

Applications of Immuno Affinity Chromatography in Various Scientific and Medical Fields

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DESCRIPTION

Immunoaffinity chromatography is a powerful and highly specific separation technique used in biochemistry and biotechnology to purify and isolate target molecules, such as proteins or antibodies, from complex biological samples. This method capitalizes on the unique binding properties of antibodies, which can specifically recognize and bind to their corresponding antigens with high affinity and selectivity. The process of immune affinity chromatography involves immobilizing an antibody or an antigen (depending on the target molecule) onto a solid support matrix, typically a resin or a bead. The target molecule, which is present in the sample mixture, is then allowed to interact with the immobilized antibody/antigen, leading to the formation of an antibody-antigen complex. The rest of the sample components are washed away, leaving the target molecule specifically bound to the immobilized antibody/antigen. One of the key advantages of immunoaffinity chromatography is its exceptional specificity. The interaction between the antibody and the target molecule is highly specific, allowing for the isolation and purification of the desired molecule even from very complex samples containing numerous other components.

This specificity is crucial for applications where high purity and selectivity are essential, such as in the isolation of therapeutic proteins or the detection of specific biomarkers in clinical samples. For example, it is often employed as an initial capture step to enrich the target molecule before subjecting the eluted fraction to additional purification steps using other chromatographic methods like size-exclusion chromatography or ion exchange chromatography. This combination of techniques enables researchers to achieve a higher degree of purification and resolve challenging purification problems. Despite its strengths, immune affinity chromatography also has some limitations. The production and immobilization of antibodies or antigens on the support matrix can be time-consuming and expensive, making the technique less practical for some applications. Additionally, harsh elution conditions may be required to dissociate the

antibody-antigen complex and recover the target molecule fully, potentially impacting the activity or integrity of the purified molecule.

Depending on the type of biomolecule and the specific analysis required, this can involve various techniques such as protein extraction, digestion (e.g., enzymatic cleavage of proteins into peptides), derivatization (e.g., adding chemical tags), or direct analysis for smaller molecules like metabolites. In mass spectrometry, the biomolecules are converted into ions because mass spectrometers can measure the Mass-to-Charge ratio (m/z) of ions effectively. There are different ionization techniques for different biomolecules, such as Electrospray Ionization (ESI) and Matrix-Assisted Laser Desorption/Ionization (MALDI) for proteins and peptides, and Electron Ionization (EI) or Chemical Ionization (CI) for small molecules.

The ionized biomolecules are then introduced into the mass analyzer. There are various types of mass analyzers, including Time-Of-Flight (TOF), quadrupole, ion trap, and Orbitrap. Each mass analyzer has its advantages and is suitable for specific applications. Once the ions are separated by the mass analyzer, they are detected by a detector, typically an electron multiplier or a microchannel plate detector. The detector records the number of ions detected at each m/z value, generating a mass spectrum. Data analysis software is used to interpret the spectrum, identify the biomolecules based on their mass and charge, and quantify their relative abundances.

CONCLUSION

In conclusion, immunoaffinity chromatography is a valuable tool in the field of molecular separation and purification. Its exceptional specificity and compatibility with other chromatographic methods make it an indispensable technique for researchers and industries involved in biotechnology, pharmaceuticals, and clinical diagnostics. As technology advances and the cost of antibody production decreases, immunoaffinity chromatography is likely to become even more prevalent and impactful in various scientific and medical fields.

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Received: 19-Jun-2023; Manuscript No. JCGST-23-26040; **Editor assigned:** 21-Jun-2023; Pre-QC No. JCGST-23-26040 (PQ); **Reviewed:** 11-Jul-2023; QC No. JCGST-23-26040; **Revised:** 20-Jul-2023, Manuscript No. JCGST-23-26040 (R); **Published:** 28-July-2023, DOI: 10.35248/2157-7064.23.14.524

Citation: Berggren V (2023) Applications of Immuno Affinity Chromatography in Various Scientific and Medical Fields. J Chromatogr Sep Tech. 14:524.

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