

Human Islet Amyloid Polypeptide: Identifying Early-stage Aggregation Mechanisms through Molecular Simulation



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Abstract

Human islet amyloid polypeptide (hIAPP) is implicated in the onset of type II diabetes and is known to aggregate into amyloid fibrils. However, it is prefibrillar species, not mature fibrils, that are proposed to be cytotoxic. In order to better understand the role of hIAPP aggregation in the onset of disease, as well as to design effective therapeutics, it is crucial to understand the mechanism of early-stage hIAPP aggregation. In this work, we use atomistic molecular dynamics simulations combined with multiple advanced sampling techniques to examine the formation of the hIAPP dimer and trimer.

Introduction & Motivation

Millions of people worldwide are afflicted by diseases in which certain proteins misfold and aggregate into a fibrillar structure known as amyloid, accumulating in a specific part of the body. These diseases include type II diabetes and numerous neurodegenerative illnesses (ex. Alzheimer's, Parkinson's, and Huntington's diseases).

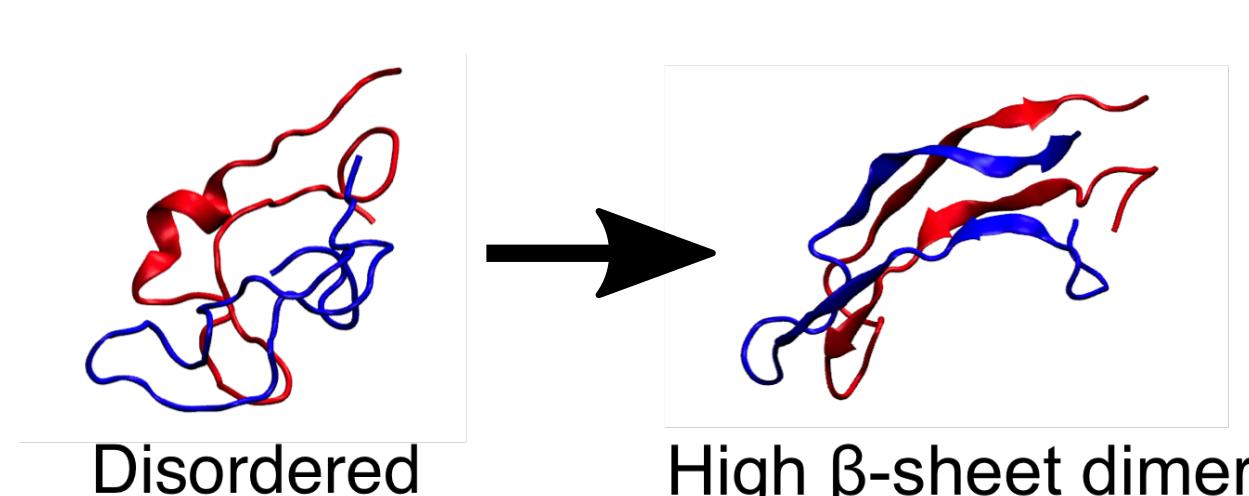
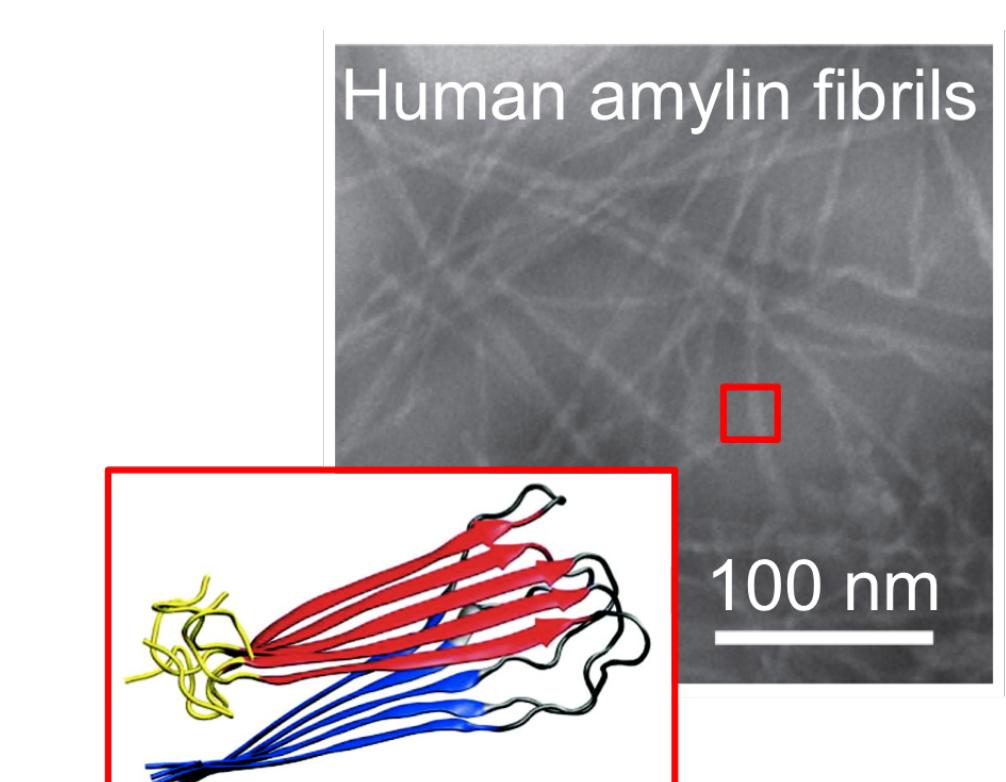
Amyloid can be toxic to cells; early-stage aggregates are most responsible for this cytotoxic effect. Atomistic molecular simulations have the potential to identify early-stage aggregation mechanisms and facilitate the study of relevant intermediate species.

