Contents

Dedication V List of Contributors XXI Preface XXVII A Personal Foreword XXXI

Volume 68a

- Part I Introduction to Lead Generation 1
- 1 Introduction: Learnings from the Past Characteristics of Successful Leads 3 Mike Hann Acknowledgments 10 References 10

2 Modern Lead Generation Strategies 13

Jörg Holenz and Dean G. Brown

- 2.1 Lead Generation Greatly Influences Clinical Candidate Quality 14
- 2.2 Screening of Compound Libraries has Undergone a Major Paradigm Change 15
- 2.3 New Chemical Modalities are Available to Tackle Difficult Targets 15
- 2.4 As Demands have Increased, New Lead Generation Methods Emerged 16
- 2.5 How do Lead Generation Chemists Meet These Challenges and Subsequently Provide Their Lead Optimization Colleagues with High-Quality Lead Series? *17*
- 2.5.1 Learnings can be Drawn from LG Project Failures 17
- 2.5.2 How Many Compounds to Screen to Generate High-Quality Leads? 18
- 2.5.3 Which Compounds to Screen to Generate High-Quality Leads? 19
- 2.5.4 Developing Project-Customized, Concerted, and Comprehensive Lead Generation Strategies will Increase LG Success Rates: the *CREATION* of Leads 20
- 2.5.5 Selecting the Target Defines LG Success Rates 21

VIII Contents

2.5.6	Lead Generation shou	Ild be Complemented by Auxiliary Technologies
	to Characterize Hits	21

- 2.5.7 Phenotypic Screens are Often Complemented by a Chemical Biology Arm 22
- 2.5.8 The Lead Generation Strategy is Defined by the Budget Allocated 22
- 2.5.9 Cost-Efficient but Information-Rich Lead Generation Strategies 23
- 2.5.10 The Revival of Potency as the Most Important Lead Criterion? 24
- 2.5.11 When has a LG Campaign Delivered Successfully? 27 References 31

The Importance of Target Identification for Generating Part II Successful Leads 35

3	"Ligandability" of Drug Targets: Assessment of Chemical		
	Tractability via Experimental and In Silico Approaches	37	
	Udo Bauer and Alexander L. Breeze		

- 3.1 Introduction 37
- The Concept of Ligandability 39 3.2
- 3.2.1 General Characteristics of Ligandable Targets 39
- 3.3 The Intersection of Ligandability and Human Disease Target Space 40
- 3.3.1 Experimental Techniques for Assessing Target Ligandability 42
- 3.3.1.1 High-Throughput Screening and Subset/"Validation Set" Screening 43
- 3.3.1.2 Fragment Screening 44
- Practical Examples of the Use of Fragment Screening for Ligandability 3.4 Assessment 50
- 3.4.1 Chemical Tractability Assessment by in silico Approaches 54
- 3.4.1.1 Pocket-Finding Algorithms 54
- 3.4.1.2 Discrimination Functions and Validation Sets 55
- Simulation-Based Methods for Identifying Interaction Potentials 56 3.4.1.3
- 3.5 Conclusions and Outlook 56 References 58

Chemistry-Driven Target Identification 63 4

Iván Cornella-Taracido, Ryan Hicks, Ola Engkvist, Adam Hendricks, Ronald Tomlinson, and M. Paola Castaldi

- 4.1 Introduction 63
- 4.2 Chemistry-Driven Target Discovery: Enabling Biology 65
- 4.2.1 Biological Samples 65
- 4.2.2 Cells Cultured in 2D 66
- 4.2.3 Cells Cultured in 3D, Organoids, and Tissues 67
- 4.2.4 Nonhuman Cells and Whole-Organism Screening 68
- 4.2.5 Functional Assays and Readouts 68
- 4.3 Chemistry for Target Discovery 71
- 4.3.1 Screening Deck Selection 71

