PARTICLE CHARACTERIZATION BY MULTI-DETECTOR HDC

Andre M. Striegel
National Institute of Standards & Technology (NIST)
andre.striegel@nist.gov

Hydrodynamic chromatography (HDC) is a liquid chromatographic technique that separates analytes on the basis of their size in solution [1]. HDC can be performed in an open tube (capillary) or in a packed column. In the latter case, the column packing material should be inert and either non-porous or of a pore size substantially smaller than the solution size of the analyte. Microcapillary HDC is currently employed chiefly for the separation of DNA and DNA fragments [2], while packed-column HDC with small-diameter (sub-2 µm) packing particles has been used, in ultra-high-pressure mode, for the separation of narrow dispersity polystyrene standards with molar mass M as high as 3 MDa [3]. Meanwhile, the use of packed-column HDC using large-diameter (15–20 µm) packing particles for the characterization of ultra-high-M polymers and particles has seen a resurgence in recent years, aided chiefly by the coupling of the HDC columns to a multiplicity of physical detection methods [4, 5]. The latter technique will be the topic of this tutorial.

The “holy grail” of particle sizing can be regarded as the accurate and precise determination of the particle size averages and distribution of a sample that is disperse in size, shape, structure, and chemistry, along with the determination of the averages, distributions, and mutual-interdependences of all these properties. While no single analytical technique will be able to provide all of these for every sample type, multi-detector HDC has demonstrated the ability to measure a number of these properties for a variety of samples. Advantages of SEC, vis-à-vis size-exclusion chromatography or field-flow fractionation, include its being gentler than the former (thus minimizing the possibility of on-column, flow-induced analyte degradation) and more affordable and easily implementable than the latter. Additionally, for broad dispersity samples, appropriate choice of column packing material particle size and pore size presents the possibility of combining multiple separation modes into a single analysis, via a combined HDC–SEC separation [6].

This tutorial will focus on how to design a packed-column HDC experiment for the purposes of characterizing particle size, shape, molar mass, and structure, and the type of information that can be obtained by combining detection methods such as refractometry, viscometry, and light scattering (both dynamic and static). Examples from the recent literature will be used to showcase the power, versatility, and limitations of the technique.