

## **Essentials of Modern HPLC/UHPLC 1: Fundamentals and Applications**

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### **Course description**

This intermediate course will provide you with an updated overview and a solid working knowledge of high-performance liquid chromatography (HPLC) and Ultra-high-pressure LC (UHPLC). The attendees will learn useful theoretical concepts, instrumental fundamentals and operating principles, column basics and selection guide, and key applications in various industries. This is the first part of a two-course series. The second part, "Essential of Modern HPLC/UHPLC 2", is a follow-on course that focuses on the practice of modern HPLC: operation, maintenance, troubleshooting, method development as well as performance, practice and potential issues of UHPLC.

### **Target Audience**

Analysts, scientists, researchers, and managers who want to get an updated introduction of modern HPLC fundamentals and its diversified applications. It is recommended that you have a good understanding of general chemistry. Some prior hands-on HPLC experience would be helpful. This course is conducted at the intermediate level.

### **Course Outline**

#### **1. Introduction and fundamental concepts**

- History, advantages, limitations, and modes
- Retention time ( $t_R$ ), retention factor ( $k$ ), separation factor ( $\alpha$ ), column efficiency ( $N$ ), column void volume ( $V_M$ ), and resolution ( $R_S$ ).
- Mobile phase factors (organic modifiers, pH, buffers), operating parameters (Flow, Gradient time ( $t_G$ ), column temperature ( $T$ )), and peak capacity ( $P_c$ )

#### **2. HPLC columns, trends and selection guides**

- Column characteristics and types, packing characteristics (support type, particle size, pore size) and bonding chemistries.
- Trends of shorter and narrower columns packed with small particles, high-purity silica, novel bonding chemistries.
- Van Deemter equation.
- Column selections guide.
- HILIC, monoliths, hybrids, sub-3, sub-2  $\mu\text{m}$  and core shell columns.

#### **3. HPLC instrumentation and operating principles**

- Solvent delivery system, injector, autosampler, detector (UV/Vis, photodiode array, fluorescence, refractive index, ELSD, CAD, conductivity, and mass spectrometer (MS), and data handling system (CDS).
- HPLC system calibration
- Concepts and ramifications of dwell volume and instrumental bandwidth (extra-column bandbroadening).

#### **4. Practical applications of HPLC/UHPLC in diversified industries**

- An overview of HPLC applications in diversified industries supported with specific case studies.
- Pharmaceutical: drug discovery to quality control, assay, impurities, chiral separations, PKDM and dissolution.
- Food: sugars, fats, organic acids, and additives.
- Environmental: US EPA methods, pesticides, and PAHs.
- Chemical: GPC, plastics, and ion-chromatography.
- Bioseparations and life sciences: proteins, peptides, peptide mapping, amino acids, oligonucleotides, nuclei acids, and PCR products.

### **About the instructor**

Dr. Michael W. Dong is a principal in MWD Consulting focusing on training and consulting services in HPLC/UHPLC, pharm analysis and drug quality. He was formerly Senior Scientist at Genentech, Research Director at Synomics Pharma, Research Fellow at Purdue Pharma, and Senior Staff Scientist at Applied Biosystems / Perkin-Elmer. He holds a Ph.D. in Analytical Chemistry from City University of New York, and a certificate in Biotechnology at U. California. Santa Cruz. He has over 100 publications and a best-seller book in HPLC. He is an editorial advisory board member of LCGC magazine and American Pharmaceutical Review.

### **Recommended textbook:**

M. W. Dong, Modern HPLC for Practicing Scientists, Wiley-Interscience, New Jersey, 2006 (ISBN-10: 047172789X). This book is used as a course reference and supplements the course presentation handouts.