Edited by Bernd Groner

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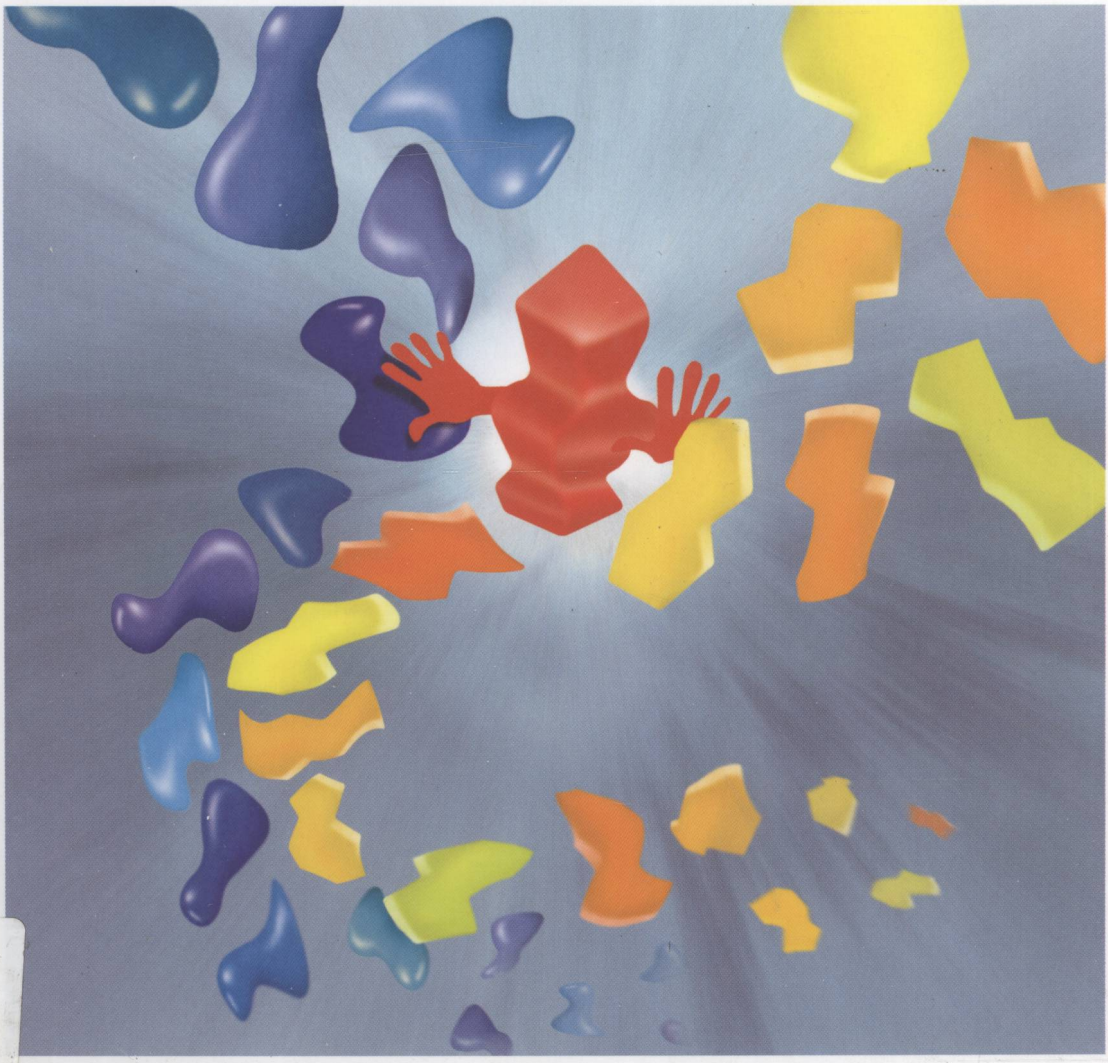
Peptides as Drugs

Discovery and Development



Peptides as Drugs

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Cover
The cover shows a cartoon illustration of two protein strands (in blue and yellow respectively) whose interaction is competitively inhibited by a peptide (in red).

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Further Reading

Sewald, N., Jakubke, H.-D.

Peptides: Chemistry and Biology

2009
ISBN: 978-3-527-31867-4

Jakubke, H.-D., Sewald, N.

Peptides from A to Z

A Concise Encyclopedia

2008
ISBN: 978-3-527-31722-6

Mannhold, R. (ed.)

Molecular Drug Properties

Measurement and Prediction

Volume 37 of Series "Methods and Pinciples in Medicinal Chemistry" edited by Mannhold, R., Kubinyi, H., Folkers, G.

2008
ISBN: 978-3-527-31755-4

Van de Waterbeemd, H. Testa, B. (eds.)