Key questions:

- What are the mechanisms behind the first steps of amyloid formation?
- Can we find structures, pathways, or transition states to potentially target therapeutically?

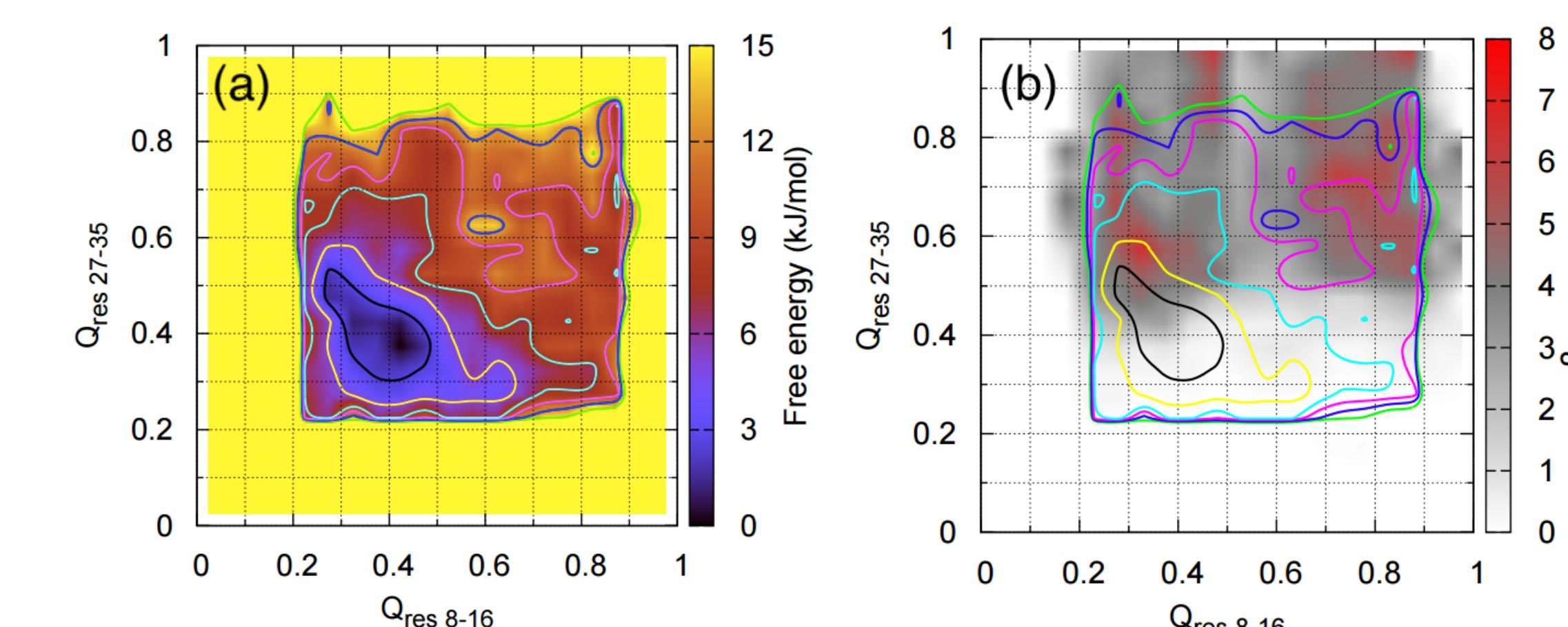
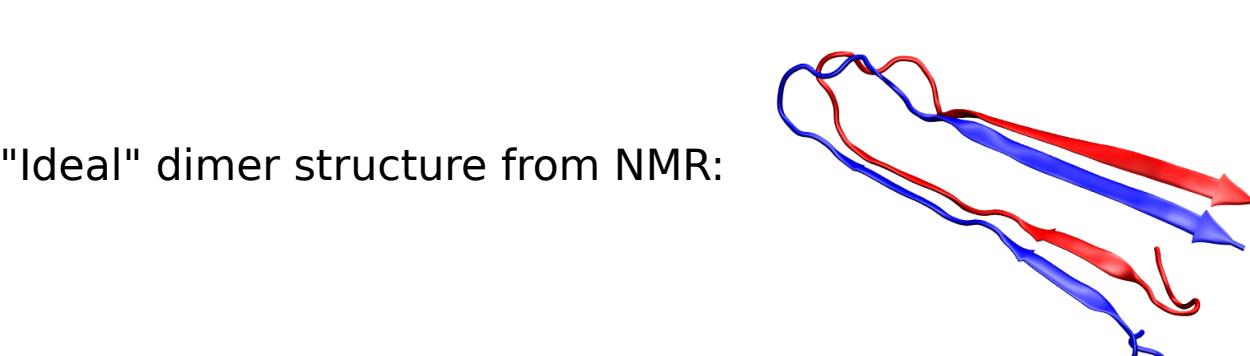
System: Human Amylin (hIAPP)

We aim to understand the early stages of amyloid formation, which are most relevant to disease pathology. We use a variety of molecular simulation techniques to study the mechanisms through which early aggregates form and their thermodynamic properties. Here we focus on human amylin (hIAPP), implicated in type II diabetes. This 37 residue polypeptide forms fibrils where individual hIAPP molecules stack in a parallel U-shape.



Bias-Exchange Metadynamics on hIAPP Dimer

Prior work applied metadynamics to the hIAPP dimer to map out a free energy landscape. However, metadynamics has disadvantages: simulation time is spent in regions irrelevant to the transition mechanism and specific aggregation pathways are not identified. Additionally, metadynamics would require an unreasonable amount of resources to study higher order aggregates.

