Investigation of Dynamic Processes in RNA-Protein Recognition

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Biomolecular recognition involves dynamic processes, conformational changes and kinetics at different, usually fast, time scales. It has been challenging to understand and predict how the structure and sequence of biomolecules contribute to these essential dynamic recognition processes. We are particularly interested in studying RRM-RNA interactions. RNA recognition motifs or RRMs are found in hundreds of RNA-binding proteins that bind to different RNA substrates with varying affinities and specificities.

The U1A protein, which is a component of the U1 snRNP and is involved in pre-mRNA splicing, is a model system for studying RNA-RRM interactions. Conformational changes occur in both RNA and protein upon binding. We are trying to understand the role of individual amino acids and base functional groups in dynamic processes that contribute to binding affinity and kinetics. We are also developing methods that will probe long-range cooperative interactions in this system.

We are using three approaches to study dynamics in the U1A-RNA recognition. First, time resolved fluorescence anisotropy to determine motions on the ns-timescale. Second, fast kinetic experiments to determine folding or unfolding as it affects association and dissociation rates. Lastly, we use computational simulations as a theoretical basis to understand the experimental results. These will provide us with a means to identify key residues and functional groups that are involved in these dynamic processes. We have used mutational studies to explore the role of several amino acids in the dynamics of an essential helix in U1A and have investigated their effects on complex formation and stability.