Advances in PROTEIN CHEMISTRY

VOLUME 72 Peptide Solvation and H-Bonds



ADVANCES IN PROTEIN CHEMISTRY

Volume 72

Peptide Solvation and H-Bonds

This page intentionally left blank

ADVANCES IN PROTEIN CHEMISTRY

EDITED BY

FREDERIC M. RICHARDS

Department of Molecular Biophysics and Biochemistry Yale University New Haven, Connecticut DAVID S. EISENBERG

Department of Chemistry and Biochemistry Center for Genomics and Proteomics University of California, Los Angeles Los Angeles, California

JOHN KURIYAN Department of Molecular and Cellular Biology University of California, Berkeley Berkeley, California

VOLUME 72

Peptide Solvation and H-Bonds

EDITED BY

Robert L. Baldwin

Department of Biochemistry Beckman Center Stanford University Medical Center Stanford, California

David Baker

Department of Biochemistry University of Washington Seattle, Washington



AMSTERDAM • BOSTON • HEIDELBERG • LONDON NEW YORK • OXFORD • PARIS • SAN DIEGO SAN FRANCISCO • SINGAPORE • SYDNEY • TOKYO Academic Press is an imprint of Elsevier



Elsevier Academic Press 525 B Street, Suite 1900, San Diego, California 92101-4495, USA 84 Theobald's Road, London WC1X 8RR, UK

This book is printed on acid-free paper. \bigotimes

Copyright © 2006, Elsevier Inc. All Rights Reserved.

No part of this publication may be reproduced or transmitted in any form or by any means, electronic or mechanical, including photocopy, recording, or any information storage and retrieval system, without permission in writing from the Publisher.

The appearance of the code at the bottom of the first page of a chapter in this book indicates the Publisher's consent that copies of the chapter may be made for personal or internal use of specific clients. This consent is given on the condition, however, that the copier pay the stated per copy fee through the Copyright Clearance Center, Inc. (www.copyright.com), for copying beyond that permitted by Sections 107 or 108 of the U.S. Copyright Law. This consent does not extend to other kinds of copying, such as copying for general distribution, for advertising or promotional purposes, for creating new collective works, or for resale. Copy fees for pre-2006 chapters are as shown on the title pages. If no fee code appears on the title page, the copy fee is the same as for current chapters. 0065-3233/2006 \$35.00

Permissions may be sought directly from Elsevier's Science & Technology Rights Department in Oxford, UK: phone: (+44) 1865 843830, fax: (+44) 1865 853333, E-mail: permissions@elsevier.com. You may also complete your request on-line via the Elsevier homepage (http://elsevier.com), by selecting "Support & Contact" then "Copyright and Permission" and then "Obtaining Permissions."

For all information on all Elsevier Academic Press publications visit our Web site at www.books.elsevier.com

ISBN-13: 978-0-12-034272-3 ISBN-10: 0-12-034272-3

 PRINTED IN THE UNITED STATES OF AMERICA

 06
 07
 08
 09
 9
 7
 6
 5
 4
 3
 2
 1

Working together to grow libraries in developing countries www.elsevier.com | www.bookaid.org | www.sabre.org

ELSEVIER BOOK AID Sabre Foundation

NEW DIRECTIONS IN THE STUDY OF PEPTIDE H-BONDS	
AND PEPTIDE SOLVATION	ix
Potential Functions for Hydrogen Bonds in Protein Structure Prediction and Design	
Alexandre V. Morozov and Tanja Kortemme	
I. Introduction II. Physical Mechanism of Hydrogen Bond Formation III. Main Approaches to Modeling Hydrogen Bonds in	2 4
Biomolecular Simulations	6
V. Conclusions and Perspectives	20
References	30 32
Backbone–Backbone H-Bonds Make Context-Dependent Contributions to Protein Folding Kinetics and Thermodynamics: Lessons from Amide-to-Ester Mutations	
Evan T. Powers, Songpon Deechongkit, and Jeffery W. Kelly	
I. Introduction	40
II. Nomenclature and Synthesis of Amide-to-Ester Mutants	42
III. Esters as Amide Replacements	44
IV. Interpretation of Energetic Data from	
Amide-to-Ester Mutants	48
V. Amide-to-Ester Mutations in Studies of Protein Function	56
VI. Amide-to-Ester Mutations in Studies of Protein	
Folding Thermodynamics	58
VII. Analysis of $\Delta\Delta\Theta_{\rm b}$ and $\Delta\Delta\Theta_{\rm f}$ values from Amide to Ester Mutants	61
VIII Amide-to-Ester Mutations in Studies of Protein	01
Folding Kinetics	68
IX. Conclusions and Future Directions	69
References	70

