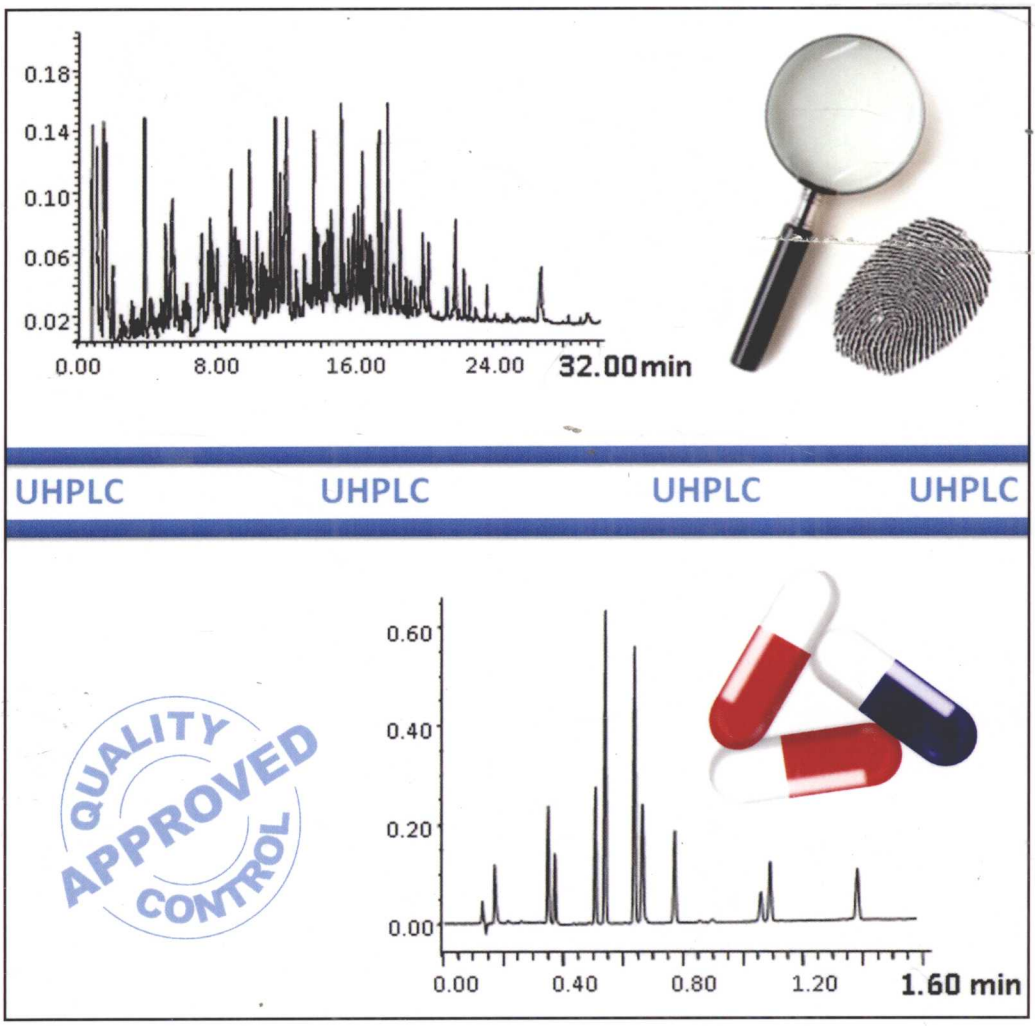
RSC Chromatography Monographs

Edited by Davy Guillarme and Jean-Luc Veuthey

UHPLC in Life Sciences



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UHPLC in Life Sciences

Edited by

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Preface

Since the initial development of modern liquid chromatography in the 1970s, this technique has received strong interest and has seen tremendous improvements in equipment (such as columns, stationary phases, pumping systems and injection valves). In addition, there has been a consistent trend towards analyses with either faster separations or enhanced resolution. During the last century, these goals were generally attained to the detriment of sensitivity, robustness and both system complexity and stability. In 2004, there was a breakthrough with the commercial introduction of columns packed with sub-2 μm fully porous particles and chromatographic systems able to withstand pressures of up to 1000 bar. Chromatography under such extreme conditions is generally described as UHPLC [ultra-high-pressure (or -performance) liquid chromatography]. Since 2004, more than 2500 papers have been published on this topic, including theoretical investigations of both the technique and its applications, from industrial, governmental and academic laboratories.

