Molecular characteristics of chickpea protein fractions: Hydrophobicity and structural analysis using Raman spectroscopy

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Chickpea protein is a mixture of protein consisting of several fractions, including salt-soluble globulin, water-soluble albumin, and dilute acid/alkali-soluble glutelin. Chickpea protein isolates (CPI) contain a high fraction of globulins, representing 80% of the total seed proteins. Globulin can be further subdivided into 11S legumin (CL) and 7S vicilin (CV) subunits. These fractions are responsible for most functional properties of CPI, but the molecular characteristics are not fully understood. This study is aimed to compare the hydrophobicity and structural properties of isolated globulin, legumin, and vicilin fractions from chickpea. Alkaline extraction-isoelectric precipitation and modified salt dissolution-precipitation method were performed to produce CPI, CL, and CV fractions. The samples were characterized by surface hydrophobicity (fluoresce method), sulfhydryl group content (Ellman's method), and electrophoresis (SDS-PAGE). In addition, both hydrophobicity and structural properties were characterized using Raman spectroscopy. The protein content in globulin, legumin and vicilin fractions reached over 91%. The results showed that the fractions had differences in their molecular characteristics. Vicilin fraction had significantly ($p<0.05$) lower surface hydrophobicity, indicating less exposed hydrophobic residues or patches; lower free and total SH groups, and disulfide bond content than legumin and globulin. According to the results of SDS-PAGE, the vicilin fractions showed subunit bands with lower MW, ranging from 50 to 10 kDa, implying heterogenous polypeptide subunits. Interestingly, Raman spectroscopy analysis exhibited a high content of both ?-helix (~46%) and total ?-sheet (~38%) secondary structure in CPI. The highest percentage of ?-sheet was found in the vicilin rather than in the legumin fraction. Results that could be interesting to study and understand its impact on digestibility. The lower hydrophobicity and disulfide bond in vicilin was detected in Raman spectra, and an increase in the intensity ratio I1360/I1340 was identified indicating a buriedness of the tryptophan residues. The results confirm that the molecular properties of vicilin fraction play an important role in influencing the structural characteristics of CPI. The characterization of hydrophobicity and structural properties of chickpea protein fractions provided a certain theoretical basis for understanding their interaction behavior and functional characteristics and the potential application of these legume proteins for the development of products