the protein samples. The presence of alginate was confirmed in AA-A and AS-A, as in the FTIR spectra of these samples the band at 1030 cm-1 was more pronounced (higher intensity in AS-A). The results showed that, while all samples exhibited precipitation at pH4, complexion with alginate was observed only in AA and AS, and that AS was capable of binding greater amount of alginate. Therefore, the method of protein isolation is of great importance for its ability to form coacervates.