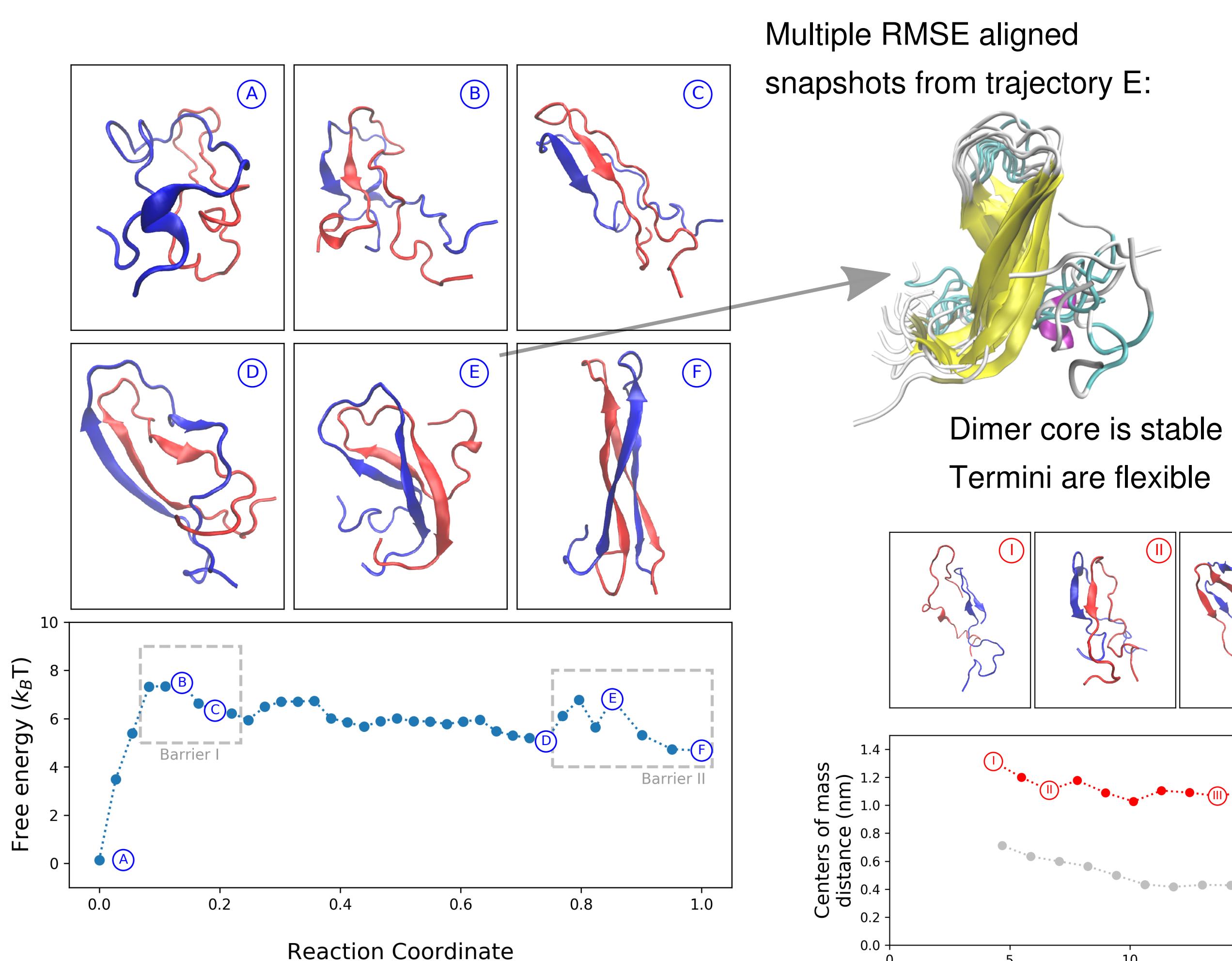


Qres x-y: RMSD similarity to "ideal" dimer in residues x-y

βRMSD x-y: Measure of parallel β-sheet character in residues x-y

Applying the String Method to hIAPP Aggregation

- We use the string method, a technique built for identifying and studying transition pathways, which has not been applied to amyloid formation or protein aggregation in general. Through this iterative method, a transition pathway is approximated between the disordered state and the highly β-sheet state.
- Finite Temperature String Method was performed using a combination of GROMACS 4.6.7, the PLUMED 2.1 plugin, and custom codes written in Python. This work was a basis for an open source string method implementation in SSAGES (Software Suite for Advanced Generalized Ensemble Simulations, MICCoM).

