

A Robust Online Extraction Approach for Bioanalysis

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High-performance liquid chromatography with tandem mass spectrometry (LC-MS/MS) has been widely utilized for the bioanalysis due to its selectivity and excellent sensitivity. An accurate and reliable LC-MS/MS assay, however, requires an effective sample cleanup procedure. The high turbulence liquid chromatography (HTLC) technology significantly reduces the time required for sample cleanup since sample extraction and analysis are performed online. Using HTLC/LC and tandem mass spectrometry (MS/MS), robust, reliable and automated methods were developed for direct injection and quantitation of biological samples, such as plasma, serum, urine and dialysate. In this approach, the dual column configuration (both focus and quick elute mode) was used to allow chromato-focusing and improve the sensitivity and selectivity of assays; an Aria TX2 parallel HTLC/LC-MS/MS on-line extraction/separation system was validated to double the throughput of a mass spectrometer. Additionally, a novel method was developed to increase the assay robustness and extend the extraction column lifetime to over 2000 human plasma samples.

Using this approach, over 300 clinical samples were prepared and analyzed a day. These robust, selective and precise methods ensured the timely analysis of all clinical samples for several projects, and they have several major advantages over conventional LC-MS/MS methods: 1) simple sample pretreatment, 2) easy to automate, 3) robust procedures, and 4) improved sensitivity and selectivity as a result of narrow peak width and low noise levels.