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SOLID-BASED EXTRACTION APPROACHES IN SAMPLE PREPARATION FOR BIOANALYSIS

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DESCRIPTION

Bioanalysis concerns the analysis of xenobiotics (similar as medicines) and endogenous compounds (similar as biomarkers) in biological systems. Both xenobiotics and biomarkers encompass low molecular weight (LMW) and High Molecular Weight (HMW) analytes. According to the analytical purpose, the sample can follow two distinct types of methodologies untargeted and targeted analysis. The former approach (untargeted) is substantially applied to acquire a most of information from the sample to segregate inter-or intra-individual variations and identify potential biomarkers. After validation of a new biomarker, the aim is generally towards quantitation with a maximum of perceptivity, selectivity, and repetition, as needed around targeted analysis. Anyhow of the type of analysis, a typical bioanalytical workflow comprises sampling and sample preparation previous to analysis, data treatment, and biological interpretation. Sampling and sample medication are the first stages of the bioanalytical process. Sampling comprises collection, handling, delivery, processing, and storehouse of the biological sample previous to sample medication. Sample medication is used to clean-up and/ or enrich the sample before analysis to enhance its discovery without fouling the analytical device. According to the nature, accessibility, volume, and stability of the natural fluid, as well as the thing of the analysis (targeted or untargeted) and other constraints (throughput, cost), different sample medication should be used.

Solid Phase Extraction (SPE) is the most generally used solid- grounded sample preparation technique in bioanalysis. SPE is based on the analyte commerce between a stationary phase and the sample. A large variety of stationary phases (reversed phase, normal phase, ion exchange) and multitudinous formats, from many milligrams to micrograms, are available. Advancements have been made towards miniaturization and numerous solid- based ME exist. Among them, solid phase microextraction consists of a stationary phase grafted on a probe. As for SPE, a large variety of stationary phases and probe dimensions are available. Alternatives to the probe are also used, with syringe needles for example, as found in microextraction by packed sorbent. The principal means of these ways are (i) application to unpredictable compounds by head space extraction, (ii) ready hyphenation with GC, and (iii) probe recycling by thermal desorption. Lately, reports have been made using SPME for *in vivo* sampling SPME probe was surgically inserted in tissues and organs enabling a rapid monitoring of a treated area without tissue lesions.

Other interesting SPE formats have been developed. The first one is the zip-tip format, in which the stationary phase is contained in the end of a 10 μL pipette tip. It has been particularly used in RP mode in the proteomics field for peptide desalting and enrichment. With Disposable Pipette Extraction (DPX), pipette tips incorporate approximately contained sorbent material, which is mixed with the sample result. Turbulent air bubble mixing creates a suspension of sorbent in the sample ensuring optimal contact and effective extraction.

In order to ameliorate the mass transfer and the extraction speed, Stir Bar Sorptive Extraction (SBSE) and Fabric Phase Sorptive Extraction (FPSE) have been developed. In SBSE, the stationary phase is grafted on the surface of a magnetic bar, which is strengthened in the sample, whereas in FPSE the stationary phase is bonded to a permeable fabric (cotton, paper). The main advantage of these approaches is the enhancement of the contact surface between the sample and the stationary phase, leading to high recovery, high enrichment, in a minimum time, and smaller extraction steps. In the proteomics field, the HMW compounds of interest can be at a very

low abundance compared to high abundant proteins. Extraction of the former can hardly be achieved by conventional SPE procedure due to their close behavior compared to the latter. Reduction approaches have therefore been developed to allow for minor HMW compounds expression. They're grounded on picky interactions between the proteins and arbitrary hexapeptides grafted on beads used as ligand theoretically each hexapeptide bind to a unique protein. Because of the blob capacity lading limits, major proteins will quickly saturate their ligand and the excess will be washed. On the other hand, minor proteins will be concentrated on globules previous to elution of all the retained proteins. Grounded on an analogous medium (antibody recognition) immunodepletion, allows suppressing major adulterant proteins from a sample. Immunodepletion columns are commercially available in microcartridge format and can deplete up to 22 major proteins from the plasma.