Modeling Polarization in Proteins and Protein–Ligand Complexes: Methods and Preliminary Results

RICHARD A. FRIESNER

I.	Introduction	80
II.	Incorporation of Polarization in Molecular	
	Mechanics Models	81
III.	Aqueous Solvation Models for Polarizable Simulations	87
IV.	Modeling Polarizability with Mixed Quantum	
	Mechanics/Molecular Mechanics Methods	89
V.	Protein Simulations in Explicit Solvent Using a	
	Polarizable Force Field	94
VI.	Conclusion	98
	References	99

Hydrogen Bonds in Molecular Mechanics Force Fields

JAN HERMANS

I.	Introduction	105
II.	Geometric Deformation	106
III.	Nonbonded Interactions	111
IV.	Conclusion	116
	References	117

Resonance Character of Hydrogen-Bonding Interactions in Water and Other H-Bonded Species

F. WEINHOLD

I.	Introduction	122
II.	Natural Bond Orbital Donor-Acceptor Description	
	of H-Bonding	125
III.	Quantum Cluster Equilibrium Theory of	
	H-Bonded Fluids	131
IV.	Recent Experimental Advances in Determining Water	
	Coordination Structure	138
V .	General Enthalpic and Entropic Principles of H-Bonding	141

VI.	Hydrophobic Solvation: A Cluster Equilibrium View	145
VII.	Summary and Conclusions: The Importance of	
	Resonance in H-Bonding and Its Possible Representation	
	by Molecular Dynamics Simulations	149
	References	150

How Hydrogen Bonds Shape Membrane Protein Structure

STEPHEN H. WHITE

Introduction	157
Structure of Fluid Lipid Bilayers	159
Energetics of Peptides in Bilayers	160
Helix-Helix Interactions in Bilayers	165
Perspectives	167
References	167
	Introduction Structure of Fluid Lipid Bilayers Energetics of Peptides in Bilayers Helix–Helix Interactions in Bilayers Perspectives References

Peptide and Protein Folding and Conformational Equilibria: Theoretical Treatment of Electrostatics and Hydrogen Bonding with Implicit Solvent Models

WONPIL IM, JIANHAN CHEN, AND CHARLES L. BROOKS, III

I.	Introduction	174
II.	Generalized Born (GB) Models	176
III.	Peptide Folding and Conformational Equilibria	184
IV.	Concluding Discussion	190
	References	192

Thermodynamics of α -Helix Formation

George I. Makhatadze

I.	First 50 Years of Study of the Thermodynamics of the	
	Helix-Coil Transition	199
II.	The Quest for Enthalpy of the Helix–Coil Transition	205
III.	Temperature Dependence of Enthalpy of the	
	Helix-Coil Transition	213

vii

IV.	Thermodynamic Helix Propensity Scale: Importance of	
	Peptide Backbone Hydration	215
V .	Other Instances When Peptide Backbone Hydration is	
	Important for Stability	216
VI.	Future Directions	218
	References	220

The Importance of Cooperative Interactions and a Solid-State Paradigm to Proteins: What Peptide Chemists Can Learn from Molecular Crystals

J. J. DANNENBERG

I. Introduction	228
II. Similarities and Differences Between Proteins/Peptides	
and Molecular Crystals	229
III. The Importance of H-Bond Cooperativity in	
Molecular Crystals	231
IV. Structural Consequences of H-Bond Cooperativity in	
Molecular Crystals	234
V. How Does the Use of the Crystal Paradigm Affect	
Protein/Peptide Study?	240
VI. Are H-Bonds Electrostatic?	242
VII. How Strong are Peptide H-Bonds?	243
VIII. Comparison with Experimental Data from Studies	
in Solution	255
IX. The Importance of a Suitable Reference State(s)	257
X. How Protein Chemists Can Deal with Problems Posed by	
Dual Paradigms	260
XI. Water, the Hydrophobic Effect and Entropy	263
XII. Concluding Remarks	267
References	267
AUTHOR INDEX.	275
SUBJECT INDEX	291