The aim of this book is to provide a single source of information with an extensive overview of UHPLC and its applications in the life sciences. The book is divided in two major sections: Chapters 1 to 7 are dedicated to the theoretical basis of UHPLC, and Chapters 8 to 14 describe its application to life science analyses.

Chapter 1 gives a general background of UHPLC and a theoretical comparison between it and other existing strategies for increasing the speed or resolution of the separation. In Chapter 2, the importance of suitable instrumentation and columns for UHPLC is critically discussed. Important aspects of transferring a method from HPLC to UHPLC conditions are discussed in Chapter 3 and include the rules used and certain inherent

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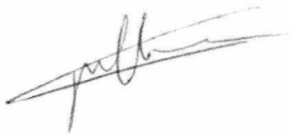
problems encountered. Chapter 4 explains the interest in UHPLC with mobile phase temperatures higher than the ambient temperature. Indeed, high temperatures provide some obvious advantages, such as reducing the mobile phase viscosity and polarity. In 2006, a new generation of columns that were packed with sub-3 μm superficially porous particles (also known as core-shell particles) was introduced to the market. This promising technology is described in detail in Chapter 5. A performance comparison between columns packed with sub-3 μm core-shell particles and sub-2 μm porous particles is presented in the same Chapter. Another recent trend in HPLC is the rapid development of hydrophilic liquid chromatography (HILIC) for the analysis of polar compounds and to provide an alternative selectivity for ionizable compounds. Because UHPLC-HILIC columns packed with porous, sub-2 μm particles are now commercially available, the advantages and limitations of the UHPLC-HILIC approach are described in Chapter 6. Finally, the last Chapter in the first section of the book is devoted to both the coupling of UHPLC to MS devices and the technological constraints that make this coupling slightly more difficult than traditional HPLC-MS. It is noteworthy that approximately 60% of the papers published on UHPLC deal with MS detection.

The second section of the book is dedicated to the application of UHPLC to life science analyses. During the first stages of drug discovery, the number of potential drug candidates is quite large. For this reason, it is important to develop high-throughput screening (HTS) methods for the rapid determination of physico-chemical properties (such as log *P*, solubility and permeability). Chapter 8 presents the attempts that were made over the last few years to achieve the HTS screening of drugs using UHPLC and UHPLC-MS technologies. The analysis of biological fluids is of prime importance for drug development, therapeutic drug monitoring and clinical/forensic/toxicological analyses. The number of biological samples that require rapid, quantitative analysis is generally large, and thus UHPLC has been widely employed in this field, which is discussed in Chapter 9. Chapter 10 provides more specific details for the doping control analysis of urine samples by UHPLC-MS and describes the well-accepted two-step strategy, which involves a screening step and a confirmation step. Seized drugs of abuse are generally in powder form, and the strategy for the rapid screening and profiling of these specific drugs is considered in Chapter 11. The need for the determination of drugs in environmental samples is shown in Chapter 12. In this case, a UHPLC-MS strategy was selected because of the combination of improved resolution from UHPLC and high selectivity and sensitivity from the MS devices. Other complex living organism of interest in drug analysis is plant material. As reported in Chapter 13, various approaches can be required, namely, fingerprinting, profiling and metabolomics of plant constituents. UHPLC is useful for all of these approaches because of its ability to provide either rapid or high-resolution analysis. Finally, there has been an important development of the metabolomic/metabonomic approaches for biomarker discovery, and UHPLC-MS is again used as a valuable tool for the determination of

biomarkers in biological fluids and tissues. This topic is extensively discussed in Chapter 14.

We would like to conclude by warmly acknowledging the different authors, who are all recognized experts in their fields, for their contributions towards making this book a reference source for UHPLC for people in both academia and industry.

Dr. Davy Guillarme



Prof. Jean-Luc Veuthey

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