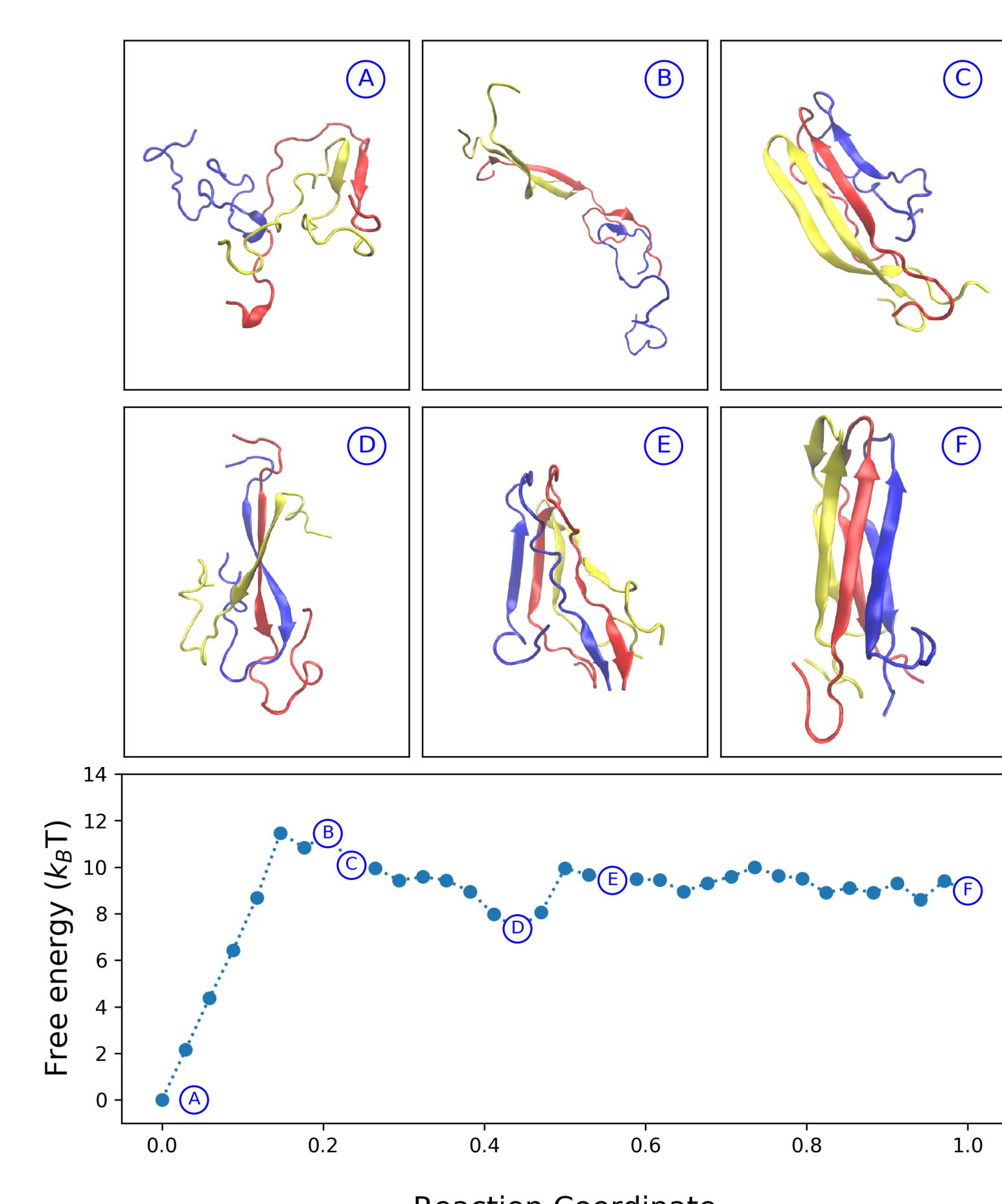