viii

NEW DIRECTIONS IN THE STUDY OF PEPTIDE H-BONDS AND PEPTIDE SOLVATION

There are two main reasons for the rapid growth of research into the energetics of peptide H-bonds and peptide solvation: to help solve the problem of protein structure prediction and to complete the quantitation of the energetics of folding. For a long time protein chemists have accepted that the hydrophobic interaction (burial of nonpolar surface area through folding) is the major source of free energy driving folding, as proposed by Kauzmann (1959), but there has always been a nagging doubt about the role of peptide H-bonds. Even if each H-bond contributes only 0 ± 1 kcal/mol, which is sometimes used as a guesstimate, approximately 70% of the peptide groups in globular proteins make H-bonds (Stickle et al., 1992) and a contribution of ± 70 kcal/mol to ΔG for folding of a 100 residue protein would make an enormous difference to its stability. The ΔH for forming an alanine peptide helix (-0.9 \pm 0.1 kcal/mol per residue) has been measured accurately by titration calorimetry and it must arise from the peptide H-bond, not from burial of nonpolar surface, because of its very small ΔCp (see chapter by Makhatadze). Thus, peptide H-bonds may make a substantial favorable contribution to the enthalpy of protein folding. Moreover, every H-bonding group must make a H-bond, either within the folded protein or else to water, because the penalty for burying a free H-bonding group is large (~6 kcal/mol; Fleming and Rose, 2005). Thus, the drive for continued rapid progress in protein structure prediction (Kuhlman et al., 2003), which requires a fuller understanding of protein-folding energetics, brings peptide H-bonds and peptide solvation into central focus.

Three chapters of this volume, by Dannenberg, Morozov and Kortemme, and Weinhold, deal with the problem of using quantum mechanics to represent H-bonds. Dannenberg reviews the lessons learned from analyzing chains of H-bonds in molecular crystals of small molecules such as formamide and urea. Morozov and Kortemme discuss the properties of H-bonds seen in protein structures and compare them to properties predicted by quantum mechanics. Weinhold tackles the structure of water by performing quantum mechanics on defined clusters of water molecules. These three chapters reach a common conclusion, namely that the long-standing electrostatic model of H-bond formation, which predicts linear H-bonds that are formed noncooperatively, is too simple and the H-bonds found in proteins, as well as in water clusters, are partly bent. The molecular mechanics force fields used commonly to simulate protein structures and dynamic behavior have fixed partial atomic charges consistent with the older electrostatic model of H-bonds. Friesner tackles the problem of making a force field that can adjust to the newer view of H-bonds by allowing the partial charges on atoms to vary through induced polarization. He reports that the first results of using this force field to represent protein docking reactions are promising as regards the H-bonds formed. Hermans comments on the recent development of a method for performing quantum mechanics on an entire protein and discusses issues raised in the three other chapters, such as whether Weinhold's "new view" of water structure can be reconciled with the older view of water–water H-bonds resembling those found in ice. Im, Chen, and Brooks provide a tool for rapid calculation of peptide solvation in proteins as they review computation of protein electrostatics by use of generalized Born methods. Solvation of the polar peptide group is electrostatic in character and may be predicted by electrostatic algorithms, provided the protein or peptide structure is known accurately.

Each of three experimental chapters deals with a recent experimental method of investigating peptide H-bonds and peptide solvation. Powers, Deechongkit, and Kelly review the first energetic results of making mutations (amide to ester) that eliminate peptide H-bonds. Makhatadze summarizes recent calorimetric studies of peptide helix formation and considers their implications for the energetics of protein folding. White discusses the energetics of shaping membrane proteins based on the results of inserting peptides into lipid bilayers. The twin problems of making H-bonds and accounting for any free H-bonding groups take on a new character when the newly formed protein resides in a nonaqueous environment. Valuable lessons can be learned about the nature of folding energetics in aqueous solution.

This list of chapters raises some questions, the most evident one being: why are peptide H-bonds treated here by quantum mechanics or by introducing a new force field while peptide solvation is handled as a problem in electrostatics? Why is not peptide solvation treated as the problem of making H-bonds between water molecules and free peptide -NH and -C=O groups? There are various answers to this question, the first being that probably quantum mechanics will soon be used to study H-bonds between water and peptide -NH and -C=O groups. Weinhold looks ahead to possible methods of tackling this problem. A second answer is that if liquid water is considered to be a giant network of H-bonded water molecules, then it is just too big for present methods of performing quantum mechanics. Weinhold suggests one approach for breaking this problem into parts. A third answer is that backbone electrostatics, which result from the large partial charges on the atoms of the peptide -NHand -C=O groups, are important in other problems besides peptide solvation, notably in the docking reactions of proteins with small ligands, other proteins, and nucleic acids. Note that amide solvation is a directly measurable quantity for small amides, as demonstrated by the pioneering work of Wolfenden (1978), and the solvation of polar groups in small molecules has been treated successfully as an electrostatic problem. The parameters of the electrostatic algorithm DelPhi are calibrated specifically to reproduce the solvation free energies of polar small molecules (Sitkoff *et al.*, 1994). When DelPhi is used to investigate how peptide solvation depends on backbone conformation and on the presence of specific neighboring residues in short peptides, surprising and interesting results are found (Avbelj and Baldwin, 2004).