- 4.3.2 Triaging and Prioritization of Chemical Matter 72
- 4.3.3 SAR Expansion and Probe Synthesis for Target Deconvolution 73
- 4.4 Small-Molecule Target Identification Techniques 75
- 4.4.1 In Silico Target Deconvolution 75
- 4.4.2 Biochemical Profiling 77
- 4.4.3 Target Deconvolution Correlational Tools 78
- 4.4.4 Subcellular Localization 79
- 4.4.5 Chemical Genetics 79
- 4.4.6 Affinity Chemical Proteomics 81
- 4.4.7 Target Corroboration 84
- 4.5 Conclusions 86 References 89
- Part III Hit Generation Methods 93
- 5 Lead Generation Based on Compound Collection Screening 95 Dirk Weigelt and Ismet Dorange
- 5.1 Introduction 95
- 5.2 Screening of Existing Collections: the General Workflow 96
- 5.2.1 High-Throughput Screening 96
- 5.2.2 Medium-Throughput Screening: Selection Methods 98
- 5.3 Generation of New Screening Compounds 99
- 5.3.1 Collection Enhancement Programs 102
- 5.3.2 Library Design and Compound Selection 102
- 5.3.2.1 Number of Dimensions 103
- 5.3.2.2 Enumeration and Filtering 104
- 5.3.2.3 Layout 106
- 5.3.3 Focus on Synthetic Feasibility 107
- 5.3.3.1 Multicomponent Reactions 107
- 5.3.3.2 Click Chemistry 108
- 5.3.3.3 Diversity-oriented Synthesis 108
- 5.3.4 Structure-driven Approaches 109
- 5.3.4.1 Privileged Structures 110
- 5.3.4.2 Structure-driven Approaches Toward Unchartered Territory 112
- 5.3.5 Target Focus 114
- 5.3.5.1 Kinases 114
- 5.3.5.2 G-Protein-Coupled Receptors 115
- 5.3.5.3 Ion Channels 116
- 5.3.5.4 Protein–Protein Interactions 117
- 5.4 Other Concepts 117
- 5.4.1 Natural Products 118
- 5.4.2 DNA-Encoded Libraries 119
- 5.4.3 Spatially Addressed Libraries 120
- 5.4.4 On-bead Screening 120
- 5.4.5 Dynamic Combinatorial Chemistry 121

X Contents

5.4.6 5.5	Cocktails and Mixtures 121 Summary and Outlook 122			
	References 123			
6	Fragment-Based Lead Generation 133 Ivan V. Efremov and Daniel A. Erlanson			
6.1	Introduction 133			
6.2	Screening Methods 135			
6.3	Hit Validation 137			
6.4	Ligand Efficiency and Other Metrics 138			
6.5	Hit Optimization 139			
6.6	Fragment Growing 140			
6.7	Fragment Linking 144			
6.8	Protein–Protein Interactions 147			
6.9	GPCRs 151			
6.10	Computational Approaches 152			
6.11	Conclusions 153			
	References 154			
7	Rational Hit Generation 159			
	Bernd Wellenzohn and Alexander Weber			
7.1	Introduction 159			
7.2	Lead Generation: Transition State and Substrate Analogs 161			
7.3	Hit Generation by Rational Library Design 165			
7.4	Hit Generation by Virtual Screening 167			
7.4.1	Structure-based VS in Enumerated Molecules 170			
7.4.2	Ligand-based VS in Nonenumerated Virtual Chemical Spaces 172			
7.5	Hit Generation by Scaffold Replacement Technologies 173			
7.6	Hit Generation by Chemogenomics Approaches 174			
7.7	Summary 178			
	References 178			
8	Competitive Intelligence-based Lead Generation and Fast Follower			
	Approaches 183			
	Yu Jiang, Ziping Liu, Jörg Holenz, and Hua Yang			
8.1	Introduction 183			
8.2	Competitive Intelligence-based Approach 185			
8.2.1	Example A: A Case Study for the Hybrid Strategy 190			
8.2.2	Example C: A Case Study for the Fused Strategy 192			
8.2.3	Example C: A Case Study for the Fused Strategy 193			
8.2.4	Example D: A Case Study for the Fused Strategy 196			
8.2.5	Example E: A Case Study for the Chimera Strategy 197			
8.3	Fast Follower Approach 201			
8.3.1	Salfanilamide-based Fast Follower Approaches 202			
8.3.2	Omeprazole-based Fast Follower Approaches 203			
8.3.3	Kimonabant-based Fast Follower Approach 210			
	Keterences 214			

