BMB 632
Probing structure and function of complex RNA-protein machines

Instructors: Kristen W Lynch, Kathy Liu, Jeremy Wilusz

Mondays, 2:10-4 pm
CRB 302

Summary:
RNA-Protein complexes or RNPs can range from simple assemblies to megadalton enzymatic machines. The latter include two of the most abundant and essential enzymatic complexes for converting genes to functional protein – the ribosome and the spliceosome. Understanding the molecular interactions that hold these RNPs together and how these complexes function has required the development of new techniques and pushed the boundaries of quantitative biochemistry. In this course we will take an in-depth look at general concepts common to many RNA binding proteins, the methods used to study protein-RNA and RNA-RNA interactions, and how the complex nature of large RNPs uniquely allow them to achieve their precise functions. The course will be a combination of both lectures and student-lead discussion of recent literature. Students will be evaluated based on their presentations of primary literature and their participation in class discussion and a final oral exam.

Syllabus:

Oct 7 Overview of RNA-Protein Machines and Discussion of Basic Methodology: primer extension, RT-PCR, RNA-Seq, CLIP-Seq

Oct 14 Ribozymes and Riboswitches: foot-printing and SHAPE

Oct 21 Ribosomes and Spliceosomes: RNA folding and Identifying RNA binding proteins (UV crosslinking, RNA affinity)

Oct 28 Structure and determinants of RNA-RBP interface: NMR and FRET (EMSA for affinity/specificity?

Nov 4 Nucleotide modifications by and in RNP complexes: snoRNAs and mRNA mods

Nov 11 Spliceosome and the study of large RNPs (Native gels, RNAse H, psoralen, EM)

Nov 18 Regulation of Splicing: Site-specific labeling, psoralen, MS2 purification

Nov 25 Single-Molecule study of Spliceosome and RISC: FRET, CoSMoS

Dec 2 CRISPR: use of RNP as tools

Dec 9 Highthroughput methods to study RNA and RNPs: RNA-Seq, Ribosome footprinting, Mass Spec, etc

EXAM DATE TBD (Likely Dec 16 or 17)