Drug Bioavailability

Estimation of Solubility, Permeability, Absorption and Bioavailability

Volume 40 of Series "Methods and Pinciples in Medicinal Chemistry" edited by Mannhold, R., Kubinyi, H., Folkers, G.

2009
ISBN: 978-3-527-32051-6

Ottow, E., Weinmann, H. (eds.)

Nuclear Receptors as Drug Targets

Volume 39 of Series "Methods and Pinciples in Medicinal Chemistry" edited by Mannhold, R., Kubinyi, H., Folkers, G.

2008
ISBN: 978-3-527-31872-8

Rognan, D. (ed.)

Ligand Design for G Protein-coupled Receptors

Volume 30 of Series "Methods and Pinciples in Medicinal Chemistry" edited by Mannhold, R., Kubinyi, H., Folkers, G.

2006
ISBN: 978-3-527-31284-9

Bertau, M., Mosekilde, E., Westerhoff, H. V.

Biosimulation in Drug Development

2008
ISBN: 978-3-527-31699-1

Fischer, J., Ganellin, C. R. (eds.)

Analogue-based Drug Discovery

2006
ISBN: 978-3-527-31257-3

Preface

The biological revolution of the past three decades has provided spectacular insights into the structure and function of macromolecules, cells and organisms, has changed our appreciation of the material basis of life, and has also provided the tools for targeted interferences with cellular processes. The technological advances which made this progress possible are based on, for example, molecular cloning, DNA sequencing, and the genetic manipulations of cells and organisms, and these methods have always preceded – or at least have accompanied – major conceptual advances.

Today, these successes in individual areas of biology are being extrapolated, and highly ambitious system biology projects have been initiated which will attempt to describe how the function of individual genes and the entirety of their interactions result in quantitative phenotypes of particular cells or organisms. The efforts to gain insights into quantitative behaviors are still restricted to simple model organisms and a few well-defined signal transduction pathways. Genetic manipulations are used to study information transmission through signaling systems, and to interpret them in the light of the resulting phenotypes. Interdisciplinary collaborations aim at identifying and predicting responses, and formulating new hypotheses concerning system functions.

Despite this remarkable progress in almost all areas of biology, reasonable predictions about gene functions in the organismal context remain very difficult. Whilst it is possible to annotate genes, and to recognize particular functional domains or subcellular localization signals, the cell-type specificity of expression, the regulation of expression, and the contribution of individual genes to particular phenotypic manifestations remain to be determined on an empirical basis. This has important implications for the development and use of drugs because, even if a drug can be developed to target a molecule that plays a central role in a particular pathological process, the adverse side effects of such a drug might still be unpredictable.

In life, if problems are very complex and the methods to approach them are still not fully developed, then there is a temptation to say "It can't be done". The Human Genome Project and targeted homologous recombination in embryonal stem cells are two such examples. But, in hindsight, advances in technologies tend to make the skeptic appear timid. Whilst the need to develop new drugs, to exploit additional drug targets and to meet medical needs is undisputed, many hurdles persist which

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might deter the use of a bold approach. One criterion which is most important for the decision makers is that of *oral bioavailability*. The design of small-molecular-weight compounds takes into consideration the "rule-of-five" ("Lipinski's rule of drug-likeness"), and this has had a profound influence on the thinking of medicinal chemists. Biologically active small molecules and drugs are many optimization steps apart, and sometimes even incompatible. Their pharmacokinetic properties clearly must be considered at an early stage in the development process, but this can also can stifle innovation. "Drug-likeness" and "druggability" represent two most valuable concepts, and the majority of the successful small-molecular-weight compounds are seen to adhere to certain rules which, if applied dogmatically, may tend to eliminate many possible target structures, even from only a theoretical consideration. It should be borne in mind that these rules are not absolute, and do not extend to biological substances – for example, in recent years monoclonal antibodies have become not only clinically successful but also commercially viable. Future drug discoveries will undoubtedly include more unconventional approaches, particularly in those cases where small-molecular-weight compounds with ideally defined properties have not yet been identified.

The versatility of protein functions and the power of peptides might represent a good starting point for considering some new options. Proteins are often composed of multiple functional domains which can operate independently of each other. Indeed, their autonomous functions are often based on the formation of distinct structures and conformations which are independent from any accompanying domains. Such domains can vary from 25 to 500 amino acids, and be stabilized with the help of metal ions or disulfide bridges. They also often embody functional units and act autonomously and, for this reason, can be taken out of context and recombined with other functional domains to yield new proteins with novel properties. Such characteristics of peptides can be exploited for practical purposes, notably the design of highly specific ligands and inhibitors. Peptide ligands may function as either agonists or antagonists, and in turn can influence protein conformations, protein interactions, or their DNA-binding properties. Upon the identification of a useful target structure, peptides recognizing this structure can be assembled into carriers or scaffolds and delivered as therapeutic proteins.

