

Contents

	Preface	XV
	About the Editors	XVII
	List of Contributors	XIX
	List of Abbreviations	XXI
	Notations	XXV
1	Introduction	1
	<i>Henner Schmidt-Traub and Reinhard Ditz</i>	
1.1	Development of Chromatography	1
1.2	Focus of the Book	3
1.3	Recommendation to Read this Book	4
	References	6
2	Fundamentals and General Terminology	7
	<i>Andreas Seidel-Morgenstern, Michael Schulte, and Achim Epping</i>	
2.1	Principles of Adsorption Chromatography	7
2.1.1	Adsorption Process	9
2.1.2	Chromatographic Process	10
2.2	Basic Effects and Chromatographic Definitions	11
2.2.1	Chromatograms and Parameters	11
2.2.2	Voidage and Porosity	12
2.2.3	Influence of Adsorption Isotherms on Chromatogram Shapes	15
2.3	Fluid Dynamics	18
2.3.1	Extra Column Effects	18
2.3.2	Column Fluid Distribution	19
2.3.3	Packing Nonidealities	19
2.3.4	Sources for Nonideal Fluid Distribution	20
2.3.5	Column Pressure Drop	21
2.4	Mass Transfer Phenomena	22
2.4.1	Principles of Mass Transfer	22
2.4.2	Efficiency of Chromatographic Separations	24
2.4.3	Resolution	27
2.5	Equilibrium Thermodynamics	30

2.5.1	Definition of Isotherms	30
2.5.2	Models of Isotherms	32
2.5.2.1	Single-Component Isotherms	32
2.5.2.2	Multicomponent Isotherms Based on the Langmuir Model	34
2.5.2.3	Competitive Isotherms Based on the Ideal Adsorbed Solution Theory	35
2.5.2.4	Steric Mass Action Isotherms for Ion Exchange Equilibria	38
2.6	Thermodynamic Effects on Mass Separation	40
2.6.1	Mass Load	40
2.6.2	Linear and Nonlinear Isotherms	41
2.6.3	Elution Modes	43
	References	45
3	Stationary Phases and Chromatographic Systems	47
	<i>Michael Schulte, Matthias Jöhnck, Romas Skudas, Klaus K. Unger, Cedric du Fresne von Hohenesche, Wolfgang Wewers, Jules Dingenen, and Joachim Kinkel</i>	
3.1	Column Packings	47
3.1.1	Survey of Packings and Stationary Phases	47
3.1.2	Generic, Designed, and Customized Adsorbents	48
3.1.2.1	Generic Adsorbents	48
3.1.2.2	Designed Adsorbents	54
3.1.2.3	Customized Adsorbents	62
3.1.3	Reversed Phase Silicas	66
3.1.3.1	Silanisation of the Silica Surface	67
3.1.3.2	Chromatographic Characterization of Reversed Phase Silicas	69
3.1.4	Cross-Linked Organic Polymers	72
3.1.4.1	General Aspects	73
3.1.4.2	Hydrophobic Polymer Stationary Phases	76
3.1.4.3	Hydrophilic Polymer Stationary Phases	76
3.1.4.4	Ion Exchange (IEX)	77
3.1.4.5	Mixed Mode	85
3.1.5	Chiral Stationary Phases	85
3.1.5.1	Antibiotic CSP	91
3.1.5.2	Synthetic Polymers	91
3.1.5.3	Targeted Selector Design	92
3.1.5.4	Further Developments	93
3.1.6	Properties of Packings and their Relevance to Chromatographic Performance	95
3.1.6.1	Chemical and Physical Bulk Properties	95
3.1.6.2	Mass Loadability	101
3.1.6.3	Comparative Rating of Columns	102
3.1.7	Sorbent Maintenance and Regeneration	103
3.1.7.1	Cleaning in Place (CIP)	103
3.1.7.2	Conditioning of Silica Surfaces	106

