Molecular Dynamics Studies of Nucleic Acid-Protein Complexes

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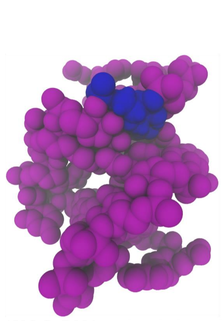
Abstract

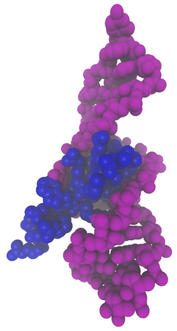
RNA-Protein complexes are an integral part of all living cells. Through the use of molecular simulation methods we aim to quantify the the strength of the interaction between these molecules. In order to quantify this interaction two different simulation methods will be used: Steered Molecular Dynamics (SMD) and Metadynamics. Through the use of SMD we can quantify the binding strength in pN by pulling the molecules apart at a constant velocity. Metadynamics allows us to look at the free energy difference between the bound state and the unbound state of these molecules.

Introduction

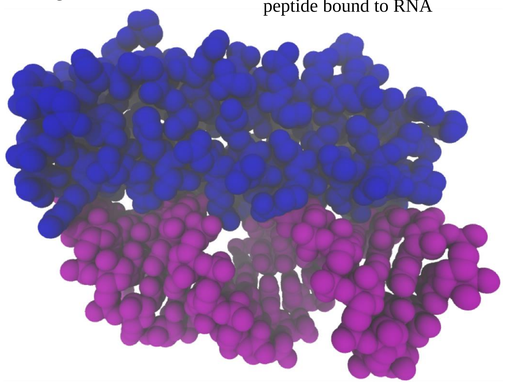
- The interactions between proteins and RNA molecules within living cells play a vital roll in a number of cellular processes.
- RNA-protein binding helps to create arguably one of the most important pieces of cellular machinery, the Ribosome
- The Ribosome is a compilation of a variety of proteins, as well as ribosomal RNA molecules, that carry out the translation of messenger RNA into polypeptide chains of amino acids
- These chains are then folded into the proteins that the cell requires for proper functioning
- Another important role of these RNA-protein complexes is to regulate post-transcriptional control of gene expression by binding ribonucleoproteins (RNPs) to RNA
- There is a wide range of RNA binding proteins (RBP)
- Quantifying the binding strength of these interfaces, as well as determining the free energy barrier of the binding process, are integral pieces of information needed to further the knowledge of RNA-Protein complexes.
- Below are 4 examples of different types of RNA-Protein complexes



1KOC. RNA molecule bound to an Arginine residue

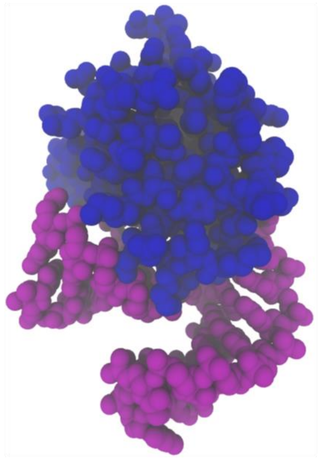


1ETG. Alpha helical arginine rich peptide bound to RNA



2AZO. Large protein bound to RNA at multiple residues

Steered Molecular Dynamics of Nucleic Acid-Protein Complexes



1AUD. RNP domain of human U1A protein in complex with RNA

SMD Technique

- SMD is a technique that allows one to apply an external force on an atom or group of atoms, while simultaneously keeping another atom or group of atoms fixed.
- This type of technique can be used to explore conformational changes in biomolecules or even the unbinding of ligands.
- Here the SMD technique is used to explore the binding strength between RNA-protein complex interfaces.

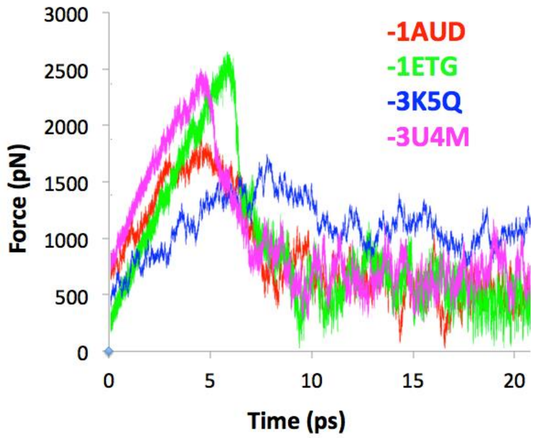
SMD Parameters

- SMD can be performed by either constant force or constant velocity pulling, here we use constant velocity.