171

Contents XI

Selective Optimization of Side Activities: An Alternative and Promising 9 Strategy for Lead Generation 221 Norbert Handler, Andrea Wolkerstorfer, and Helmut Buschmann 9.1 Introduction 221 9.1.1 Drug Selectivity and Unwanted or Desired Side Effects 222 9.2 Definition, Rational, and Concept of the SOSA Approach 223 9.2.1 Multiple Ligands and Polypharmacology 224 Safety and Bioavailability 225 9.2.2 9.3 Drugs in Other Drugs: Drug as Fragments 225 9.4 Drug Repositioning and Drug Repurposing 226 9.4.1 Old Drugs 226 9.5 The SOSA Approach and Analog Design 227 9.6 Patentability and Interference Risk of the SOSA Approach 230 9.6.1 Analogization, Optimization, and Isosterism 230 9.7 Case Studies and Examples 231 9.7.1 Sulfonamides 231 9.7.2 Morphine Analogs 232 9.7.3 Warfarin 232 9.7.4 Sildenafil (Viagra) 232 9.7.5 Thalidomide Analogs 233 9.7.6 Bupropion 234 9.7.7 Chlorpromazine 235 9.7.8 Chlorothiazide 235 9.7.9 Propranolol 235 9.7.10 Minaprine Analogs 236 9.7.11 Viloxazine Analogs 237 9.7.12 Methylation in the SOSA Strategy of Drug Design 237 9.7.13 Discovery of New Antiplasmodial Compounds 239 Drugs Acting on Central Nervous System Targets as Leads for 9.7.14 Non-CNS Targets 241 9.7.15 Mexiletine Derivatives as Orally Bioavailable Inhibitors of Urokinase-Type Plasminogen Activator 242 9.7.16 Amiloride Analogs as Inhibitors of the Urokinase-type Plasminogen Activator 245 Flavonoids with an Oligopolysulfated Moiety: A New Class of 9.7.17 Anticoagulant Agents 246 9.7.18 Clioquinol 249 9.8 Conclusions 251 References 252 10 Lead Generation for Challenging Targets 259 Jingiao Wan, Dengfeng Dou, Hongmei Song, Xian-Hui Wu, Xuemin Cheng, and Jin Li Introduction 259 10.1 10.2 DNA-Encoded Library Technology in Lead Generation 260

- XII Contents
 - 10.2.1 Background 260
 - 10.2.2 DNA-Recorded Synthesis-Assisted Libraries 262
 - 10.2.3 DNA-Templated Synthesis-Assisted Libraries 264
 - 10.2.4 Encoded Self-Assembling Chemical Libraries 266
 - 10.2.5 Summary and Perspective 267
 - 10.3 Stapled Peptide 276
 - 10.3.1 Background 276
 - 10.3.2 Structure, Design, and Synthesis of Stapled Peptide 278
 - 10.3.2.1 Stapled Peptide Structure 278
 - 10.3.2.2 Stapled Peptide Design 280
 - 10.3.2.3 Stapled Peptide Synthesis 282
 - 10.3.3 Stapled Peptide Solution α-Helix Conversion Measurement 283
 - 10.3.4 Stapled Peptide Affinity Evaluation and α-Helix Content Correlation 284
 - 10.3.4.1 Surface Plasmon Resonance Binding Assays 284
 - 10.3.4.2 Fluorescence Polarization Assay 284
 - 10.3.4.3 Stapled Peptide Affinity and α-Helix Content Correlation 285
 - 10.3.5 Stapled Peptide Permeability 286
 - 10.3.6 Peptide Stability Assay 288
 - 10.3.7 Outlook 288
 - 10.4 Phenotypic Screening 289
 - 10.4.1 Introduction 289
 - 10.4.2 Basics for Establishing a Phenotypic Screen 291
 - 10.4.2.1 Identify a "Druggable" Phenotype and the Type of Readout 291
 - 10.4.2.2 Assay Design 291
 - 10.4.2.3 Hit Selection and Secondary Assay 291
 - 10.4.3 Typical Phenotypic Assays 292
 - 10.4.3.1 Cell-Viability Assay 292
 - 10.4.3.2 Fluorescent Imaging Plate Reader Technology 293
 - 10.4.3.3 High-Content Screening 293
 - 10.4.4 In Vitro Phenotypic Screening 293
 - 10.4.4.1 Classic Phenotypic Screening 293
 - 10.4.4.2 Patient-Derived Stem Cell in Drug Discovery 294
 - 10.4.4.3 Phenotypic Screening on iPSC-Derived Disease Models 295
 - 10.4.4.4 High-Content Cytotoxicity Screening by iPSC-Derived Hepatocytes 296
 - 10.5 Summary 297 References 298

