523d Investigation of Protein Adsorption and Similarity Measurement for Protein Binding Behavior in Hydrophobic Interaction Chromatography (Hic) System

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Hydrophobic Interaction Chromatography (HIC) is widely employed for protein purification. In this paper, the adsorption of proteins in HIC systems was investigated using a variety of experimental and theoretical techniques. The adsorption isotherms of a wide range of proteins under various resin and salt conditions (0.5 M to 1.5 M ammonium sulfate) were determined using high throughput batch experiments. Various classes of proteins exhibited qualitatively different adsorption behavior. In order to gain insight into these classifications, novel molecular descriptors were developed which include both property and surface distribution effects. These descriptors were then employed to study the "similarities" of the different classes of proteins. In addition, classification software was employed to determine the key molecular descriptors associated with various types of adsorption behavior. This approach was carried out to evaluate both linear and non-linear adsorption behavior. Finally, Raman and circular dichroism (CD) experiments were carried out to examine protein conformational and/or orientation changes associated with the adsorption of different classes of proteins in HIC systems.