3.1.7.3	Sanitization in Place (SIP)	108
3.1.7.4	Column and Adsorbent Storage	108
3.2	Selection of Chromatographic Systems	109
3.2.1	Definition of the Task	114
3.2.2	Mobile Phases for Liquid Chromatography	118
3.2.2.1	Stability	118
3.2.2.2	Safety Concerns	118
3.2.2.3	Operating Conditions	121
3.2.2.4	Aqueous Buffer Systems	123
3.2.3	Adsorbent and Phase Systems	125
3.2.3.1	Choice of Phase System Dependent on Solubility	127
3.2.3.2	Improving Loadability for Poor Solubilities	128
3.2.3.3	Dependency of Solubility on Sample Purity	130
3.2.3.4	Generic Gradients for Fast Separations	131
3.2.4	Criteria for Choosing NP Systems	132
3.2.4.1	Pilot Technique Thin-layer Chromatography	134
3.2.4.2	Retention in NP Systems	134
3.2.4.3	Solvent Strength in Liquid–Solid Chromatography	136
3.2.4.4	Selectivity in NP Systems	138
3.2.4.5	Mobile-Phase Optimization by TLC Following the PRISMA Model	139
3.2.4.6	Strategy for an Industrial Preparative Chromatography Laboratory	148
3.2.5	Criteria for Choosing RP Systems	153
3.2.5.1	Retention and Selectivity in RP Systems	155
3.2.5.2	Gradient Elution for Small amounts of Product on RP Columns	156
3.2.5.3	Rigorous Optimization for Isocratic Runs	157
3.2.5.4	Rigorous Optimization for Gradient Runs	161
3.2.5.5	Practical Recommendations	164
3.2.6	Criteria for Choosing CSP Systems	167
3.2.6.1	Suitability of Preparative CSP	168
3.2.6.2	Development of Enantioselectivity	169
3.2.6.3	Optimization of Separation Conditions	171
3.2.6.4	Practical Recommendations	172
3.2.7	Downstream Processing of Mabs using Protein A and IEX	174
3.2.8	Size Exclusion (SEC)	179
3.2.9	Overall Chromatographic System Optimization	181
3.2.9.1	Conflicts During Optimization of Chromatographic Systems	181
3.2.9.2	Stationary Phase Gradients	184
	References	189
4	Chromatography Equipment: Engineering and Operation	199
	<i>Abdelaziz Toumi, Jules Dingenen, Joel Genoet, Olivier</i>	
	<i>Ludemann-Hombourger, Andre Kiesewetter, Martin Krahe,</i>	
	<i>Michele Morelli, Henner Schmidt-Traub, Andreas Stein, and Eric Valery</i>	
4.1	Introduction	199
4.2	Engineering and Operational Challenges	201

4.3	Chromatography Columns Market	207
4.3.1	Generalities – The Suppliers	207
4.3.2	General Design	208
4.3.3	High- and Low-Pressure Columns	210
4.3.3.1	Chemical Compatibility	211
4.3.3.2	Frits Design	211
4.3.3.3	Special Aspects of Bioseparation	215
4.4	Chromatography Systems Market	217
4.4.1	Generalities – The Suppliers	217
4.4.2	General Design Aspects – High Performance and Low-Pressure Systems	217
4.4.3	Material	219
4.4.4	Batch Low-Pressure Liquid Chromatography (LPLC) Systems	220
4.4.4.1	Inlets	220
4.4.4.2	Valves to Control Flow Direction	220
4.4.4.3	Pumps	221
4.4.4.4	Pump(s) Valves and Gradient Formation	222
4.4.5	Batch High-Pressure Liquid Chromatography (HPLC) Systems	224
4.4.5.1	General Layout	224
4.4.5.2	Inlets and Outlets	224
4.4.5.3	Pumps	226
4.4.5.4	Valves and Pipes	227
4.4.6	Batch SFC Systems	228
4.4.6.1	General Layout	228
4.4.6.2	Inlets	230
4.4.6.3	Pumps, Valves, and Pipes	231
4.4.7	Continuous Systems – Simulated Moving Bed	231
4.4.7.1	General Layout	231
4.4.7.2	A Key Choice: The Recycling Strategy	232
4.4.7.3	Pumps, Inlets, and Outlets	233
4.4.7.4	Valves and Piping	233
4.4.8	Auxiliary Systems	233
4.4.8.1	Slurry Preparation Tank	234
4.4.8.2	Slurry Pumps and Packing Stations	234
4.4.8.3	Cranes and Transport Units	235
4.4.8.4	Filter Integrity Test	235
4.5	Process Control	236
4.5.1	Standard Process Control	236
4.5.2	Advanced Process Control	237
4.5.3	Detectors	240
4.6	Packing Methods	243
4.6.1	Column and Packing Methodology Selection	243
4.6.2	Slurry Preparation	244
4.6.3	Column Preparation	246
4.6.4	Flow Packing	246