11 Collaborative Approaches to Lead Generation 307

Fabrizio Giordanetto, Anna Karawajczyk, and Graham Showell

- 11.1 Introduction 307
- 11.2 Creativity 308
- 11.3 Speed 308
- 11.4 Risk Sharing 308
- 11.5 Intellectual Property 309
- 11.6 Costs 309

- 11.7 Management 310
- 11.8 Lilly's Open Innovation Drug Discovery 310
- 11.9 Molecular Library Program 312
- 11.10 EU Openscreen 314
- 11.11 European Lead Factory 315
- 11.12 Medicines for Malaria Venture 317
- 11.13 Open Source Malaria Project 320
- 11.14 Drugs for Neglected Diseases Initiative 320
- 11.15 Open Lab Foundation 321
- 11.16 Scientists Against Malaria 322
- 11.17 Open Source Drug Discovery 323
- 11.18 TB Alliance 323
- 11.19 Summary 324 References 325

Volume 68b

Dedication V List of Contributors XXI

- Part IV Converting Hits to Successful Leads 329
- 12 A Medicinal Chemistry Perspective on the Hit-to-Lead Phase in the Current Era of Drug Discovery 331 Dean G. Brown
- 12.1 Introduction 331
- 12.2 Active to Hit Processes 333
- 12.3 Target Potency: Energetics of Binding 336
- 12.4 Addressing Vast Chemical Space: HtL Strategies 345
- 12.5 Matched Pair Analysis 348
- 12.6 The Role of Hydrophobicity and HtL 351
- 12.7 Probing H-Bond Donors and Acceptors 353
- 12.8 Structure Based DD in HtL 356
- 12.9 Statistical Molecular Design 358
- 12.10 Hit to Lead is not Lead Optimization 359
- 12.11 Summary 362 References 363
- 13 Molecular Recognition and Its Importance for Fragment-Based Lead Generation and Hit-to-Lead 367 Thorsten Nowak
- 13.1 Introduction 367
- 13.2 Brief Summary of the Main Factors that Govern Molecular Interactions 368

XIV Contents

13.3	Thermodynamics	of Molecular	Interactions	and Imp	pact on 1	Hit Fi	nding
	and Optimization	369					

- 13.4 Enthalpy as a Key Decision Tool in Medicinal Chemistry 371
- 13.5 Importance of Enthalpic Interactions: Drivers of Selectivity and Specificity? 373
- 13.6 Fragment Screening Hit Optimization: Fragment Linking 374
- 13.7 Interstitial Waters and Their Usefulness: Case Studies on HSP-90 381
- 13.8 Fragments to Find Hot Spots in Binding Pockets 385
- 13.9 Nonclassical Hydrogen Bonds – Interactions of Halogen Atoms with Π-Systems and Carbonyl Groups: Factor Xa and Cathepsin L 386
- 13.10 Binding Mode Dependency of the Experimental Conditions and Chemical Framework of Ligand 390
- 13.11 Cooperativity in Binding: DAO or DAAO D-Amino Acid Oxidase 391 References 394
- 14 Affinity-Based Screening Methodologies and Their Application in the Hit-to-Lead Phase 401
- Stefan Geschwindner
- 14.1 Introduction 401
- 14.2 Nuclear Magnetic Resonance Spectroscopy 402
- 14.3 Optical Biosensors: Surface Plasmon Resonance and Optical Waveguide Grating 404
- 14.4 Isothermal Titration Calorimetry 407
- 14.5 Thermal Shift Assay 411
- 14.6 Mass Spectrometry Approaches 412
- 14.7 Encoded Library Technologies 414
- 14.8 Emerging Technologies: Microscale Thermophoresis and Backscattering Interferometry 417 References 418
- 15 Predictive Methods in Lead Generation 425 Matthew D. Segall and Peter Hunt
- 15.1 Introduction 425
- 15.2 Compound Property Prediction 427
- 15.3 Multiparameter Optimization: Identifying High-Quality Compounds 430
- 15.3.1 Drug-like Properties 430
- 15.3.2 Filters 431
- 15.3.3 Desirability Functions and Probabilistic Scoring 432
- 15.3.4 Pareto Optimization 435
- 15.3.5 Example 436
- 15.4 De Novo Design: Guiding the Exploration of Novel Chemistry 439
- 15.4.1 Example Application 442
- 15.5 Selection: Balancing Quality with Diversity 443
- 15.6 Conclusions 445 References 447

