High-Throughput Screening in Drug Discovery

Edited by
Jörg Hüser

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Foreword

Random screening of comprehensive compound collections constitutes a major source of novel lead structures reflected by industry's ongoing commitment to invest in extensive compound libraries and screening technologies. During the last decade, High-Throughput Screening (HTS) has evolved to become an innovative multidisciplinary branch in biological chemistry combining aspects of medicinal chemistry, biology, and laboratory automation. While basic medicinal chemistry techniques and views are largely conserved throughout industry, HTS strategies differ to a great extend. Each strategy can be justified by scientific rationale. However, it is also the result of different scientific backgrounds, different therapeutic areas, different technical expertise within a group, and different ways HTS is integrated within the overall discovery process in a given organization. For most strategies, the close interrelation between HTS and the molecular target approach to drug discovery renders the validity of a disease link for a selected biomolecular target an essential prerequisite for success. As a consequence, a critical assessment of HTS has to incorporate also reflections on the discovery process from target selection to appropriate screening cascades. A different approach employing phenotypic readouts, e.g. cell proliferation, has a long tradition in screening, particularly for chemotherapeutic principles in cancer and antiinfectives research. Similarly, chemical genetics makes use of small molecule perturbation of specific cellular responses to unravel the underlying gene and pathways function. Within the later paradigm, High-Throughput Screening techniques have gained increasing relevance also in academic research.

The current book presents a collection of review-style papers written by experts in the field intended to provide insights into selected aspects of the experimental lead discovery process in High-Throughput Screening. It is by no means claimed to comprehensively cover the entire field. A number of aspects have been discussed in previous volumes within this series on "Methods and Principles in Medicinal Chemistry". It complements this book series by illustrating HTS as one of the technologies of great relevance to the medicinal chemist and molecular pharmacologist working in pharmaceutical or academic research.

I am personally thankful to the Series Editors not only for providing the opportunity to present High-Throughput Screening within a single dedicated

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volume, but also for their patience during the preparation of this volume. In addition, the continuous support of my colleagues, Stefan Mundt, Nils Griebenow and Peter Nell, is gratefully acknowledged.

Wuppertal, July 2006
Jörg Hüser

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Preface

Biological "trial and error" testing of large collections of small molecules for a specific pharmacological effect is the classical route to discover novel lead compounds which subsequently serve as templates for further optimization in medicinal chemistry programs. During the past two decades, the advent of recombinant DNA technologies together with improved assay techniques and high-performance laboratory automation dramatically changed the pharmacological screening process. Nowadays, high-throughput screening (HTS) has gained attention also in the academic environment. Using HTS of compound collections in combination with cell-based tests, small molecule modulators of relevant biochemical and signal transduction pathways should be identified. Following identification of the biological target, the small molecule modulator (inhibitor or activator) substitutes mutational analysis to unravel the target protein function. The vision of this approach, referred to as "Chemical Genetics", is to identify a small molecule partner for every gene product. In the future, pharmaceutical drug research might be stimulated by Chemical Genetics by revealing novel drug targets and initial lead structures.

The present volume provides fascinating insights into this important part of the early drug discovery process. Four most important issues relevant to HTS are covered: a) concepts of pathway/phenotypic versus target-based screening, b) automation technologies, c) assay technologies, and d) data analysis.

Part I contrasts the two approaches of "Chemical Genetics" using pathway assays dependent on a initially not defined set of possible drug receptor sites and the target-based lead finding process commonly used in pharmaceutical research. Caroline Shamu uses case studies to introduce the basic concept, the assay technologies and selected results. Jörg Hüser and colleagues summarize the concepts of target-directed screening for lead discovery and discuss strengths and weaknesses of random/diversity screening when compared to knowledge-based *in silico* methodologies.

In **Part II**, John Comley provides a general overview of laboratory automation technologies covering assay carriers, liquid handling automats, signal detection instrumentation, and robotic integration.

Part III focuses on HTS assay technologies. For the discovery of novel lead candidates the choice of the appropiate assay technology and its technical realiza-

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tion will determine the overall quality of the screening experiment. There are two general appoaches for pharmacological assays: Assays measuring the binding of a candidate molecule to the target receptor ("binding tests") and assays monitoring the function of a target (or pathway) to visualize a possible modulation by small molecules. In chapter 4, Jörg Hüser and colleagues focus on principles of functional cell-based test systems. Improved readout techniques together with the rich molecular biology toolbox have rendered cell-based assays an important methodology for the targeted discovery of pharmacological lead compounds in HTS. Designed cell-based HTS assays ideally combine high specificity for and superior sensitivity towards the targeted receptor. In addition, measuring receptor function rather than binding allows one to monitor all possible drug-receptor interactions including allosteric modulation and reveals additional information on ligand efficacy, i.e. agonism or antagonism. Chapter 5 by William Mallender and colleagues gives an overview on functional biochemical tests, providing examples for the most important drug target classes approached by enzyme assays (proteases, kinases and others). In chapter 6, Peter Lipp and Lars Kästner introduce and critically discuss "Image-based High-content Screening", a recently emerging technology using subcellular imaging for target-based and pathway assays.

The last set of papers (**Part IV**) covers various aspects of HTS data analysis. Chapter 7, by Hanspeter Gubler, introduces concepts of HTS data management, assay quality assurance, and analysis. It touches on some fundamental statistical consideration relevant to handling large sets of compound activity data. Chapter 8 (Peter Nell and Stefan Mundt) illustrates the use of chemoinformatic tools, e.g. structural clustering of active compound sets, aiming to discriminate between specific hits and compounds acting through unspecific mechanisms and providing clues for preliminary SARs or pharmacophoric elements (i.e. molecular fragments contributing to activity). In chapter 9, Roger Smith and Nils Griebenow discuss pro's and con's of focused library screening, including methodologies and strategies to design such subsets of the available compound file and its use for lead discovery. The data analysis section is concluded by a paper by Jeremy Caldwell describing the consequent exploitation of large activity databases derived from functional cell-based screening by data mining technologies to reveal molecules acting through unexpected mechanisms.

The series editors are grateful to Jörg Hüser for his enthusiasm to organize this volume and to work with such a fine selection of authors. We believe that this book adds a fascinating new facet to our series on "Methods and Principles in Medicinal Chemistry". Last, but not least we thank the publisher Wiley-VCH, in particular Renate Dötzer and Dr. Frank Weinreich, for their valuable contributions to this project and to the entire series.

July 2006

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