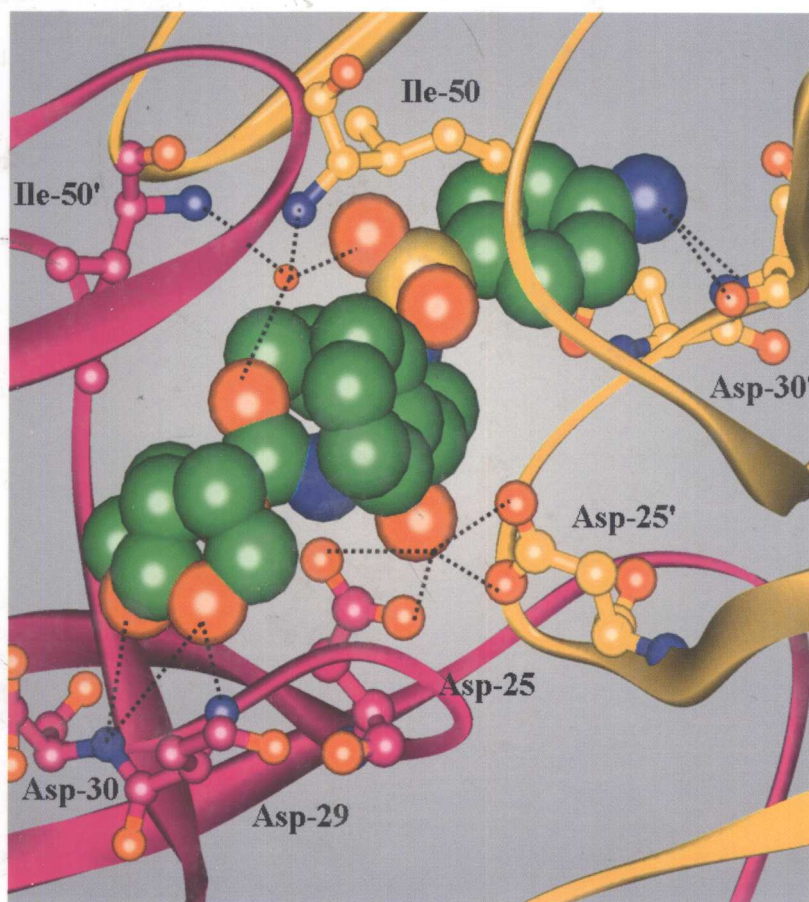


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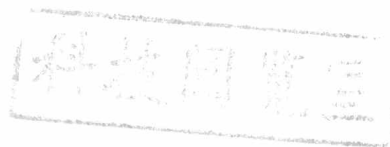
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Aspartic Acid Proteases as Therapeutic Targets



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X-ray structure of darunavir-bound HIV-1 protease (Tie, Y.; et al. 2004). Darunavir carbons are shown in green. Hydrogen bonds are shown as dotted lines.

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Preface

The digestive enzymes pepsin and chymosin (formerly called rennin), both being known since long, are members of the aspartic protease family. While these enzymes cleave many proteins and peptides in a relatively nonspecific manner, the substrate-specific aspartic protease renin converts angiotensinogen to angiotensin I, the precursor of the hypertensive peptide angiotensin II. For this purpose, research on renin inhibitors as potential antihypertensive drugs started in the early 1970s, based on Pauling's concept of transition-state mimics as effective enzyme inhibitors. Umezawa's discovery of pepstatin, a picomolar to nanomolar inhibitor of several aspartic proteases, confirmed the importance of a replacement of the scissile amide bond by such a transition state-imitating group. Some early peptide-like renin inhibitors showed nanomolar activities but lacked sufficient bioavailability, a problem that continued for the next decades. In the meantime, the search for aspartic proteases inhibitors was significantly stimulated by the discovery that HIV protease also belongs to the aspartic protease family, like pepsin and renin. Research on this new target was supported by the concept of chemogenomics, that is, by transferring successful design strategies to the new target. Based on the accumulated experience from the search for renin inhibitors, saquinavir, indinavir, and ritonavir resulted as first drugs in the mid-1990s. These peptide-like compounds were followed by some nonpeptidic analogues, all of them bearing a transition state partial structure. Finally, after thousands of man years in research in different companies, the first bioavailable renin inhibitor, aliskiren, was approved and marketed in 2007. Much effort and hope go now into the search for β - and γ -secretase inhibitors as drugs to prevent the development and progression of Alzheimer's disease, as well as plasmepsin inhibitors to treat malaria. This book by Arun K. Ghosh treats all these topics in much detail, starting from the function and physiological role of the corresponding protease, discussing its structural biology, and finally the medicinal chemistry of ligand and drug design.