- **16 Lead Quality** *451*
 - J. Willem M. Nissink, Sebastien Degorce, and Ken Page
- 16.1 Introduction 451
- 16.2 Properties in Drug Design 452
- 16.2.1 Primary Activity Assays 453
- 16.2.2 Physicochemical Properties 453
- 16.2.3 DMPK 454
- 16.2.4 Safety 454
- 16.2.5 Overall Profiles 456
- 16.3 Optimizing Properties: Useful Rules, Guides, and Simple Metrics for Early-Stage Projects 457
- 16.3.1 Rules for Potency: Ligand Efficiency Measures 457
- 16.3.2 Rules for Safety 462
- 16.3.3 Rules for DMPK and Mode of Administration: Early-Stage Structure-Based Profiling 464
- 16.3.3.1 Simple Design Rules for Good DMPK 464
- 16.3.3.2 Other DMPK Design Rules 465
- 16.3.4 Multiobjective Optimization 466
- 16.4 Predicted Dose to Man as a Measure of Early- and Late-Stage Lead Quality *467*
- 16.4.1 Introduction 467
- 16.4.2 Description of Models and Data 469
- 16.4.3 Data Supporting Technique 471
- 16.4.3.1 Matching eD2M Doses with Normalized Observed Clinical Doses 472
- 16.4.3.2 Matching C_{max} Values from eD2M and Clinical Studies 472
- 16.4.4 Flagging Potential Candidate Drugs Using eD2M 473
- 16.4.5 Determining Properties that Drive eD2M Predictions for a Series 474
- 16.5 Summary 480 References 481

Part V Hypothesis-driven Lead Optimization 487

- 17 The Strategies and Politics of Successful Design, Make, Test, and Analyze (DMTA) Cycles in Lead Generation 489 Steven S. Wesolowski and Dean G. Brown
- 17.1 DMTA Cycles: Perspectives from History 490
- 17.2 Test: What Assays, in What Order, and Why? 494
- 17.3 Additional Advice for "Test" Component of DMTA 496
- 17.4 Design: What to Make and Why? 496
- 17.5 Additional Advice for "Design" Component of DMTA 500
- 17.6 Make: Challenges and Strategies for Synthesis 501
- 17.7 Additional Advice for the "Make" Component of DMTA 502

