Our research program focuses on the developments and applications of theoretical/computational methods to chemical and physical problems in biology. Currently active research topics, which are further elaborated below, are:

1. Protein/Peptide Interactions in Biological Membranes
2. Membrane Protein NMR Structure Calculation/Refinement
3. Modeling and Simulation of Glycoconjugates
4. Bacterial Outer Membranes and Interactions with Proteins
5. CHARMM-GUI / ST-analyzer Development

1. Protein/Peptide Interactions in Biological Membranes

Understanding the delicate balance of forces governing helix-helix and helix-lipid interactions of transmembrane (TM) proteins is central to understanding membrane structure and function. These membrane constituent interactions form the basis for many vital cellular processes, including TM-induced signaling, transport of ions and small molecules, energy transduction, and cell-cell recognition. My lab has published novel methods and their applications that aim to address the complicated energetics and molecular mechanisms of helix-lipid and helix-helix interactions at the atomic level. With these initial accomplishments, we will continue to address (1) *hydrophobic mismatch (helix-lipid interactions)*, (2) *helix-helix interactions in membranes*, (3) *influence of helix tilting on ion channel gating*, (4) *TM-induced signaling* (**Figure 1**), and (5) *influence of TM peptide dynamics on experimental observables*, which can complement experimental efforts on these important biomedical systems.

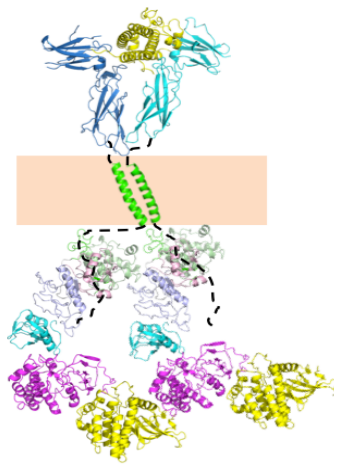

Figure 1. The model of a hGHR on the membrane surface (*orange*).

2. Membrane Protein NMR Structure Calculation/Refinement

Membrane proteins with one or a few TM helices are abundant and often involved in important TM-induced signaling and regulation through conformational changes of TM domains and/or formation of hetero- and homo-oligomers. The structure determination of these membrane proteins provides invaluable insights into their unique functions, but it is still challenging to obtain such structural information in bilayer environments. In addition to conventional NOE-based NMR methods, measuring orientational restraints is an emerging technique to overcome some critical problems existing in the conventional methods. Such orientational restraints are residual dipolar coupling (RDC) in solution NMR, and ^{15}N chemical shift anisotropy (σ) and ^{1}H-^{15}N dipolar coupling (ν) in solid-state NMR (ssNMR).

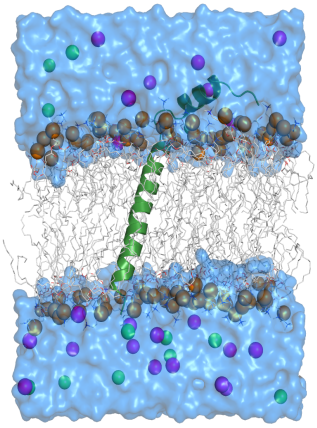

Figure 2. Simulation system of Pf1 coat protein in an explicit DOPC/DOPG bilayer. Protein is shown in cartoon, lipids are in sticks, lipid phosphorus atoms (orange) and ions in spheres, and water in surface.

We have published novel restraint potentials for RDC, σ, and ν in order to translate such restraints into protein structures. We will continue to use the RDC and ssNMR restraint potentials in the context of *ensemble dynamics* to further explore structural heterogeneity and dynamics embedded in such experimental observables. In addition, we also continue to use various NMR

restraints to perform structure refinement of membrane proteins in a realistic membrane environment (**Figure 2**) to obtain not only the structural information, but also protein-lipid interaction and protein dynamics, which are closely related with the protein's function.

