TOWARD DEVELOPING A THERMODYNAMIC MODEL OF BINDING-INDUCED CONFORMATIONAL TRANSITIONS IN SHORT, DISORDERED PROTEIN REGIONS

Justin Drake, University of Texas Medical Branch
2016–2017 Graduate Fellow

EXECUTIVE SUMMARY

Intrinsically disordered regions (IDRs) in proteins are highly dynamic and often undergo conformational transitions upon binding a protein target. The thermodynamics underlying these transitions, particularly conformational entropy and its contribution to protein binding, remain elusive. In this work, we use molecular dynamics simulations to sample the conformational states of a model disordered polypeptide as a function of chain length. Backbone dihedral angles for each polypeptide were measured from the simulation trajectories and used to calculate the conformational entropy using the Quasi-Harmonic Analysis (QHA), Boltzmann Quasi-Harmonic (BQH) Analysis, and two variants of the Mutual Information Expansion (MIE) method. Our results suggest that short IDRs may provide a significant source of free energy that proteins may tap through order-disorder transitions to modulate or regulate protein binding. We are currently working to develop a model that more fully describes the energetics of these conformational transitions and the extent to which they contribute to protein binding processes.

RESEARCH CHALLENGE

For much of the twentieth century, it was widely held that the three-dimensional structure of a protein dictates its cellular function. While true for a large group of proteins, at the turn of the twenty-first century, seminal papers (1, 2) opened the door to an entirely new class of proteins that rely on highly dynamic, flexible (i.e., disordered) regions to carry out their functions. These
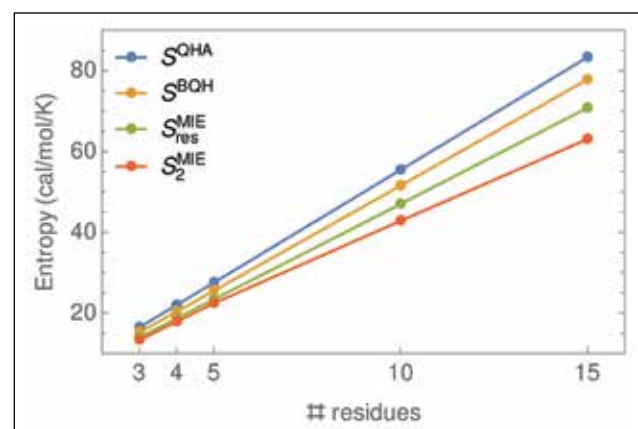

Figure 1: Conformational entropy calculated from the dihedral angles of successively longer oligoglycine polypeptides using the QHA, BQH, and two variants of the MIE method.

intrinsically disordered regions (IDRs) found within proteins play an important role in protein-signaling networks and regulation and have been implicated in a number of diseases (1, 2). Upon binding a target protein, IDRs often undergo conformational transitions to a more ordered or disordered state and may directly interface with the target or be located distally to the binding interface (3, 4). These conformational transitions involve relatively short IDRs on the order of ~10 residues (4). To successfully target drugs to IDRs or genetically engineer IDRs with certain therapeutic properties to treat various diseases, we need a complete understanding of the thermodynamics associated with binding-induced conformational transitions of short IDRs.

METHODS & CODES

Conformational entropy (i.e, a proxy for disorder) of IDRs associated with order-disorder transitions is believed to play an important role in protein-binding processes (3). However, it is notoriously difficult to measure or approximate conformational entropy using traditional solution biophysics techniques. In this work, we use all-atom molecular dynamics simulations to sample the conformational states of successively longer oligoglycine polypeptides (Gly$_N$ where N = 3, 4, 5, 10, and 15 residues), and use a number of methods to calculate the dihedral angle contribution to the conformational entropy. Oligoglycine is an ideal protein backbone model and is found in varying lengths in a number of IDRs. Simulations were performed with the NAMD (5) molecular dynamics (MD) package and Amber ff12SB (6) force field. Backbone dihedral angles for each model were measured from the simulation trajectories and used to calculate the conformational entropy using QHA (7), BQH (8), and two variants of the MIE (9) method.

RESULTS & IMPACT

The structural and thermodynamic properties of IDRs depend on chain length. In this work, we have calculated the conformational entropy (S) of an oligoglycine model as a function of chain length using four methods that account in different ways for the reduction in entropy due to correlated dihedral motions. Fig. 1 shows that conformational entropy scales remarkably linearly with chain length, with slopes ranging between 4.1 and 5.6 cal/mol/K per residue. This is consistent with the experimentally measured loss of backbone entropy (4.5 cal/mol/K per residue) upon protein folding (10). S^{QHA} is a strict upper bound on the conformational entropy. S_{res}^{MIE} and S_2^{MIE} differ in that the former accounts only for

dihedral angle correlations within each residue, whereas the latter considers all possible pairs of dihedral angles along the backbone, yielding a slight reduction in the slope of S_2^{MIE} (4.1 cal/mol/K/residue) compared to S_{res}^{MIE} (4.7 cal/mol/K/residue). We note that S_{res}^{MIE} is the simplest and most computationally efficient method of the four. Coupling of the dihedral angles within each residue appears to be the most significant source of correlations affecting the conformational entropy (Fig. 2). Preliminary data also suggest that oligoglycine conformational entropy is relatively insensitive to constraints on end-to-end distance, radius of gyration, and solvent-accessible surface area.