As noted earlier, the theoretical chapters on representing H-bonds in proteins, as well as in water clusters, agree that a paradigm shift is under way. Morozov and Kortemme trace the evolution of the change in viewpoint. Linear electrostatic H-bonds, formed by atoms with fixed partial charges, are very convenient for rapid computation using standard force fields and the paradigm shift will not take place without a struggle. That peptide H-bonds are linear was assumed by Pauling and co-workers (1951) in their landmark paper predicting the structure of the α -helix. It is sometimes said that the business of a scientist is to introduce new ideas into the field. Readers of this volume will find that long-standing and basic assumptions of structural biology are being challenged.

Acknowledgment

I thank Jan Hermans for discussion.

Robert L. Baldwin

References

- Avbelj, F. A., and Baldwin, R. L. (2004). Origin of the neighboring residue effect on peptide backbone conformation. *Proc. Natl. Acad. Sci. USA* 101, 10967–10972.
- Fleming, P. J., and Rose, G. D. (2005). Do all backbone polar groups in proteins form hydrogen bonds? *Protein Sci.* 14, 1911–1917.
- Kauzmann, W. (1959). Factors in interpretation of protein denaturation. Adv. Protein Chem. 14, 1–63.
- Kuhlman, B., Dantas, G., Ireton, G. C., Varani, G., Stoddard, B. L., and Baker, D. A. (2003). Design of a novel globular protein fold with atomic-level accuracy. *Science* **302**, 1364–1368.
- Pauling, L., Corey, R. B., and Branson, H. R. (1951). The structure of proteins: Two hydrogen-bonded helical configurations of the polypeptide chain. *Proc. Natl. Acad. Sci. USA* 37, 205–211.

- Sitkoff, D., Sharp, K. A., and Honig, B. (1994). Accurate calculation of hydration free energies using macroscopic solvent models. J. Phys. Chem. 98, 1978–1988.
- Stickle, D. F., Presta, L. G., Dill, K. A., and Rose, G. D. (1992). Hydrogen bonding in globular proteins. J. Mol. Biol. 226, 1143–1159.
- Wolfenden, R. (1978). Interaction of the peptide bond with solvent water: A vapor phase analysis. *Biochemistry* **17**, 201–204.

POTENTIAL FUNCTIONS FOR HYDROGEN BONDS IN PROTEIN STRUCTURE PREDICTION AND DESIGN

By ALEXANDRE V. MOROZOV* AND TANJA KORTEMME[†]

*Center for Studies in Physics and Biology, Rockefeller University, New York, New York 10021; [†]Department of Biopharmaceutical Sciences and California Institute for Quantitative Biomedical Research, University of California, San Francisco, San Francisco, California 94142

I.	Introduction	39
II.	Physical Mechanism of Hydrogen Bond Formation	4
III.	Main Approaches to Modeling Hydrogen Bonds in Biomolecular	
	Simulations	6
	A. Potentials Derived from Hydrogen Bonding Geometries Observed	
	in Crystal Structures	6
	B. Molecular Mechanics: Comparison with the Structure-Derived,	
	Orientation-Dependent Potential	9
	C. Quantum Mechanics: Comparison with Molecular Mechanics and the	
	Structure-Derived Potential	12
IV.	Applications of Hydrogen Bonding Potentials	20
	A. Protein Structure Prediction and Refinement	20
	B. Prediction of Protein–Protein Interfaces	24
	C. Protein Design	27
V.	Conclusions and Perspectives	30
	References	32