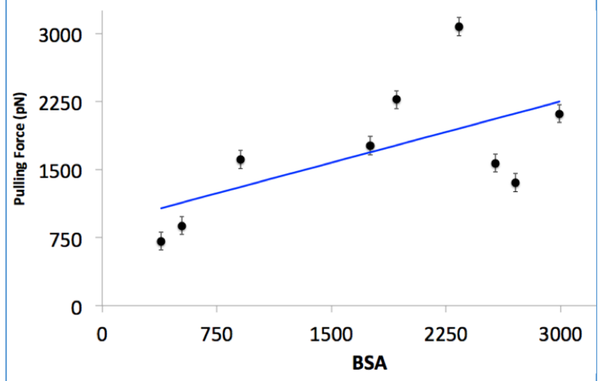
Parameter	Value
System sizes	12k to 92k atoms
Pulling velocity	0.000001 Å/ps
Spring constant k	7 kcal/mol/Å^2
Ensemble	NPT
Temperature	310K
Fixed atoms	Phosphorous atoms in nucleic acid backbone
SMD atoms	All protein atoms (excl. H) within 4 Å of any nucleic acid atoms

SMD Results

- Below are the force pulling curves for the SMD simulations of four of the nine complexes tested.
- The peak of the curve represents the force required to break the interaction between the protein and its nucleic acid.

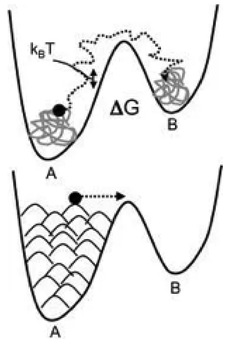


- Below is a scatter plot of the breaking force versus the buried surface area (BSA) of the complex.
- BSA is defined as the surface area of the complex that is not exposed to the solvent.
- It is hypothesized that a larger BSA will correspond to a larger breaking force.



- While there is some correlation between BSA and breaking force it's clearly not the only factor.
- One known factor that can affect breaking force is the presence of charged amino acid residues.
- Residues such as ARG or LYS that carry a positive charge can interact strongly with the negatively charged backbones in nucleic acids, increasing the binding strength.

Nucleic Acid Protein Complexes with Metadynamics

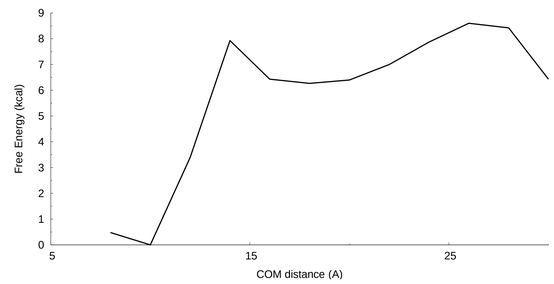


- Metadynamics is an enhanced sampling method used to explore the free energy landscape in a large collective coordinate space
- The height and width of the energy Gaussians are manipulated and added at a certain frequency to explore the depth of the free energy wells
- Because it's a history dependent potential the free energy landscape can be reconstructed iteratively by a sum of Gaussians
- This allows for the RNA-Protein complexes to explore rare metastable states that it may not be able to reach in a standard simulation

Metadynamics Results

Below is a free energy plot based on the center of mass distance collective variable (CV) for the 1KOC system.

- The CV ranges from 8 to 30
- The Gaussian width used is 2
- Well tempered metadynamics is used
- NPT ensemble



- As the Arginine molecule moves away from the RNA the free energy of the system increases.
- The lowest free energy of the system seems to be that of the initial configuration.
- With more systems currently being tested, the picture will become clearer as to how the free energy barrier relates to the binding affinity of the complex.

Acknowledgements

- All images were created using VMD software
- All simulations were performed using NAMD software
- We would also like to thank the University of New Hampshire for access to the supercomputer Trillian