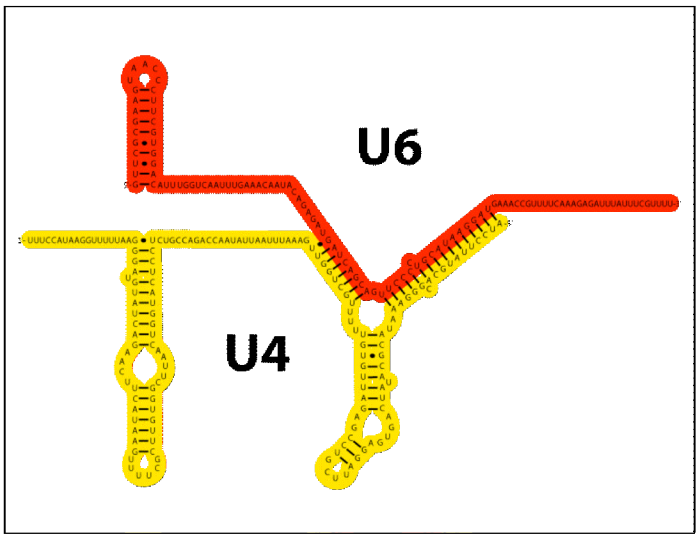
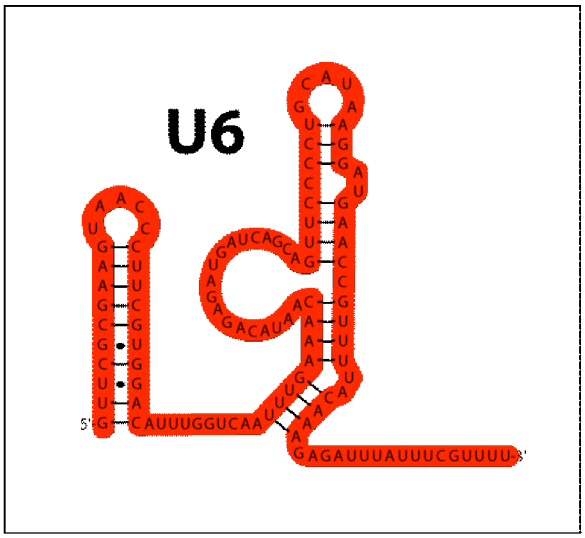
Rader Lab Research

The detailed, atomic mechanism by which enzymes function has been studied in many protein enzymes and even a few RNA enzymes. A new challenge in understanding enzyme mechanism lies in the elaborate, multi-molecular enzymes such as those that catalyze translation, transcription, and pre-messenger RNA (pre-mRNA) splicing. The complex that carries out pre-mRNA splicing (the removal of non-coding sequences from pre-mRNA to generate messenger RNA) is known as the spliceosome, and consists of 5 small nuclear RNAs (snRNAs) and roughly 100 proteins. The mechanism by which the spliceosome removes the non-coding sequences (introns) from the pre-mRNA is characterized by dynamic changes in interactions between the RNA and protein molecules. Research in my lab aims to understand these dynamic processes at a molecular level, using techniques such as fluorescence resonance energy transfer (FRET) and x-ray crystallography.



The spliceosomal components (the snRNAs and their associated proteins, as well as transiently associated proteins) associate with their pre-mRNA substrate with the U4 and U6 snRNAs extensively base-paired to each other (see figure, U6 red, U4 yellow). The base pairing between U4 and U6 is required for entry of U6 into the splicing complex. Formation of base pairs between U4 and U6 is catalyzed in *S. cerevisiae* by the 51 kDa protein Prp24, a member of the RNA-binding domain (aka RBD or RNA recognition motif, RRM) family of proteins. During formation of the base-paired U4/U6 complex, a large stem loop at the 3' end of U6 snRNA (see figure of free U6) must be completely unwound to allow formation of the mutually exclusive base pairs with U4. My lab seeks to address the mechanism by which Prp24 brings U4 and U6 together in the base-paired complex.



Once U6 has been delivered to the splicing apparatus, the base pairs with U4 are disrupted and U4 becomes weakly associated with the rest of the splicing complex (see figure). Free U4 (*ie* U4 not associated with U6) is also believed to contain secondary structure elements that must be unwound prior to base pair formation with U6 (see figure of free U4). While it is known that a number of proteins are associated with U4, their function in base pair formation or subsequent steps of spliceosome assembly remains largely unexplored. The function of the U4-associated proteins is another area of interest in my lab.