XVI Contents

17.8	Analyze: Making Sense of What's Been Done and Formulating Sensible Plans for the Next Designs 502
17.9	Additional Advice for "Analyze" Component of DMTA 508
17.10	Results: Do Lead Optimization Teams Get What
	They Need? 508
	References 509
Part VI	Recent Lead Generation Success Stories 513
10	
18	Lead Generation Paved the way for the Discovery of a Novel H_3 inverse
	Agonist Clinical Candidate 515
10.1	Christophe Genicot and Laurent Provins
18.1	Introduction 515
18.2	Hit Identification 517
18.3	Lead Generation 521
18.3.1	Exploration of Oxazoline Substitution 523
18.3.2	Rigidification of Propoxy Linker 531
18.3.3	Caralysiana 520
18.3.4	Conclusions 550
18.4	Conclusions 542
16.5	Admouledgements 544
	Poforoncos 544
	Keleichtes 344
19	Vorapaxar: From Lead Identification to FDA Approval 547
	Samuel Chackalamannil and Mariappan Chelliah
19.1	Introduction 547
19.2	Background Information on Antiplatelet Agents 549
19.3	Thrombin Receptor (Protease-activated Receptor-1) Antagonists as a
	Novel Class of Antiplatelet Agents 550
19.4	Mechanism of Thrombin Receptor Activation 550
19.5	Preclinical Data Supporting the Antiplatelet Effect of Thrombin
	Receptor Antagonists 551
19.6	Himbacine-derived Thrombin Receptor Antagonists 552
19.6.1	Lead Identification 552
19.6.2	Lead Generation of Himbacine-derived Thrombin Receptor
	Antagonist Hit 553
19.6.2.1	Structure–Activity Relationship Studies 555
19.6.2.2	First-Generation Thrombin Receptor Antagonists 556
19.6.2.3	<i>In vivo</i> Metabolism of Himbacine Derivatives 558
19.6.2.4	Generation of Aryl Himbacine Leads 561
19.6.2.5	Second-Generation Leads that Incorporate Heteroatoms in the
	C-ring 562
19.6.2.6	Identification of nor-seco Himbacine Lead 564

- 19.6.3 Discovery of Vorapaxar (SCH 530348) 565 19.6.3.1 Clinical Studies of Vorapaxar 567 197 Conclusions 569 Abbreviations 570 Acknowledgments 570 References 571 20 Lead Generation Approaches Delivering Inhaled B2-Adrenoreceptor Agonist Drug Candidates 575 Michael Stocks and Lilian Alcaraz 20.1Introduction 575 20.2 Lead Generation Exercises to Discover B2AR Agonist Clinical Candidates 577 20.3 AstraZeneca Lead Generation Exercises to Discover B2AR Agonist Clinical Candidates 587 20.4 Summary 593 References 593 21 GPR81 HTS Case Study 597 Eric Wellner and Ola Fjellström 21.1 General Remarks 597 21.2 The Target 598 21.3 Screening Cascade 599 21.4 Compound Selection (10 K Validation Set) 602 HTS 606 21.5 CSE 608 21.5.121.5.2 Single-Concentration Counterscreen 614 21.5.3 Clustering 615 21.5.4 Cluster Expansion and Nearest Neighbours 618 21.6 Hit Evaluation 618 21.6.1 Potency, Efficacy, and Curves 618 21.6.2 Binding Kinetics 621 Concentration–Response Counterscreen 622 21.6.3 21.6.4 Hit Assessment 622 21.6.4.1 Size and Lipophilicity Efficiency Assessment 622 21.6.4.2 Secondary Pharmacology Assessment 626 21.6.5 Secondary Screening Cascade and Hit Expansion 630 21.6.6 Biological Effect Assay 634 21.7 Alternative Lead Generation Strategies 638 21.7.1 Pepducins and Other Modified Peptides 641 21.8 Conclusions 645 References 646 22 Development of Influenza Virus Sialidase Inhibitors 651 Mauro Pascolutti, Robin J. Thomson, and Mark von Itzstein
 - 22.1 Introduction 651

- **XVIII** Contents
 - 22.2 Targets for Anti-influenza Drug Development: Receptor Binding and Receptor Cleavage 652
 - 22.2.1 Targeting Receptor Binding by Haemagglutinin 654
 - 22.2.2 Targeting Receptor Destruction by Sialidase 655
 - 22.2.3 Influenza Virus Sialidase: Structure and Mechanism 656
 - 22.3 Development of Influenza Virus Sialidase Inhibitors 658
 - 22.3.1 The Development of Zanamivir: Proof of Concept and First-in-Class Sialidase Inhibitor Drug 659
 - 22.3.1.1 Template Selection 659
 - 22.3.1.2 Structure-based Inhibitor Design 662
 - 22.3.1.3 X-Ray Crystallographic Confirmation of Inhibitor Binding Mode 665
 - 22.3.1.4 Selectivity for Influenza Virus Sialidase over Human Sialidases 666
 - 22.3.1.5 Efficacy against Virus Replication 667
 - 22.3.1.6 Mode of Administration of the Highly Polar Drug 667
 - 22.3.1.7 Modifying the Presentation of Zanamivir: Prodrugs and Multivalency 668
 - 22.3.2 Sialidase Inhibitor Development on Noncarbohydrate Scaffolds 671
 - 22.3.2.1 A Sialidase Inhibitor Based on a Cyclohexene Scaffold: The Development of Oseltamivir 671
 - 22.3.2.2 A Sialidase Inhibitor Based on a Cyclopentane Scaffold: The Development of Peramivir 673
 - 22.3.3 Monitoring Resistance to Influenza Virus Sialidase Inhibitors 675
 - 22.4 Summary and Future Directions 676 References 676
 - 23 The Discovery of Cathepsin A Inhibitors: A Project-Adapted Fragment Approach Based on HTS Results 687

