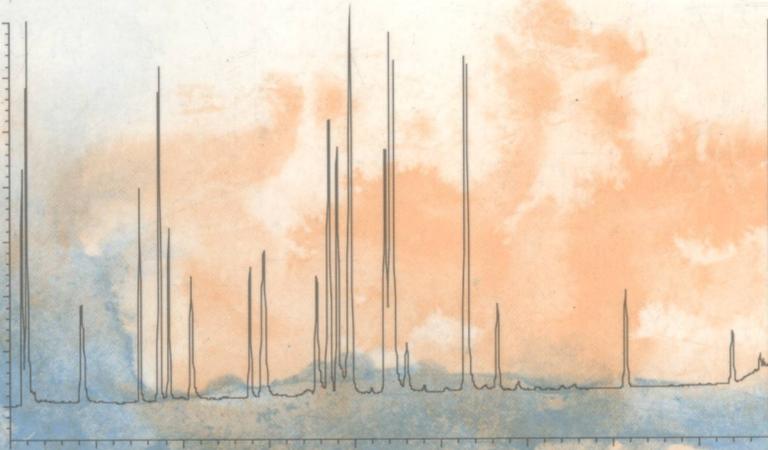


# Modern HPLC **FOR** Practicing Scientists



Michael W. Dong

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# MODERN HPLC FOR PRACTICING SCIENTISTS

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Michael W. Dong

Synomics Pharmaceutical Services, LLC  
Wareham, Massachusetts



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"[...] a concise and 'to-the-point' text, covering the broad topic of HPLC. While the book is not intended to be a comprehensive treatise, it addresses all major topics in HPLC and provides updated, practical information not found in other introductory texts. I found the author's use of bullet points, tabulation and figures extremely effective in conveying practical 'take-home' messages and providing sound and definitive guidance."

—Henrik T. Rasmussen, PhD

"This is a great introductory book on liquid chromatography. I especially like the intuitive explanations and clear figures."

—John W. Dolan, PhD

"Dong's book is especially written for the pharmaceutical industry. I am sure that it will be highly welcomed by the practitioners in this field, not only because of its numerous relevant examples of drug separations but also because of its focus on regulatory aspects."

—Veronika Meyer, PhD

# MODERN HPLC FOR PRACTICING SCIENTISTS

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# PREFACE

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The idea for writing this basic HPLC book was probably born in the New York City subway system while I was a graduate student in the 1970s. Amidst the rumbling noise of the subway, I was reading “the green book”—*Basic Gas Chromatography* by McNair and Bonnelly—and was immediately impressed with its simplicity and clarity. In the summer of 2004, I had just completed the editing of *Handbook of Pharmaceutical Analysis by HPLC* with Elsevier/Academic Press, and was toying with the idea of starting a book project on Fast LC and high-throughput screening. Several phone conversations with Heather Bergman, my editor at Wiley, convinced me that an updated book on modern HPLC, modeled after “the green book,” would have more of an impact.

This book was written with a sense of urgency during weekends and weekday evenings . . . through snow storms, plane trips, allergy seasons, company restructuring, and job changes. The first draft was ready in only 10 months because I was able to draw many examples from my previous publications and from my short course materials for advanced HPLC in pharmaceutical analysis given at national meetings. I am not a fast writer, but rather a methodical one who revised each chapter many times before seeking review advice from my friends and colleagues. My goal was to provide the reader with an updated view of the concepts and practices of modern HPLC, illustrated with many figures and case studies. My intended audience was the practicing scientist—to provide them with a review of the basics as well as best practices, applications, and trends of this fast-evolving technique. Note that this basic book for practitioners was written at both an introductory and intermediate level. I am also targeting the pharmaceutical analysts who constitute a significant fraction of all HPLC users. My focus was biased towards reversed-phase LC and pharmaceutical analysis. The scope of this book does not allow anything more than a cursory mention of the other applications.

Writing a book as a sole author was a labor of love, punctuated with flashes of inspiration and moments of despair. It would have been a lonely journey without the encouragement and support of my colleagues and friends. First and foremost, I would like to acknowledge the professionalism of my editor at John Wiley, Heather Bergman, whose enthusiasm and support made this a

happy project. I also owe much to my reviewers, including the 10 reviewers of the book proposal, and particularly to those whose patience I tested by asking them to preview multiple chapters. They have given me many insights and valuable advice. The list of reviewers is long:

Prof. David Locke of City University of New York (my graduate advisor); Prof. Harold McNair of Virginia Tech, whose “green book” provided me with a model; Prof. Jim Stuart of University of Connecticut; Drs. Lloyd Snyder and John Dolan of LC Resources; Drs. Raphael Ornaf, Cathy Davidson, and Danlin Wu, and Joe Grills, Leon Zhou, Sung Ha, and Larry Wilson of Purdue Pharma; Dr. Ron Kong of Synaptic; Drs. Uwe Neue, Diane Diehl, and Michael Swartz of Waters Corporation, Wilhad Reuter of PerkinElmer; Drs. Bill Barbers and Thomas Waeghe of Agilent Technologies; Drs. Krishna Kallary and Michael McGinley of Phenomenex; Dr. Tim Wehr of BioRad; John Martin and Bill Campbell of Supelco; Dr. Andy Alpert of PolyLC; Margie Dix of Springborn-Smithers Laboratories; Dr. Linda Ng of FDA, CDER; and Ursula Caterbone of MacMod.

Finally, I acknowledge the support and the unfailing patience of my wife, Cynthia, and my daughter, Melissa, for putting up with my long periods of distraction when I struggled for better ways for putting ideas on paper. To them, I pledge more quality time to come after 2006.

*Norwalk, Connecticut*

MICHAEL W. DONG

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### 1.1 INTRODUCTION

#### 1.1.1 Scope

High-performance liquid chromatography (HPLC) is a versatile analytical technology widely used for the analysis of pharmaceuticals, biomolecules,