For several years, it has been our goal to include monographs on therapeutically relevant targets and their modulators in our book series "Methods and Principles in Medicinal Chemistry." After volumes on G protein-coupled receptors (volume 24), voltage-gated ion channels (volume 29), ligand design for GPCRs (volume 30),

antitargets (volume 38), nuclear receptors (volume 39), epigenetic targets (volume 42), and drug transporters (volume 44), this monograph is the first one in an important family of therapeutically relevant enzymes, the aspartic proteases.

We would like to express our appreciation for Arun Ghosh for his enthusiasm and perseverance in bringing this project to fruition. We wish to thank Joseph Vacca for his initial efforts toward this goal. A team of leading scientists discusses the above-mentioned aspartic proteases, as well as some others, and their inhibitors. We are very grateful to all these authors for their excellent contributions, as well as to Frank Weinreich and Nicola Oberbeckmann-Winter for their ongoing engagement in our series "Methods and Principles in Medicinal Chemistry," in which this book will surely be as well accepted as all previous volumes.

April 2010

Raimund Mannhold, Düsseldorf
Hugo Kubinyi, Weisenheim am Sand
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A Personal Foreword

The human genome features a family of 15 active aspartic acid proteases that play a central role in the pathogenesis of many human diseases. The therapeutic inhibition of these aspartic acid proteases could conceivably provide a basis for novel treatment of human diseases. The disease targets include renin for hypertension, pepsin for gastric ulcers, HIV protease for AIDS, β - and γ -secretases for Alzheimer's disease, plasmepsins for malaria, cathepsin D and E for neoplastic diseases, and candida proteases for fungal infections. The catalytic aspartic acids in the active site motif are responsible for polarizing a bound water molecule that affects the proteolysis of the scissile bond of the substrate through a tetrahedral transition-state intermediate. Subsequently, mechanism-based design of nonhydrolysable dipeptide isosteres, mimicking the transition-state cleavage the scissile bond led to the evolution of potent substrate-derived inhibitors. Early studies on X-ray structures of fungal aspartic proteases bound to inhibitors, especially pepstatin-related structures, provided important molecular insights into the ligand-binding site interactions. This, in turn, provided important impetus for medicinal chemistry efforts toward the design and synthesis of a variety of transition-state isostere-derived peptidomimetic aspartyl protease inhibitors. Early efforts in the development of aspartic protease inhibitor drugs focused on the design of renin inhibitors for the treatment of hypertension. However, first-generation inhibitors were peptide-like, and due to their unfavorable pharmacological properties, early renin inhibitors never made it to clinical development. With the advent of AIDS and the discovery of human immunodeficiency virus (HIV) as the etiological agent, HIV-protease was recognized to play a critical role in the HIV life cycle. This led to massive research efforts in both academic and industrial laboratories in the quest for orally bioavailable protease inhibitor (PI) drugs as effective chemotherapeutics for AIDS. By the mid-1990s, the first aspartic acid protease inhibitor drug, saquinavir, received regulatory approval for the treatment of HIV/AIDS in combination with reverse transcriptase inhibitors. Soon after, a number of other first-generation HIV protease inhibitors were approved for use in highly active antiretroviral therapy (HAART). This was indeed a turning point in the management of HIV/AIDS. HAART treatment regimens significantly reduced mortality and improved the quality of life for HIV/AIDS

patients. However, the emergence of multidrug-resistant HIV strains required the search for a better long-term treatment with novel drugs. New-generation PIs have been developed to address the issue of drug resistance.