4.6.5	Dynamic Axial Compression (DAC) Packing	249
4.6.6	Stall Packing	250
4.6.7	Combined Method (Stall + DAC)	250
4.6.8	Vacuum Packing	252
4.6.9	Vibration Packing	253
4.6.10	Column Equilibration	254
4.6.11	Column Testing and Storage	254
4.6.11.1	Test Systems	254
4.6.11.2	Hydrodynamic Properties and Column Efficiency	256
4.6.11.3	Column and Adsorbent Storage	257
4.7	Process Troubleshooting	257
4.7.1	Technical Failures	258
4.7.2	Loss of Performance	259
4.7.2.1	Pressure Increase	259
4.7.2.2	Loss of Column Efficiency	262
4.7.2.3	Variation of Elution Profile	263
4.7.2.4	Loss of Purity/Yield	264
4.7.3	Column Stability	265
4.8	Disposable Technology for Bioseparations	265
4.8.1	Market Trend	265
4.8.2	Prepacked Columns	266
4.8.3	Membrane Chromatography	267
4.8.4	Membrane Technology	269
	References	270
5	Process Concepts	273
	<i>Malte Kaspereit, Michael Schulte, Klaus Wekenborg, and Wolfgang Wewers</i>	
5.1	Discontinuous Processes	273
5.1.1	Isocratic Operation	273
5.1.2	Flip-Flop Chromatography	275
5.1.3	Closed-Loop Recycling Chromatography	276
5.1.4	Steady-State Recycling Chromatography	278
5.1.5	Gradient Chromatography	279
5.1.6	Chromatographic Batch Reactors	281
5.2	Continuous Processes	283
5.2.1	Column Switching Chromatography	283
5.2.2	Annular Chromatography	283
5.2.3	Multiport Switching Valve Chromatography (ISEP/CSEP)	284
5.2.4	Isocratic Simulated Moving Bed (SMB) Chromatography	286
5.2.5	SMB Chromatography with Variable Process Conditions	290
5.2.5.1	VariCol	290
5.2.5.2	PowerFeed	291
5.2.5.3	Partial-Feed, Partial-Discard, and Fractionation-Feedback Concepts	292
5.2.5.4	Improved/Intermittent SMB (iSMB)	293

5.2.5.5	ModiCon	294
5.2.5.6	FF-SMB	294
5.2.6	SMB Chromatography with Variable Solvent Conditions	294
5.2.6.1	Gradient SMB Chromatography	295
5.2.6.2	Supercritical Fluid SMB Chromatography	296
5.2.7	Multicomponent Separations	296
5.2.8	Multicolumn Systems for Bioseparations	298
5.2.8.1	Sequential Multicolumn Chromatography (SMCC)	298
5.2.8.2	Multicolumn Countercurrent Solvent Gradient Purification (MCSGP)	299
5.2.9	Countercurrent Chromatographic Reactors	301
5.2.9.1	SMB Reactor	301
5.2.9.2	Processes with Distributed Functionalities	302
5.3	Choice of Process Concepts	304
5.3.1	Scale	305
5.3.2	Range of k'	306
5.3.3	Number of Fractions	306
5.3.4	Example 1: Lab Scale; Two Fractions	306
5.3.5	Example 2: Lab Scale; Three or More Fractions	308
5.3.6	Example 3: Production Scale – Wide Range of k'	309
5.3.7	Example 4: Production Scale; Two Main Fractions	310
5.3.8	Example 5: Production Scale; Three Fractions	311
5.3.9	Example 6: Production Scale; Multi-Stage Process	312
	References	315
6	Modeling and Model Parameters	321
	<i>Andreas Seidel-Morgenstern, Henner Schmidt-Traub, Mirko Michel, Achim Epping, and Andreas Jupke</i>	
6.1	Introduction	321
6.2	Models for Single Chromatographic Columns	322
6.2.1	Classes of Chromatographic Models	322
6.2.2	Derivation of the Mass Balance Equations	324
6.2.2.1	Mass Balance Equations	325
6.2.2.2	Convective Transport	327
6.2.2.3	Axial Dispersion	327
6.2.2.4	Intraparticle Diffusion	327
6.2.2.5	Mass Transfer	328
6.2.2.6	Adsorption Kinetics	329
6.2.2.7	Adsorption Equilibrium	329
6.2.3	Equilibrium (“Ideal”) Model	330
6.2.4	Models with One Band Broadening Effect	334
6.2.4.1	Dispersive Model	334
6.2.4.2	Transport Model	336
6.2.4.3	Reaction Model	337
6.2.5	Lumped Rate Models	338