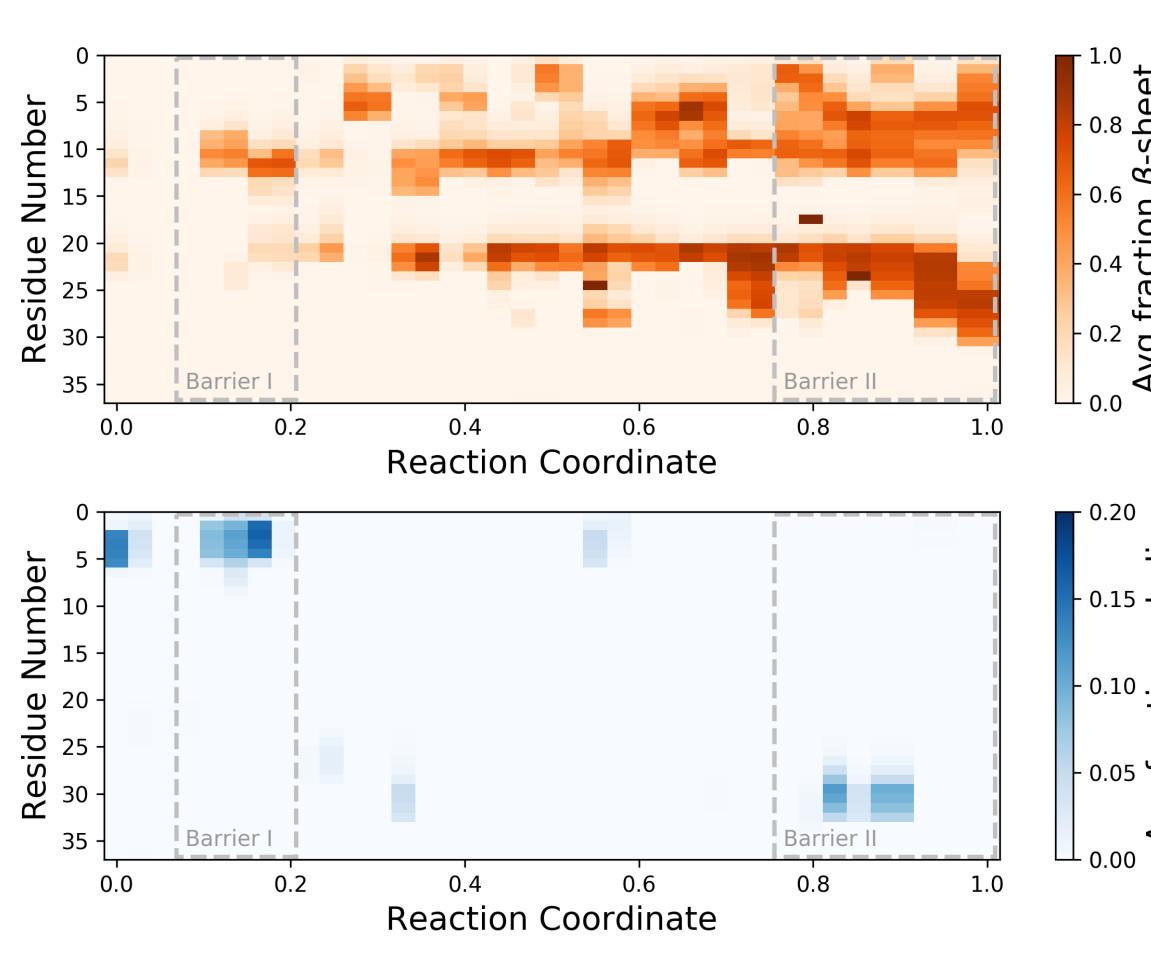


