Development of a Structure Prediction Method for G-Protein Coupled Receptors

Thesis by

Spencer E. Hall

In Partial Fulfillment of the Requirements

for the Degree of

Doctor of Philosophy

California Institute of Technology

Pasadena, California

2005

(Defended May 18, 2005)

Acknowledgments

First and foremost I have to thank my wife for her patience in being a student's wife for nine years, and her love and support during my years at school. I would also like to thank my parents for teaching me the value of hard work and the love of learning. I also have to thank my children for their boundless enthusiasm for their father going to school, especially my son Thomas who loves the fact that he now gets to go to school just like his father.

I must next thank Vaidehi Nagarajan for her encouragement, insight, and willingness to let me take her idea and work with it. Vaidehi was not only interested in my research, but in my growth as a student and scientist. I must also thank William Goddard III, my professor, for taking a mathematician as a chemistry graduate student and allowing me to work on a biochemistry project. Many a time, I would run out of possible solutions to a problem only to have Goddard propose four solutions that would all work and lead towards new avenues for research.

My thanks go to the members of the methods group that started with Rene and Wely, and now includes so many more like Peter Freddolino, Peter Huskey, Victor Kam, David Zhang, Jiyoung Heo, and others I am forgetting for their programming and input into the development of tools for MembStruk. I enjoyed the company and discussions with my office companions David Zhang and Yashar Kalani, who made spending time in the super air conditioned office fun. I also want to thank every member of the Biogroup who helped me find all the bugs and fine tune my software and methods.

Abstract

G-Protein Coupled Receptors (GPCRs) form a major target class of membrane proteins for therapeutic drug design, and the challenge is to design subtype specific drugs. Hence the knowledge of three-dimensional structure is critical to drug design for GPCRs. Since GPCRs are membrane bound proteins, there is only one crystal structure for a GPCR, namely bovine rhodopsin. The prediction of structure and function of G-protein-coupled receptors will allow for designing drugs with minimal side effects.

The focus of my thesis is the development of computational methods for prediction of structure of GPCRs and application of these methods (MembStruk) for a class of important drug targets such as chemokine receptors. MembStruk method is a hierarchical method ranging from coarse grain optimization of the trans-membrane helices to fine grain optimization of the structure in explicit lipid bilayer. The first two chapters of the thesis details the computational steps involved in MembStruk and its application to validating the method for bovine rhodopsin. The first chapter presents the method development in the most current version of the MembStruk method, version 4.30, and its application to bovine rhodopsin. The final predicted structure for bovine rhodopsin deviates from the crystal structure trans-membrane main chain atoms by 2.66 Å coordinate root mean square deviation (CRMSD), and the residues in the binding site of 11cis-retinal is only 1.37 Å CRMSD from the crystal structure for the main chain atoms. The second chapter of this thesis details the computational methods for optimization of the rotation and translation of the trans-membrane regions. These methods of rotation and translation of transmembrane helices has been further extended to the comparison of structures of two membrane proteins, and applied to the comparison

of crystal structures of bovine rhodopsin and bacteriorhodopsin. The third chapter details the graphical user interface that has been developed to automate the various steps of the MembStruk method.

Olfactory receptors are GPCRs and the molecular analysis for the recognition of odorants is very important in understanding the mechanism of olfaction. In a blind study prior to experiments, in collaboration with Dr. Bozza of Rockefeller University, I applied the MembStruk method to understanding the binding of odorants to rat and mouse olfactory receptor I7. Chapter 4 describes the application of the MembStruk method to rat and mouse I7 olfactory receptor and the binding of 65 odorants to this receptor. The last chapter describes the use of MembStruk method in predicting the structure and function of important drug targets, namely chemokine receptors CCR5 and CXCR4.

Table of Contents

Chapter 3: MembStruk GUI version 4.10

Chapter 4: Predicted 3D Structures for mouse I7 and rat I7 olfactory receptors and comparison of predicted odor recognition profiles with experiment

Appendix A: The method in detail used to build Rhodopsin

Appendix B: I7

List of Figures

Thesis: Introduction

Chapter 1

Chapter 2

Chapter 3

Chapter 4

Chapter 5

Appendix B

Appendix C

List of Tables

Chapter 4

List of Graphs

Chapter 3

Appendix A