

Chromatography in the biotech industry: Purification of biomolecules.

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Chromatography is a separation technique that has revolutionized the way scientists purify and analyze complex mixtures. In the biotech industry, chromatography plays a critical role in the purification of biomolecules such as proteins, nucleic acids, and carbohydrates. The purification of biomolecules is an essential step in the development of biopharmaceuticals, gene therapy, and other biotech products. In this article, we will explore how chromatography is used in the biotech industry for the purification of biomolecules. Proteins are the most common biomolecules that require purification in the biotech industry. Proteins are large, complex molecules that are involved in various biological processes such as catalysis, signaling, and transport. Purified proteins are used in the development of biopharmaceuticals such as vaccines, antibodies, and enzymes. Chromatography is the preferred method for protein purification because it offers high resolution, scalability, and reproducibility [1].

There are different types of chromatography techniques that can be used for protein purification, including size exclusion chromatography, ion exchange chromatography, hydrophobic interaction chromatography, and affinity chromatography. Each technique relies on different principles of separation, such as size, charge, hydrophobicity, or specific binding. Size exclusion chromatography (SEC) separates proteins based on their size and shape. Larger proteins elute first, followed by smaller ones. SEC is useful for removing impurities such as aggregates, fragments, or smaller proteins that might interfere with the function or stability of the target protein. SEC can also be used for the analysis of protein oligomerization or conformational changes [2].

Ion exchange chromatography (IEC) separates proteins based on their charge. Proteins can be positively or negatively charged depending on their isoelectric point (pI) and the pH of the solution. In IEC, proteins are adsorbed to a stationary phase that carries an opposite charge, either positively or negatively. The adsorbed proteins are then eluted by changing the pH or ionic strength of the eluent. IEC is useful for the separation of proteins with different pI or for the removal of contaminants such as endotoxins or DNA. Hydrophobic interaction chromatography (HIC) separates proteins based on their hydrophobicity. Proteins have different surface properties that can affect their solubility in aqueous solutions. HIC relies

on the interaction between the hydrophobic regions of the protein and a hydrophobic stationary phase, such as a resin or a membrane. The more hydrophobic proteins are adsorbed first, while the less hydrophobic ones are eluted later. HIC can be used for the separation of proteins with similar charges or pI, or for the removal of host cell proteins or aggregates [3].

Affinity chromatography (AC) separates proteins based on their specific binding to a ligand that is immobilized on a stationary phase. The ligand can be a protein, a peptide, a nucleic acid, a carbohydrate, or a small molecule that interacts with the target protein in a selective and reversible manner. AC can achieve high levels of purity and specificity, but it requires the design and production of a suitable ligand, which can be expensive and time-consuming. AC can also be used for the isolation of protein complexes, protein-protein interactions, or protein-DNA interactions. Chromatography can be performed using different formats, such as column chromatography, batch chromatography, or membrane chromatography. Column chromatography is the most common format, where the stationary phase is packed into a column and the eluent is passed through the column under pressure [4,5].

References

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