3. Modeling and Simulation of Glycoconjugates

As one of the four fundamental classes of macromolecules (along with nucleic acids, amino acids, and lipids) that comprise living systems, glycans come in a diversity of sequences and structures by linking individual sugar units in a multitude of ways. Understanding the impact of glycosylation on structure, dynamics, and function of proteins and lipids is an emerging and challenging problem in biology. Given the increasing availability of glycosylation information by mass spectrometry, as well as the difficulties in characterizing structure and dynamics of glycoconjugates, there is a pressing need for reliable computer modeling and simulation. Our long-term goal is to develop a unified toolset for glycoconjugate modeling and simulation and to apply it to biologically important glycoconjugates. We have made efforts to build glycan fragment database for the survey of the PDB glycan structures (**Figure 3**) and to develop a fragment-based glycan modeling toolset. The toolset and databases will be freely available through http://www.glycanstructure.org, our glycan web portal.

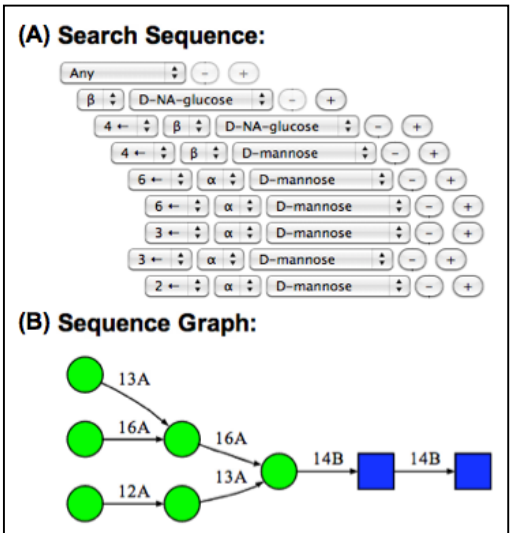

Figure 3. (A) An intuitive search tool based on a glycan sequence. (B) The sequence graph is automatically updated as sugars are added or removed in (A).

In particular, we are interested in roles of glycans in glycan-binding proteins (GBPs) in three different areas: (1) What are the roles of glycans in protein-protein interactions; (2) Are there any specific, well-defined interactions between glycans and GBPs? If so, can we use such protein binding sites to detect the potential GBPs; (3) What are the structures and dynamics of glycolipids in membrane bilayers? We are getting some exciting preliminary results on these questions and will continue to address these problems by PDB structure survey, modeling, and simulations.

4. Bacterial Outer Membranes and Interactions with Proteins

Because the bacteria's outer membrane (OM) acts as an effective barrier against the permeation of both hydrophobic and hydrophilic compounds, gram–negative cell permeation is the biggest challenge to the discovery of novel antibiotics for bacterial infections and antibiotic resistance worldwide. The bacterial OM is a unique and highly asymmetric lipid bilayer composed of phospholipids in the inner leaflet and lipopolysaccharide (LPS) in the outer leaflet (**Figure 4**). Despite the direct relationship of gram-negative bacteria to public health, our molecular-level understanding of how the bacterial OMs behave and work for various types of bacteria, how membrane proteins behave in the OMs, and how known drug molecules and potential drugs can enter through the OMs is rudimentary at best. We aim to address these questions by all-atom modeling and simulation of the bacterial OMs and proteins. We believe that we are in a good position to conduct this project because we have all the techniques in hand that are necessary to build complex *in silico* OM-protein systems.

Figure 4. The cover image for this issue illustrates a typical *E. coli* outer membrane and the molecular system used to represent that complexity in molecular dynamics simulations.

