system show a strong propensity for extended inter-strand base pairing and the formation of stable intermediates. In contrast, a sequentially similar, but non-functional system shows almost no potential for extended inter-strand base pairing. This observation was consistent even when the system was subjected to increased temperatures and external pulling forces that greatly increased the rate of complex formation in the known attenuator system. These results suggest that changes in the energetic landscape of inter-strand base pairing can be captured by coarse-grained MD simulations and provide a route for the prediction of new, orthogonal sequences capable of transcriptional attenuation in synthetic systems.

1199-Pos Board B150
U4-snoRNA Regulates Formation of the U6 Telosm within the U4/U6 Di-snoRNA
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Many of the splicedosomal small nuclear RNAs (snRNAs) undergo large-scale structural rearrangements during splicedosomal assembly, activation and catalysis. This is exemplified by the highly conserved U6 snRNA which can exist in isolation in the U6 small nuclear ribonucleoprotein (snRNP); basepaired with U4 in the U4/U6 di-snRNP, U4/U6/U5 tri-snRNP, and splicedosomal B complex; or basepaired with the pre-mRNA intron and the U2 snRNA in the splicedosomal spliceosome. Through genetic evidence and structure probing (1, 2), it was previously proposed that the U4/U6 di-snRNA consisted of a three-helical junction core domain of U4/U6 and U6-strand regions. Using single-molecule Förster resonance energy transfer (smFRET), we discovered a dynamic equilibrium between RNA conformations formed by the flanking single stranded regions. Our smFRET data is consistent with the presence of two mutually exclusive structures: (1) a U4/U6 di-snRNA containing both basepaired U4/U6 and the U6 telosm and (2) a U4/U6 di-snRNA with extended basepairing located near U4/U6 stem I. These data show that the U6 telosm persists in the U4/U6 di-snRNA and that its formation can be regulated by novel basepaipring interactions with U4. This suggests conservation of mechanism between the yeast and human U4/U6 di-snRNAs. Finally, we speculate that telosm formation can be dynamically regulated by splicedosomal proteins to aid in tri-snRNP disassembly and splicedosomal activation_rationalizing previous observations concerning the mechanism of the U4/U6 RNA helicase Br2.

References

1200-Pos Board B151
A Ribosomal Frameshifting Structure in the CCR5 mRNA Leads to Mirna-Stimulated Simultaneous-Mediated mRNA Decay
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RNA structure-based signals are used by viruses to affect programmed ribosomal framashefiting (PRF) yielding extended fusion protein products in a fraction of translation processes. Computationally identified – 1 PRF signals in cellular mRNAs have been predicted to lead to premature termination of translation in the vast majority of cases. We found that a – 1 PRF signal predicted in the mRNA of the human CCR5 cytokine receptor is a pseudoknotted structure that is responsible for redirecting translation toward a premature termination codon (PTC), ultimately destabilizing the mRNA via the nonsense mediated mRNA decay pathway (NMD) and possibly another decay pathway. A chemo-kine receptor, CCR5 is also a co-receptor used by HIV-1 to enter its target CD4+ T-cells. We built a 3D model of the – 1 PRF structure and validated its stability in molecular dynamics simulations (MD). The structure is a two-steemed pseudoknot, with the larger of the two stem domains consisting of multiple half-turrel he-lical segments, separated by asymmetric single strands, which enable bending of the larger stem region and bridging of the two stems. MD predicted this larger region and results 3’ at oil the structure to be very stable, which is consistent with the experimental results indicating that it may form triple base interactions with miR-1224. Such interactions can increase the stability of the whole – 1 PRF structure, ultimately resulting in an increased fraction of paused ribosomes. Another miRNA, miR-141, is also predicted to interact with the same region. Experiments have also demonstrated that several other cytokine receptor

mRNA – 1 PRF signals are controlled via miRNAs. Altogether these results indi-cate a novel mechanism of cellular gene expression regulation via a – 1 PRF structural signal further mediated by the sequence-specific miRNA interactions. Funded in part by HHSN261200800001E.

1201-Pos Board B152
Exploring RNA Condensation
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DNA and RNA molecules both serve as genetic material inside viruses but the structure of packaged RNA molecules remain elusive. Moreover, RNA has more diverse functional roles than DNA. Differences between DNA and RNA duplexes are most evident upon addition of a condensing agent, such as the trivalent ion cobalt hexammine (CoHex). Under conditions where DNA condenses into insoluble precipitates, RNA molecules remain in solution (Physical Review Letters 2011, 106, 108101). In fact, x-ray scattering experiments suggest that in the presence of CoHex, short RNA helices form end-to-end stacked soluble structures. We study the physical origin of nucleic acid condensation by comparing four 25 base-paired helical constructs: DNA, RNA and a DNA-RNA hybrid of similar sequences and a homopolymeric DNA sequence. Recently, we used UV spectroscopy and Circular Dichroism (CD) to connect condensation propensity to spatial location of CoHex ions determined from molecular dynamics simulations (Nucleic Acids Research 2014, 42, 10823). Here, we use solution x-ray scattering to examine the pre-condensed phase and explore conditions that might effectively condense and package RNA.

1202-Pos Board B153
Alternative Base-Pairing and Conformational Sampling in Loop a of the Hairpin Ribozyme
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The catalytic cycle of the hairpin ribozyme begins with the ‘‘docking’’ between loops A and B and the consideration of this step as the rate limiting step for the nucleolytic catalysis presents an intriguing question. The kinetic rates of docking previously reported by ourselves and others indicate that conformational sampling may be a factor that limits the docking transition rates. To assess conformational sampling in the pre-docking form of the hairpin ribozyme, we have run several explicit solvent Molecular Dynamics (MD) simulations of hairpin ribozyme loop A domain totaling 2.4 μs. We observed three dominant conformers and other minor states identified using hydrogen bonding and base stacking in the loop region. Targeted Molecular Dynamics (TMD) was used to model the pathway between a pair of conformations using CHARMm36/NAMD. The Molecular Mechanics Poisson-Boltzmann Surface Area (MMPB/SA) approximation was applied to predict conformational free energy and energy differences between various conformations. MMPB/SA correctly predicted the major conformations to be lower in free energy than the minor conformations. The observation of multiple conformations with different energies seem to underscore the formation of a rugged energy landscape with multiple accessible shallow energy minima for loop A of the hairpin ribozyme, consistent with a possible role for conformational sampling in the observed slow rates of docking.

Membrane Physical Chemistry I

1203-Pos Board B154
Influence of Ether Bonds and Branched Lipid Tails on Stability of Membranes to Pore Formation
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Archaeal lipid membranes have a number of unique structural features that distinguish them from those of bacteria and eukaryotes allowing archaeca to survive in harsh environments, such as high temperature, increased acidity and pressure. To date the data on the impact of certain peculiarities of archaeal lipid membranes to Pore Formation

Funded in part by HHSN261200800001E.