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Pseudobioaffinity ligands coupled to high throughput support matrix for purification of proteins from preparative to analytical validation important aspect in DSP of biopharma

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The term "Pseudobiospecific affinity chromatography" was coined by Prof. M A Vijayalakshmi in 1989 (TIBTECH 1989), which cover classes of ligands which have both structural and functional recognition of proteins based on their aminoacid sequence and three dimensional structure. Pseudobiospecific AC systems (e.g. amino acids, metal-chelates and triazine dyes) are highly economic and robust, can be fine-tuned to excellent specificities and medium dissociation constants (10-7 - 10-5). Ligand and Matrix are the two chromatography components that guide molecular interactions in any AC system. The ligand governs thermodynamic aspect of the chromatographic system which includes binding specificity, binding strength, ligand concentration etc. The support matrix governs the high-throughput-hydrodynamic aspect which includes the porosity of the support, particle size, etc. "MONOLITHS" are new stationary phase materials introduced in 1990's as "non-particulate homogeneous methacrylate material with high pore interconnectivity and lack of interestial voids" containing mega pores. They can be prepared in different forms like disks (CIM®: Convective Interactive Media), radio flow columns, capillaries and microfluidics. Due to the high pore interconnectivity, the flow is convective which results in efficient mass transfer of molecules and without any diffusion limitation like in the agarose based system. Thus a flow independent binding system gives very high capacity and binding, even at very high flow rates like 5 column volumes per minute. The CIM systems are hydrophilic and versatile such that any ligand can be coupled as is being done with agarose matrices and with same chromatographic buffer systems. Chromatographic runs are done seconds to minutes not in hours and days. Apart from these, monoliths possess other advantages like ease of preparation, low dead volumes, chemical and mechanical stability and compatibility to get hyphenated with conventional chromatography equipment's.

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