



## P.03 Structure and evolution of high-density protein systems

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This project aims to examine how whey proteins typically behave at the sub-molecular level under highly concentrated conditions where competition exists between the molecules for hydration-water. Freeze-dried bovine-derived beta-lactoglobulin (BLG, type-A) whey protein powder is dissolved at varying protein solution concentrations in two different aqueous buffer systems (0.2M Na<sub>2</sub>HPO<sub>4</sub> and 0.1M Citric Acid, and Phosphate buffer). These protein solutions are buffered at pHs above and below the isoelectric point pI (pH ~ 5.1). Infrared (ATR FTIR) and circular dichroism (CD) spectroscopic techniques are used to study the micro-structural arrangements of the protein when solutions of proteins are condensed towards the intended levels for high-protein foods. Changes in the protein secondary structure with respect to increased concentration and the possible reversibility of any molecular structural change via dilution with buffer was examined. ATR FTIR found significant secondary structure change occurring between 10mg/ml and 50mg/ml protein concentration with very little change occurring for concentrations higher than 50mg/ml. This may be explained by the structure evolving from a less dense to a denser more compact inter-molecular  $\beta$ -sheet formation. Between 10mg/ml and 50mg/ml there was noticeable loss in  $\alpha$ -helix structure signal and a simultaneous gain in random coil signal which may be interrelated (i.e. one structure replacing the other with increasing concentration). This is the concentration range over which the vast majority of secondary structure change seems to happen with little change occurring beyond 50mg/ml. These changes were verified by the CD method with the additional suggestion of aggregates occurring with increasing concentration.