6.2.5.1	Transport-Dispersive Model	338
6.2.5.2	Reaction-Dispersive Model	339
6.2.6	General Rate Models	340
6.2.7	Initial and Boundary Conditions of the Column	343
6.2.8	Models of Chromatographic Reactors	344
6.2.9	Stage Models	344
6.2.10	Assessment of Different Model Approaches	346
6.2.11	Dimensionless Model Equations	348
6.3	Modeling HPLC Plants	350
6.3.1	Experimental Setup and Simulation Flow Sheet	350
6.3.2	Modeling Extra Column Equipment	351
6.3.2.1	Injection System	351
6.3.2.2	Piping	352
6.3.2.3	Detector	352
6.4	Calculation Methods	353
6.4.1	Analytical Solutions	353
6.4.2	Numerical Solution Methods	353
6.4.2.1	General Solution Procedure	353
6.4.2.2	Discretization	354
6.5	Parameter Determination	357
6.5.1	Parameter Classes for Chromatographic Separations	357
6.5.1.1	Design Parameters	357
6.5.1.2	Operating Parameters	358
6.5.1.3	Model Parameters	358
6.5.2	Determination of Model Parameters	359
6.5.3	Evaluation of Chromatograms	361
6.5.3.1	Moment Analysis and HETP Plots	362
6.5.3.2	Parameter Estimation	369
6.5.3.3	Peak Fitting Functions	370
6.5.4	Detector Calibration	374
6.5.5	Plant Parameters	375
6.5.6	Determination of Packing Parameters	376
6.5.6.1	Void Fraction and Porosity of the Packing	376
6.5.6.2	Axial Dispersion	377
6.5.6.3	Pressure Drop	378
6.5.7	Isotherms	379
6.5.7.1	Determination of Adsorption Isotherms	379
6.5.7.2	Determination of the Henry Coefficient	382
6.5.7.3	Static Isotherm Determination Methods	382
6.5.7.4	Dynamic Methods	385
6.5.7.5	Frontal Analysis	385
6.5.7.6	Analysis of Disperse Fronts (ECP/FACP)	390
6.5.7.7	Peak Maximum Method	391
6.5.7.8	Minor Disturbance/Perturbation Method	392
6.5.7.9	Curve Fitting of the Chromatogram	394

6.5.7.10	Prediction of Mixture Behavior from Single-Component Data	395
6.5.7.11	Data Analysis and Accuracy	396
6.5.8	Mass Transfer	398
6.5.9	Identification of Isotherms and Mass Transfer Resistance by Neural Networks	399
6.6	Experimental Validation of Column Models	401
6.6.1	Batch Chromatography	401
6.6.2	SMB Chromatography	404
6.6.2.1	Model Formulation and Parameters	404
6.6.2.2	Experimental Validation of SMB Models	410
	References	418
7	Model-Based Design, Optimization, and Control	425
	<i>Henner Schmidt-Traub, Malte Kaspereit, Sebastian Engell, Arthur Susanto, Achim Epping, and Andreas Jupke</i>	
7.1	Basic Principles and Definitions	425
7.1.1	Performance, Costs, and Optimization	425
7.1.1.1	Performance Criteria	426
7.1.1.2	Economic Criteria	428
7.1.1.3	Objective Functions	429
7.1.2	Degrees of Freedom	430
7.1.2.1	Optimization Parameters	430
7.1.2.2	Dimensionless Operating and Design Parameters	430
7.1.3	Scaling by Dimensionless Parameters	435
7.1.3.1	Influence of Different HETP Coefficients for Every Component	436
7.1.3.2	Influence of Feed Concentration	437
7.1.3.3	Examples for a Single Batch Chromatographic Column	438
7.1.3.4	Examples for SMB Processes	440
7.2	Batch Chromatography	442
7.2.1	Fractionation Mode (Cut Strategy)	442
7.2.2	Design and Optimization of Batch Chromatographic Columns	444
7.2.2.1	Design and Optimization Strategy	444
7.2.2.2	Process Performance Depending on Number of Stages and Loading Factor	447
7.2.2.3	Other Strategies	452
7.3	Recycling Chromatography	453
7.3.1	Design of Steady-State Recycling Chromatography	454
7.3.2	Scale-Up of Closed Loop Recycling Chromatography	457
7.4	Conventional Isocratic SMB Chromatography	461
7.4.1	Optimization of Operating Parameters	462
7.4.1.1	Process Design Based on TMB Models (Shortcut Methods)	463
7.4.1.2	Process Design Based on Rigorous SMB Models	471
7.4.2	Optimization of Design Parameters	476

7.5	Isocratic SMB Chromatography under Variable Operating Conditions	481
7.6	Gradient SMB Chromatography	490
7.7	Multicolumn Systems for Bioseparations	495
7.8	Advanced Process Control	497
7.8.1	Online Optimization of Batch Chromatography	498
7.8.2	Advanced Control of SMB Chromatography	501
7.8.2.1	Purity Control for SMB Processes	502
7.8.2.2	Direct Optimizing Control of SMB Processes	503
7.8.3	Advanced Parameter and State Estimation for SMB Processes	509
	References	510

Appendix A: Data of Test Systems 519

Index 527