Sven Ruf, Christian Buning, Herman Schreuder, Wolfgang Linz, Dominik Linz, Hartmut Rütten, Georg Horstick, Markus Kohlmann, Katja Kroll, Klaus Wirth, and Thorsten Sadowski

- 23.1 General Background 687
- 23.2 Cathepsin A enzyme 687
- 23.2.1 Structural Biology and Catalytic Mechanism 687
- 23.2.2 Structural and Catalytic Functions of CatA 689
- 23.2.3 Tissue Distribution and Substrates 689
- 23.2.4 Natural Products and Synthetic Peptides as Inhibitors of CatA 690
- 23.3 CatA and the Link to Cardiovascular Disease 691
- 23.4 Lead Discovery 692
- 23.4.1 High-Throughput Screening and Data Analysis 692
- 23.4.2 Evaluation of Hit Series 693
- 23.4.2.1 Covalent Inhibitor Series 693
- 23.4.2.2 Malonamide Series 697
- 23.4.2.3 Pyrazolone Hit Series 698
- 23.4.3 Explorative Chemistry Delivers a Novel Lead Structure 699
- 23.4.3.1 Crystal Structure of 9b Bound to CatA 705

- 23.5 Lead Optimization 705
- 23.6 Toward an *in vivo* Proof of Concept 711
- 23.7 Summary and Conclusions *713*

References 714

24 Lead Structure Discovery for Neglected Diseases: Product Development Partnerships Driving Drug Discovery 717

Jeremy N. Burrows and Takushi Kaneko

- 24.1 Introduction 717
- 24.2 Malaria and Medicines for Malaria Venture 719
- 24.3 Malaria Lead Generation Strategy 719
- 24.4 Hit Identification Strategies 722
- 24.5 Optimization of a Marketed Antimalarial Chemotype 723
- 24.6 Target-Based Approaches 723
- 24.7 Asexual Blood-Stage Phenotypic Screening 724
- 24.8 Whole-Cell Screening: Results 725
- 24.9 Repositioning of Clinical Candidates Developed for Other Indications 726
- 24.10 Case Studies 727
- 24.10.1 Dihydroorotate Dehydrogenase (DHODH) 727
- 24.10.2 Whole-Cell Screening 728
- 24.11 Screening for Malaria Eradication 729
- 24.12 Tuberculosis and the Global Alliance for Tuberculosis Drug Development (TB Alliance) 729
- 24.13 Target Product Profiles 730
- 24.14 TB Alliance's Mission 730
- 24.15 Hit Generation Strategies for TB 732
- 24.16 Examples of Phenotypic Screens 733
- 24.17 Conclusions 741 References 741
- 25 A Fragmentation Enumeration Approach to Generating Novel Drug Leads 747

Pravin S. Iyer and Manoranjan Panda

- 25.1 Introduction 747
- 25.2 Principle 748
- 25.3 Research Methodology 748
- 25.3.1 Fragmentation 749
- 25.3.1.1 Origin of Parent Molecules 749
- 25.3.1.2 Cores and Daughters 749
- 25.3.1.3 Nonflat Cores 751
- 25.3.2 Intelligent Recombination and Enumeration 754
- 25.4 Evaluation 754
- 25.4.1 Preliminary Experimental Evaluation 755
- 25.4.2 In Silico Evaluation 755

XX Contents

25.4.3	Virtual Screening Using Enzyme–Ligand Docking	756
25.5	Summary 758	
	References 759	

Index 761