Abstract

Hydrogen bonds are an important contributor to free energies of biological macromolecules and macromolecular complexes, and hence an accurate description of these interactions is important for progress in biomolecular modeling. A simple description of the hydrogen bond is based on an electrostatic dipole–dipole interaction involving hydrogendonor and acceptor–acceptor base dipoles, but the physical nature of hydrogen bond formation is more complex. At the most fundamental level, hydrogen bonding is a quantum mechanical phenomenon with contributions from covalent effects, polarization, and charge transfer. Recent experiments and theoretical calculations suggest that both electrostatic and covalent components determine the properties of hydrogen bonds. Likely, the level of rigor required to describe hydrogen bonding will depend on the problem posed. Current approaches to modeling hydrogen bonds include knowledge-based descriptions based on surveys of hydrogen bond geometries in structural databases of proteins and small molecules, empirical molecular mechanics models, and quantum mechanics-based electronic structure calculations. *Ab initio* calculations of hydrogen bonding energies and geometries accurately reproduce energy landscapes obtained from the distributions of hydrogen bond geometries observed in protein structures. Orientation-dependent hydrogen bonding potentials were found to improve the quality of protein structure prediction and refinement, protein–protein docking, and protein design.

I. INTRODUCTION

Accurate modeling of hydrogen bonding interactions is critical for progress in protein structure prediction, protein-protein docking, and protein design. While the large number of hydrogen bonds in proteins and protein interfaces underlines their importance, there may be no net gain in free energy for hydrogen bond formation in protein folding and binding; the formation of hydrogen bonds between protein atoms results in the loss of hydrogen bonds made with water. Most polar groups in the protein interior form hydrogen bonds to satisfy their hydrogen bonding potential (Baker and Hubbard, 1984; McDonald and Thornton, 1994). These requirements result in considerable energetic and structural constraints and are in part responsible for the regular backbone-backbone hydrogen bonding patterns of α -helix and β -sheet regular secondary structure elements (Pauling and Corey, 1951). Similarly, hydrogen bonds, particularly side chainside chain hydrogen bonds, are thought to play important roles in the specificity of macromolecular interactions (Lumb and Kim, 1995; Petrey and Honig, 2000) and need to be taken into account in the prediction of protein interaction preferences. Hydrogen bonds may be crucial for enabling a unique three-dimensional protein conformation or binding mode in protein design applications (Looger et al., 2003; Lumb and Kim, 1995).

What is needed for an accurate description of hydrogen bonding interactions within and between proteins? The physical nature of hydrogen bonds is complex, and calculation of electrostatics, polarization, exchange repulsion, charge-transfer, and coupling contributions to hydrogen bonding energetics (Kollman, 1977; Morokuma, 1971; Singh and Kollman, 1985; Umeyana and Morokuma, 1977) from first principles is not straightforward for biological macromolecules. Likely, the level of rigor required to explain certain molecular properties in question will depend on the problem posed. Which simplifications can be made in which context? An example discussed in detail in this chapter is the orientation dependence of hydrogen bonds, which has been a subject of considerable debate. An electrostatic dipole–dipole model of a hydrogen bond would predict a linear arrangement of the donor and acceptor dipoles. However, a "lone pair" concept would imply directionality of the hydrogen bond (Fig. 1a). What are the structural and energetic characteristics of hydrogen bonds in protein structures and how can a model be devised that reproduces them?

Any simplified description of hydrogen bonds in biological molecules needs to be tested by comparing its predictions against a large body of experimental data, preferably obtained from macromolecules. A direct comparison of predicted and observed hydrogen bonding energies in biological macromolecules is not straightforward because the individual components of the free energy cannot readily be measured independently in experiments. More feasible but less direct strategies rely on the vast information available on protein sequences and structures and use concepts from computational protein design, protein structure prediction, and protein–protein docking. The structure prediction and docking tests



FIG. 1. Mechanism and orientation dependence of hydrogen bond formation. A, acceptor; D, donor; H, hydrogen; AB, acceptor base. (a) Orientation dependence of hydrogen bond formation. Hydrogen bond formation along lone-pair directions would predict hydrogen bonding geometries such as the one shown on the left, whereas an electrostatically dominated mechanism based on a dipole–dipole interaction (see b) would favor the linear arrangement on the right. (b) Simple description of hydrogen bonding interactions as the interaction of two dipoles with atom-centered partial point charges. Shaded spheres represent electron density at H and AB shifted along the H–D and AB–A covalent bonds toward more electronegative atoms, resulting in the appearance of partial charges on all four atoms. (c) Schematic representation of hydrogen bond geometry. D, donor atom; H, hydrogen atom; A, acceptor atom; AB, acceptor base; R₁, R₂, atoms bound to the acceptor base. Geometric parameters used here to describe hydrogen bonds are as follows: δ_{HA} (Å), distance between hydrogen and acceptor atom; X (degree), angle at the acceptor atom; θ (degree), angle at the hydrogen atom; X (degree), dihedral angle around the A–AB axis.