hIAPP Dimerization Pathway

Configurations obtained from string method match with previously predicted hallmarks of hIAPP aggregation:

1. β-sheets formed first in intermediate turn region
2. β-sheets formed in C-terminus before N-terminus

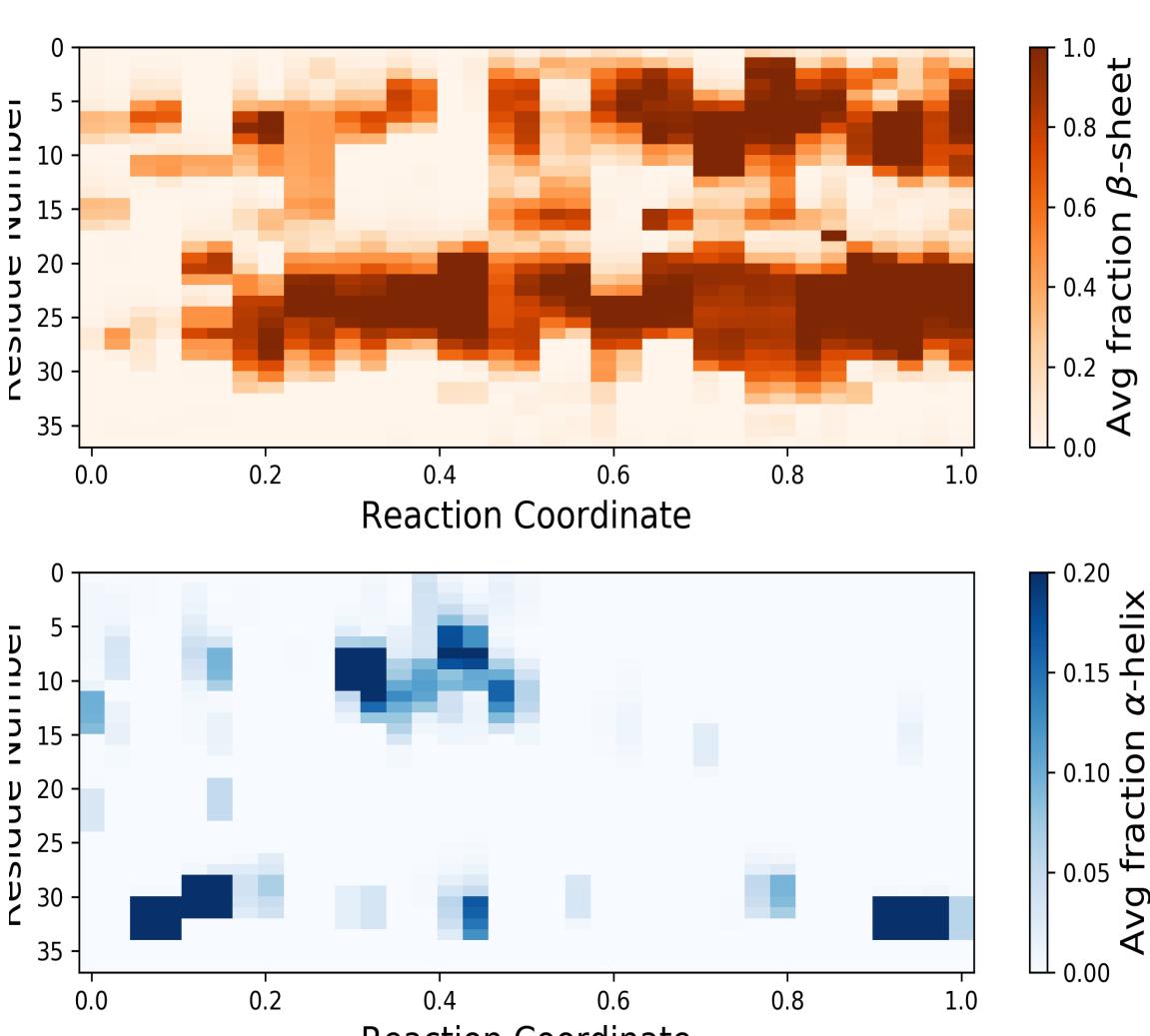
Final dimer found to be entropically stabilized.



hIAPP Trimerization Pathway

Configurations from trimer string method calculations show:

1. "Disordered" configuration contains the dimer intermediate, suggesting stepwise aggregation mechanism
2. Again intermediate β-sheets formed in the turn region



Next Steps

What happens when a disordered monomer approaches a full dimer?

What is the effect of physiological salt concentration?