

430f The Potential for Exploiting Surface Hydrophobicity Differences for Recovery of Recombinant Proteins from Corn Using Aqueous Two-Phase Partitioning

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Corn is a potential host for large scale production of recombinant proteins . Aqueous two phase (ATP) partitioning scales well and offers opportunities for integration of extraction and recovery. ATP partitioning targeted at hydrophobicity differences for separating foreign proteins from corn was assessed. Six model proteins: chymotrypsin A, cytochrome c, ribonuclease A, lysozyme, α -lactalbumin and bovine serum albumin were partitioned in polyethylene glycol (PEG) – Na₂SO₄ systems to study the controlling factor for ATP partitioning. A series of such APS containing from 0 – 7% NaCl was used. The MW and pI of the model proteins were obtained from protein databank. Each proteins' surface hydrophobicity (SH) was estimated from literature measures based on hydrophobic interaction chromatography (HIC), reverse phase chromatography (RPC), (NH₄)₂SO₄ precipitation and molecular simulation. Partition coefficients in APS containing above 2% NaCl correlated with SH but not MW or pI and the best correlation was obtained between log (K) and SH calculated from (NH₄)₂SO₄ precipitation data. Log (K) also correlated well with SH determined by HIC if lysozyme was removed from the data set. Based on the correlations between log (K) of the model proteins and their SH from either HIC or (NH₄)₂SO₄ precipitation, the net average SH of aqueous extractable corn endosperm proteins and corn germ proteins were determined by their partitioning in NaCl added PEG – Na₂SO₄ systems. The net average SH of corn endosperm protein is near that of α -lactalbumin and higher than corn germ protein whose net average SH is near that of ribonuclease A. This result establishes the hydrophobicity of a recombinant protein that would permit recovery by partitioning for each fraction in which the protein might be expressed.