The clinical success in the realm of HIV protease inhibitor drugs stimulated an intense effort to find new inhibitor drugs for other aspartic protease targets. The search for a renin inhibitor drug was reinvigorated. The design, development, and approval of the first orally bioavailable renin inhibitor, aliskiren, marked the beginning of a new treatment for hypertension. This development has intensified further design and development of novel classes of renin inhibitors. The design of γ -secretase and β -secretase inhibitors for the treatment of Alzheimer's disease (AD) has also been a very active area of research in academic and pharmaceutical laboratories and several inhibitors are now in clinical development. Furthermore, drug design efforts against plasmepsins for malaria treatment are being actively made in many laboratories.

The X-ray crystal structures of numerous human aspartic acid proteases have now been determined at high resolution. In addition, detailed kinetic and subsite specificity studies provided in-depth knowledge of their catalytic mechanisms. These studies immensely contributed to the structure-based design and synthesis of a broad range of aspartic acid protease inhibitors for the treatment of many human diseases. Today, aspartic acid protease enzymes, as well as medicinal chemistry in the quest of novel inhibitor drugs, have become one of the most prolific areas of research.

This book is a comprehensive collection aimed at providing a thorough overview of the field of medically important aspartic acid proteases. It covers broad research areas with particular emphasis on the chemical and biological relevance of the targets, design of molecular probes, and problems and resolutions related to selectivity, toxicity, and oral bioavailability. Furthermore, it encompasses structure-activity studies, critical molecular insights from protein-ligand X-ray structures, and the development of various design tools. The first four chapters provide a general overview of aspartic acid proteases and their relevance to various disease targets. Chapters 5–9 describe the design, synthesis, X-ray structures, and clinical development of the first- and second-generation HIV-1 protease inhibitors for the treatment of HIV/AIDS. Chapter 9 provides a clinical perspective on protease inhibitors and their use in the treatment of HIV infection and AIDS. Chapters 10 and 11 cover the design and development of aliskiren as a first-in-class treatment of hypertension and current drug design efforts in renin inhibitors. The chemistry and biology of γ -secretase and inhibitors of γ -secretase for the treatment of AD are described in Chapters 12 and 13. The next four chapters address the emergence of β -secretase as a possible target for Alzheimer's disease intervention, drug design efforts, development of tools for selectivity, issues of blood-brain barrier penetration, and the design of extensive structural classes of inhibitors. The plasmepsins as antimalarial targets and related inhibitor design efforts are described in Chapters 18 and 19. The last chapter provides an overview of fungal aspartic acid proteases and their relevance as targets for antifungal agents.

This book is written by the foremost experts in the field of aspartic acid proteases. Each chapter is well illustrated and provides an up-to-date account of the subject materials. This book will be an excellent resource to medicinal chemists, biochemists, pharmacologists, and to those working in related fields. I am very grateful to all authors for their insightful contribution to this work. I hope that their efforts and research expertise will pave the way for further advancement in the aspartic acid protease research field. I very much enjoyed working with Dr. Frank Weinreich, Dr. Nicola Oberbeckmann-Winter, and the Wiley-VCH editorial team. I personally want to express my sincere appreciation for their help, support, toward the completion of this work. I would like to thank Dr. Joseph Vacca for his initial efforts and help on this book. I am grateful to Dr. Jordan Tang, my friend and collaborator, for help and suggestions. I would also like to thank my research colleagues, Dr. Bruno Chapsal, Dr. Sean Fyvie, Mr. Zach Dawson, and Mr. David Anderson for their help with proofreading of chapters and Ms. Heather Miller for her help with manuscript preparation and organization. Finally, I wish to thank my family, my wife Jody, and my three children for their love, support, and inspiration. They are my spirit.

January 2010

Arun K. Ghosh, Purdue University

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