

# Hydrophilic Interaction Liquid Chromatography (HILIC) and Advanced Applications

*"Having a printed book in one's hands is always an opportunity for the imagination, and if the book is pleasant and very useful, it's the best of these."*

Since Alpert [1] coined the term hydrophilic interaction liquid chromatography (HILIC) to define the highly efficient chromatographic separation of very polar molecules that takes place through the interaction between an aqueous organic mobile phase and a polar stationary phase, the technique has been fine tuned, and accounts of many of its applications have been published.

In this information field, I commend the excellent effort of Perry G Wang (Research Chemist, Office of Regulatory Science, US FDA) and Weixuan He (Associated Director, Product and Process Development, at Meda Pharmaceuticals), who compiled and edited the state-of-the-art in their book *Hydrophilic Interaction Liquid Chromatography (HILIC) and Advanced Applications*, published by CRC Press (Taylor & Francis Group, Boca Raton, FL, USA, 2011).

This is a well-organized piece that takes us through 23 chapters ranging from the most basic concepts up to the very specific applications developed for recognized scientific teams who make contributions to the book.

Chapters 1, 2, 3, 11 and 17 offer the basic concepts for understanding HILIC, comparing diverse aqueous normal-phase chromatographies with true HILIC, explaining the relationship between  $\log k'$  and percentage organic volume in the mobile phase, using the calculation of  $\log D$  for choosing internal standards during method development, giving a point of view on ion-exchange chromatography as well as HILIC, exposing the effects of buffer concentration and temperature on HILIC performance, and describing the retention and selectivity of several polar phases.

Although the book is not divided into sections, its organization allows us to realize that chapters are grouped by similar items. Chapters 4, 5 and 6 afford advanced applications in the food and safety field; the separation of proanthocyanidins and other plant polyphenols,

the use of HILIC for checking final quality, nutrients and the safety of diverse food products, and toxic analysis episodes during seafood consumption.

Material on environmental applications, included in Chapters 7 and 8, explores the search for pesticides and drugs in wastewater and soil.

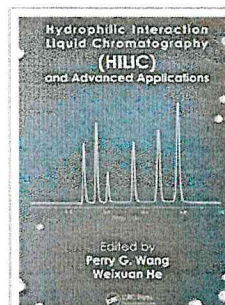
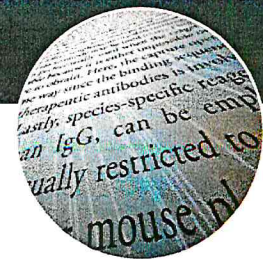
For all of those who love clinical applications, Chapters 9 and 10 delve into the hard work of biological sample processing previous to HILIC and depict the effort of scientists employing this type of chromatography for selecting and quantifying a huge variety of biomarkers for several diseases. Chapter 18 introduces us to the cumbersome analysis of very polar drugs for pharmacokinetic purposes.

In Chapters 12 through 16, five research groups describe their works in the field of pharmaceutical applications. HILIC has become a solution for the analysis of polar impurities during R&D and for the characterization of very polar acid or basic drugs. On these pages, two examples of HILIC-UPLC are shared: analysis of ioversol and of hydrazine-based mildronate.

Two special chapters are included for biochemical applications (Chapters 19 and 21): the separation of very hydrophilic mono- and di-phosphorylated nucleotides, and the separation of carbohydrates.

Finally, and to avoid the idea that HILIC is solely for small molecules, Chapters 20, 22 and 23 are presented in the area of proteomics. HILIC has been utilized for the characterization of post-translational protein modifications such as glycosylation and phosphorylation.

The great amount of applications included that have coupled HILIC with MS is not surprising, and there are several physicochemical reasons for this. First, the high percentage of organic solvent – usually acetonitrile – used in the mobile phase decreases viscosity, with a consequent decrease in pressure. Thus, high flow-rates can be used, rendering shorter run



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