At a temperature of 300°K, the conformational entropy of these short IDR models (e.g., ~21 kcal/mol for Gly^{15}) provides a significant free energy reservoir that proteins may tap through order-disorder transitions to modulate or regulate protein binding. We will incorporate our current results with those reported in last year's Blue Waters Annual Report on the solvation thermodynamics of oligoglycines to create a more complete model of the thermodynamic contribution to protein binding energetics associated with these conformational transitions. We will then evaluate this current model in the context of biologically relevant examples.

WHY BLUE WATERS

Statistically converged estimates of conformational entropy require extensive sampling of the conformational states of a protein or polypeptide during molecular dynamics (MD) simulations. Access to GPU-optimized MD packages such as NAMD for use with Blue Waters XK nodes, assistance from the project staff to develop an efficient workflow, and the high-throughput of Blue Waters was critical to performing our research in a tractable amount of time. Additionally, the Blue Waters Graduate Fellowship provided me the opportunity to successfully propose, manage, and partially fund my doctoral research, and expanded my understanding of high-performance computing. These experiences are instrumental in my pursuit of becoming an independent researcher.

PUBLICATIONS AND DATA SETS

Ou, S.C., J. A. Drake, and B. M. Pettitt, Nonpolar Solvation Free Energy from Proximal Distribution Functions. *J. Phys. Chem. B*, 121 (2017), pp. 3555–3564.

Drake, J.A., R. C. Harris, and B. M. Pettitt, Solvation Thermodynamics of Oligoglycine with Respect to Chain Length and Flexibility. *Biophysical Journal*, 111 (2016), pp. 756–767.
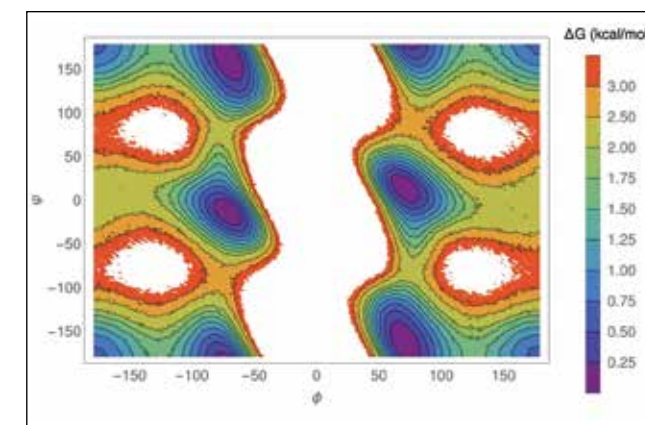
Drake, J.A., and B.M Pettitt, Solvation thermodynamics of oligoglycine: implications for aggregation and collapse. The *4th Annual NCSA Blue Waters Symposium* (Sunriver, Ore., June 12–15, 2016).

Drake, J.A., and B.M. Pettitt, Solvation thermodynamics of oligoglycine with respect to chain length and flexibility: implications for aggregation and collapse. *72nd Annual American Chemical Society Southwest Regional Meeting: Innovations in Energy and Medicine* (Galveston, Tex., November 10–13, 2016).

Drake, J.A., and B.M. Pettitt, The protein backbone: how structural and thermodynamic properties scale with length. *251st American Chemical Society National Meeting and Exposition* (San Diego, Calif., March 13-17, 2016).

Drake, J.A., and B.M. Pettitt, The protein backbone: how structural and thermodynamic properties scale with length. *21st Annual Structural Biology Symposium* (Sealy Center for Structural Biology and Molecular Biophysics, Galveston, Tex., April 23, 2016).

Drake, J.A., and B.M. Pettitt, Protein Backbone Thermodynamics: Towards Estimating Conformational Entropy. *21st Annual Structural Biology Symposium* (Sealy Center for Structural Biology and Molecular Biophysics, Galveston, Tex., April 23, 2016).

Drake, J.A., and B.M. Pettitt, Protein Backbone Thermodynamics: Towards Estimating Conformational Entropy. *20th Annual Structural Biology Symposium* (Sealy Center for Structural Biology and Molecular Biophysics, Galveston, Tex., May 2, 2015).

Figure 2: Representative free energy map of the two backbone dihedral angles (φ, ψ) in a glycine residue highlights correlations between the two. The free energy is color coded and relative to the most populated dihedral pair state. The underlying pair probability distributions of the map are used in MIE entropy calculations.

A sixth-year doctoral student in biochemistry and molecular biology at the University of Texas Medical Branch at Galveston, Justin Drake is working under the direction of B. Montgomery Pettit. He expects to graduate in December 2017.