

Chromatography for Chiral Separation

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ABOUT THE STUDY

The development of separation techniques for the analysis and resolution of chiral medicines and solutes has been a focus of pharmaceutical research for many years. The use of proteins as chiral binding agents in High-Performance Liquid Chromatography (HPLC) has gained special attention in such research. CSPs that use serum transport proteins (e.g., human serum albumin, bovine serum albumin, and alpha1-acid glycoprotein), enzymes (e.g., penicillin G acylase, cellobiohydrolases, and -chymotrypsin), or other types of proteins such as (e.g., ovomucoid, antibodies, and avidin or streptavidin) are examined.

The usage of molecular chirality may be traced back to Louis Pasteur's work with tartaric acid in the mid-1800's. In the late 1970's Yoshio Okamoto developed a helical polymer of triphenylmethyl methacrylate and utilised this material to construct chromatographic columns that could be used for chiral separation. Since then, much work has been directed into developing Chiral Stationary Phases (CSPs) for use in chromatography for the separation and study of chiral substances. A large number of pharmacological drugs are chiral compounds. Despite the fact that these chemicals are frequently offered as racemic combinations, approximately two-thirds of the medications now available on the market are chiral.

Even while the two enantiomers of a chemical will have much of the same physical qualities, how they interact with a biological system may change dramatically, potentially leading to varying degrees of activity or adverse effects. S-ibuprofen, for example, is almost 100 times more effective than R-ibuprofen in inhibiting cyclooxygenase 1 and 2, and R-thalidomide is a sedative whereas S-thalidomide is a teratogen. Furthermore, the S-enantiomers of barbiturates including pentobarbital, hexobarbital, and mephobarbital are sedatives/hypnotics, whilst the R-enantiomers are inert. These distinctions have necessitated the inclusion of pure enantiomers in pharmacological formulations and/or the investigation of the effects that the various enantiomers may have in the body. This, in turn, has increased the demand for analytical methodologies and preparative procedures for analysing and separating chiral chemicals in pharmaceutical samples.

Several approaches for chiral separation of pharmaceuticals and amino acids for chemical analysis have been developed. Gas chromatography, thin layer chromatography, High-Performance Liquid Chromatography (HPLC), supercritical fluid chromatography, and capillary electrophoresis have all been used. HPLC-based methods have aroused the curiosity of researchers. To create CSPs for HPLC, a variety of stereoselective binding agents have been utilised. The application of these compounds in a liquid chromatographic system is considered as the affinity chromatography or High-Performance Affinity Chromatography (HPAC). Proteins, polysaccharides, macrocyclic antibiotics, and ion-exchange ligands are examples of such binding agents.

Many proteins have the ability to distinguish between different versions of a chiral chemical. This is due to the fact that proteins are chiral in nature, both in terms of their core structure (being formed of L-amino acids) and their secondary or higher order structures. Proteins ranging from enzymes to serum transport proteins and glycoproteins have been immobilised and utilised to create CSPs for HPLC. These separations are carried out based on variations in the binding strength and number or quantity of binding sites for chiral chemicals with the protein. The capability to deal with a wide range of chiral compounds, the ability to employ aqueous mobile phases, and the ability to immediately analyse most chiral compounds without the requirement for derivation are all advantages of utilising protein-based CSPs.

CONCLUSION

Protein-based CSPs, on the other hand, might have a poor loading capacity, be costly to manufacture, and have limited stability, making them more suitable for analytical work than preparative separations. Separation and analysis of chiral pharmaceuticals has become more significant in the discovery and development of novel pharmacological medications. Chromatographic approaches that use proteins as chiral selectors have recently been shown to be effective tools for separating chiral medicines or solutes and studying their interactions with proteins.

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