5. Protein-Ligand and Protein-Protein Interaction

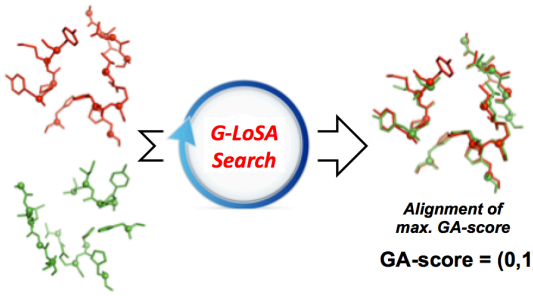

Figure 5. G-LoSA local structure alignment.

Molecular recognition by protein mostly occurs in a local region on the protein surface. Thus, an efficient computational method for accurate characterization of protein local structural conservation is necessary to better understand biology and drug design. We have developed a novel local structure alignment tool, G-LoSA (Graph-based Local Structure Alignment). G-LoSA aligns protein local structures in a sequence order independent way and provides a GA-score, a chemical feature-based and size-independent structure similarity score (**Figure 5**). Our benchmark validation shows the robust performance of G-LoSA to the local structures of diverse sizes and characteristics, demonstrating its universal applicability to local structure-centric comparative biology studies. In particular, G-LoSA is highly effective in detecting conserved local regions on the entire surface of a given protein. In addition, the applications of G-LoSA to identifying template ligands and predicting ligand and protein binding sites (BS) illustrate its strong potential for computer-aided drug design. G-LoSA is a useful computational method for exploring interesting biological problems through large-scale comparison of protein local structures and facilitating drug discovery research and development. The program G-LoSA is freely available to academic users at http://compbio.lehigh.edu/GLoSA.

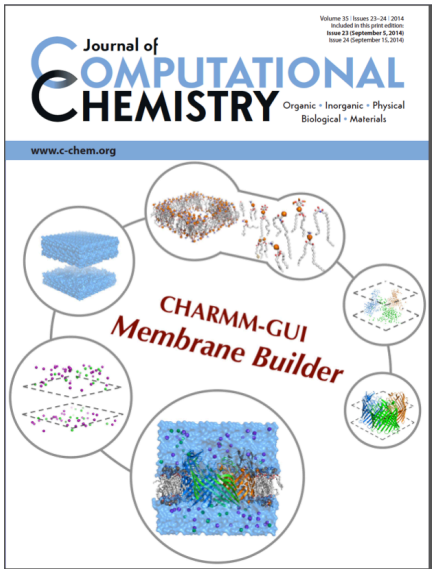

Figure 6. *Membrane Builder* is most popular in CHARMM-GUI.

6. CHARMM-GUI / ST-analyzer Development

CHARMM-GUI, http://www.charmm-gui.org, is a web-based graphical user interface to prepare molecular simulation systems as well as CHARMM and NAMD input files to facilitate the usage of common and advanced simulation techniques. Since its original development in 2006, CHARMM-GUI has been widely adopted for various purposes and now contains a number of different modules designed to setup a broad range of simulations (**Figure 6**). It has been used for the entire simulation community world-wide (**Figure 7**) and check http://scholar.google.com/citations?user=WEqu3RYAAAAJ for CHARMM-GUI citations). Currently, we are trying to add more modules as well as to provide simulation inputs for GROMACS, CHARMM/OpenMM, OpenMM, and other simulation programs.

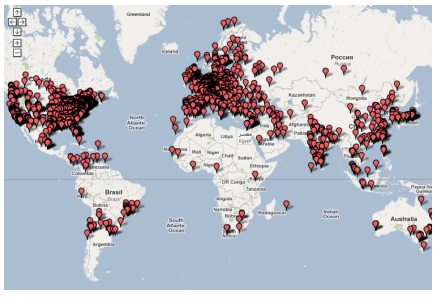

Figure 7. Geographical visitors to CHARMM-GUI.

Although many laboratories routinely perform MD simulations, analyzing MD trajectories is still time-consuming and often a difficult task. ST-analyzer, http://compbio.lehigh.edu/st-analyzer, is a standalone graphical user interface (GUI) toolset to perform various trajectory analyses with several outstanding features compared to other existing analysis tools.