The design and production of *Peptides as Drugs* appears at first sight much like many of the seemingly insurmountable problems associated with novel approaches. Yet, this volume shows not only that such problems are currently being addressed, but also that many important contributions have in fact already been elucidated. It is inevitable that, in the foreseeable future, peptides will find their way into the drug repertoire, at which point "It can't be done" will be replaced by "It has been done". And, perhaps unsurprisingly, when this new class of drugs reaches the clinic and helps to improve existing therapies, then everybody will be convinced it was "... a good idea to begin with".

May 2009 *Bernd Groner*

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Contents

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Peptides as Drugs: Discovery and Development

Bernd Groner

> *"The necessity to exploit new drug targets and the suitability of peptides as drugs."*

1.1
Discovery of New Potential Drug Targets and the Limitations of Druggability

Complex networks of interacting proteins constitute the signaling pathways which mediate the intracellular propagation of biological information. Signals can originate from the cell surface and be relayed to sites in the cell where a biological response is triggered. The recognition of receptors by specific ligands is usually the initiating event which regulates cellular homeostasis, but also causes cellular responses such as proliferation, migration, angiogenesis, immune responses and cell death. The transient assembly of higher-order protein complexes, mediated by specific protein interaction domains and often regulated by secondary protein modifications, underlies the signaling mechanisms. In many instances platform proteins, for example, bring together enzymes and substrates and, in turn, recruit negative regulators which assure the transient nature of the signaling processes. The high complexity of these interactions makes them susceptible to disturbances arising from mutations in participating components, and the deregulation of specific protein–protein interactions has been recognized as the cause of diverse diseases. Conversely, the aberrant interaction of proteins is often a hallmark of diseased cells, and the inhibition of interactions required either to initiate or to maintain a particular disease state provides challenging opportunities for targeted therapies.

The importance of specific protein interactions has not only been recognized for diseases which originate from mutations in endogenous genes, but also extends to exogenous causes of disease and pathogenic microorganisms. Today, many disease-causing organisms and diseases are starting to be understood in molecular detail [1], with almost 600 microbial genomes having already been sequenced and 1800 others currently under investigation. This has led to the

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identification of virulence genes, metabolic pathways and cell-surface proteins as new targets for antimicrobial drug development and candidate vaccines. New directions are primarily set by technologies aimed at the elucidation of global gene expression patterns, and these high-throughput molecular profiling techniques have accelerated the discovery of drug targets. Genomics, transcriptomics and proteomics not only play a decisive role in the investigation of infectious diseases, but also are becoming increasingly important in the understanding of multigenic human diseases such as diabetes, heart diseases and cancer [2]. However, the task to integrate such global and descriptive analyses into manageable models has only just begun, and the large and unwieldy datasets available not only still preclude the rational prediction of gene functions in an organismal context, but also hamper predictions about the benefits and side effects of targeted drugs.

The present limitations concerning the evaluation and interpretation of datasets collected from global gene expression patterns are not deterring progress, however. Today, large-scale efforts are under way to gain insights into whole genome alterations that distinguish cancer cells from their normal counterparts [3, 4]. Cancer cells exhibit multiple genetic alterations in their DNA sequences, in the number of individual genes, and in their epigenetic DNA and histone modifications. These alterations cause both the activation and inhibition of biological events, interpretable in the context of the pathophysiology of cancer cells [5]. The comparison of the human genome which is present in normal, healthy cells with that present in breast, colon, pancreatic cancer cells and glioblastomas, has shown that hundreds of genes can be present in mutated forms. Although about 60 genes have been found to be altered in individual tumors, the mutations varied when individual tumors were compared. Initially, it seems difficult to distinguish molecular alterations which are causal and drive tumor-related phenotypes, such as cell proliferation, cell death, metabolism, metastasis, angiogenesis or immune evasion, and those which are correlative and do not contribute to tumor formation. Nevertheless, consistent patterns could be identified. The most important mutations affect a limited number of cellular signaling pathways. The suggestion is that, interference with these pathways – but not necessarily the targeting of mutated gene products – might represent the most promising approach to therapy [3].

Elucidation of the functions of many of mutated gene products supports the overriding importance of deregulated pathways. Both, oncogenes and tumor suppressor genes control crucial points in the cell cycle, transitions from a resting stage (G_0 or G_1) to a replicating phase (S), inhibit cell growth, and stimulate cell death when induced by cellular stress [6]. The cells continuously respond to 'prods' emanating from external and internal signals. The oncogene and tumor suppressor gene products are usually components of signaling cascades and are integrated in networks of protein interactions. These protein interactions are most frequently regulated through post-translational modifications [7]. Proteins such as histones, p53, RNA polymerase II, tubulin, Cdc25C and tyrosine kinases can be modified at multiple sites through phosphorylation, acetylation, methylation, ubiquitination, sumoylation, and citrullination. These modifications can act in a com-