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Research Interests: Investigation of protein and peptide structures and dynamics using mass spectrometry; gas phase ion molecule chemistry. Interdisciplinary spectroscopy laboratories for physical chemistry.

My group uses mass spectrometry combined with hydrogen/deuterium (H/D) exchange and computational chemistry to characterize the molecular interactions and dynamics in macromolecules (specifically amino acids, peptides and proteins). Mass spectrometry is an emerging as an effective probe macromolecule structure in both solution and the gas phase. The long-term goal of these projects is to take advantage of these techniques to develop a greater understanding of non-covalent interactions and molecular dynamics of large biological molecules in both solution and the gas phase. In addition, I have an NSF funded project to develop new spectroscopy lab and class materials for the physical chemistry curriculum. *This summer I will have students continuing work on 1) extending our knowledge of the mechanism of H/D exchange of peptides in the gas phase using mass spectrometry (1-2 students anticipated) 2) using LC-MS coupled with to probe solution structures of proteins (1-2 students anticipated)). 3) Developing interdisciplinary project based spectroscopy laboratories for physical chemistry (1 student anticipated)*

For the first two projects, using mass spectrometry, we have two different instruments. An electrospray ionization ion trap mass spectrometry[1] will be used to study the gas phase hydrogen/deuterium exchange of small peptides. The LC-Q-TOF instrument will be utilized to probe solution structure, ligand binding and protein dynamics by hydrogen/deuterium exchange mass spectrometry. Interested students are encourage to read the reviews in reference 1c and d for information on the electrospray process used on both instruments and to consult the posters outside my office, prior to coming in for discussion.

Gas phase hydrogen exchange This project focuses on the fundamental structures of biological molecules in the absence of solvent.[2,] Characterization of the gas phase structure of proteins is important for understanding the degree to which interactions with water play a role in determining three dimensional protein structures. This builds on work from the early 1990's when Fred McLafferty showed that proteins appear to adopt compact structures in the gas phase.[3] Accompanying commentary by Peter Wolynes discusses how in the absence of solvent "the stability of the native protein is not compromised… the classical secondary structures should be even more thermodynamically stable."[4] This summer we will focus on hydrogen deuterium exchange of peptides, as understanding the exchange of simple systems is necessary for complete analysis of proteins. The mass spectrometry part of these studies is carried out using a Finnigan Electrospray Ionization Ion Trap Mass spectrometer. (ESI-ITMS). Electrospray ionization allows the volatilization of large molecules for study by mass spectrometry. H/D exchange of labile protons in a molecule (typically the hydrogen attached to nitrogen and oxygen) results in a mass shift by 1 amu for each hydrogen exchanged and is easily followed by

mass spectrometry. Reference 2 includes recent work published by our group using this technique.

Hydrogen/Deuterium exchange mass spectrometry studies have been extensively used to probe gas phase structures of proteins and peptides.[5,6] To assist analysis of protein H/D exchange, a thorough understanding of the mechanisms and intrinsic rates of exchange are necessary. In past summers we have investigated the rates of exchange of small di and tri peptides with exchangeable side chains, primarily focusing on peptides with aromatic and acidic side chains, and recently introducing alkali metals in place of the proton. We use solid state synthetic techniques to create custom peptides, and electronic structure calculations to determine the low energy conformations of the molecules and to map out the potential energy surface for exchange. These combinations of techniques allow us to start to investigate the subtle effects of amino acid position on exchange. This summer we will continue these investigations continue to branch out in complexity of peptides and side chains. These studies will combine synthesis, mass spectrometry, kinetic fitting and computational chemistry. In addition we may expand these studies of structure using the new Xevo Q-Tof mass spectrometer to investigate the collision induced dissociation of these peptides. No previous experience is necessary; students will learn techniques involved in studying structures of large and small molecules using mass spectrometry.

Probing solution structure, ligand binding and protein dynamics by and hydrogen/deuterium exchange mass spectrometry (Reference 8 gives 2 review articles on this technique) Solution phase H/D exchange coupled with mass spectrometry can also be used to probe protein structure and interactions.[7] At least one project this summer will couple solution H/D exchange with protein digestion and using the LC-Q-TOF system to study the extent of protein dynamics and ligand binding in proteins.[8] The ligand binding event typically initiates some sort of structure change, which changes the extent of HDX and by localizing the area in the protein where the HDX has changed one can determine the areas involved in binding. The HDX reaction is also very sensitive to the shape and movement of the protein. The more dynamic a protein is in solution, the greater the rate of exchange. These dynamic events are important to characterize as they can influence the function of the proteins. This technique can be augmented by performing rapid solution proteolysis prior to mass spectrometry to determine specific regions of the protein protected from exchange.[9] Current work in the lab involves applying this technique to the protein MTHFR and its mutants (see professor Elizabeth Trimmer's research description). Previous experience in biochemistry is helpful for this project.

Developing interdisciplinary project based spectroscopy laboratories for physical chemistry Through a recent grant from the NSF-TUES program we have acquired a suite of instrumentation to carry out spectroscopic and kinetic studies. A variety of experiments are under development including 1) Size determination of quantum dots. Quantum dots are nanoscale materials with luminescent properties reflecting their size. They provide an opportunity for students to study nanomaterials with potential practical applications in both biological and material science. 2) experiments on exotic kinetic systems as relevant to atmospheric chemistry, and 3) Raman spectroscopy for forensic analysis. This project wil involve working with nanoparticle systems, as well as giving students experience with

fluorescence spectroscopy and computational modeling. No experience with physical chemistry is necessary for this project.

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