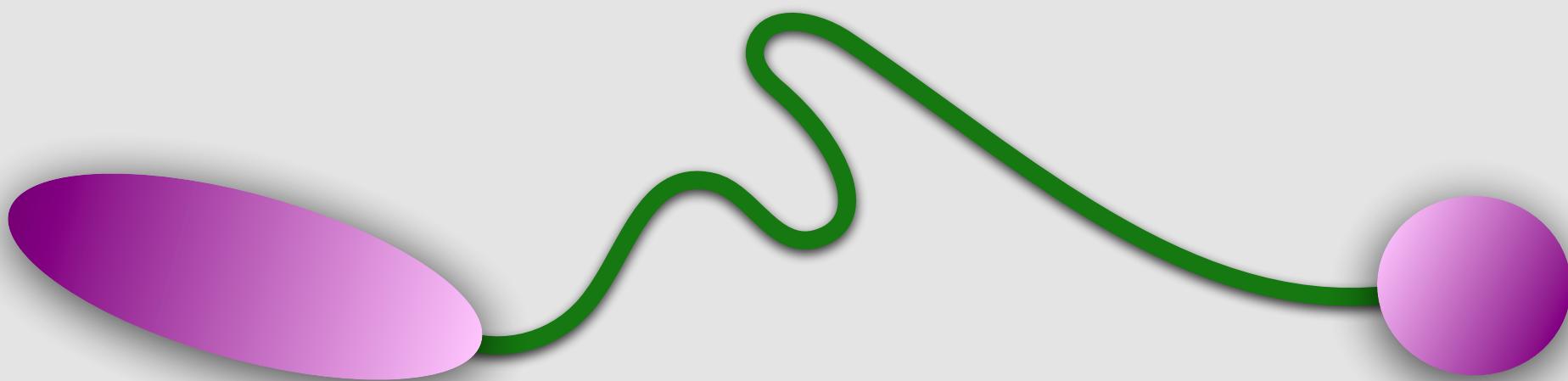
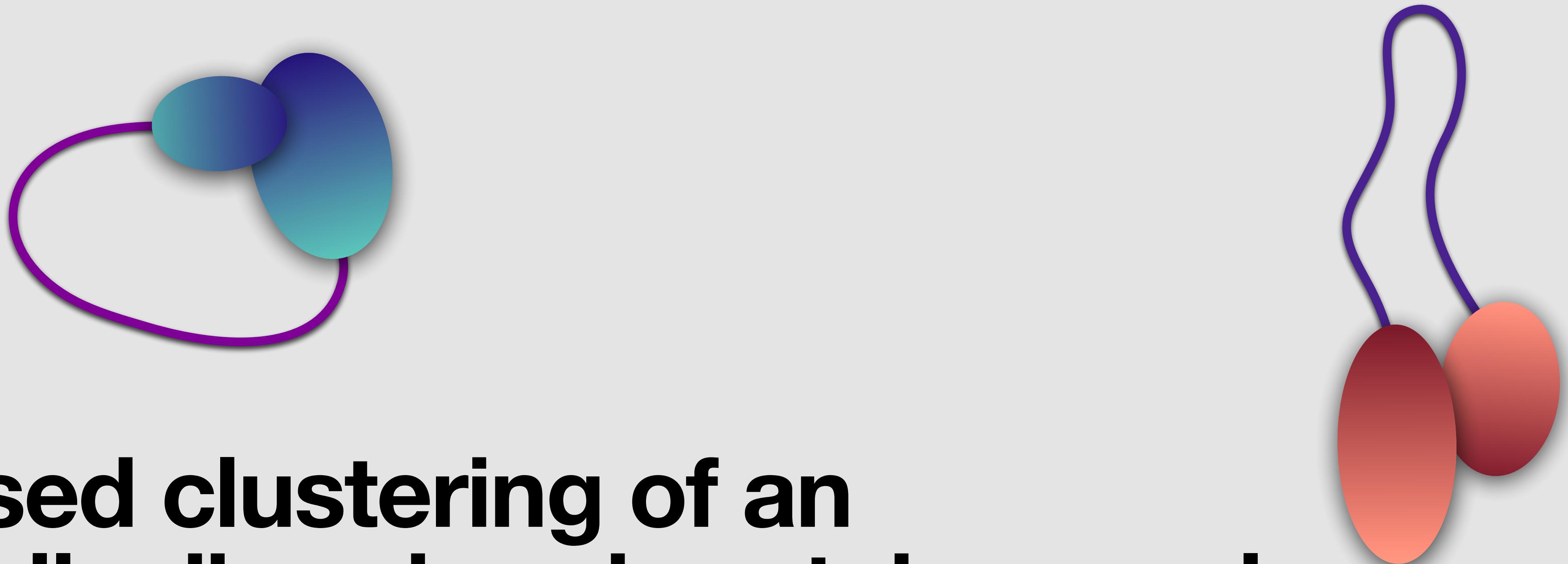
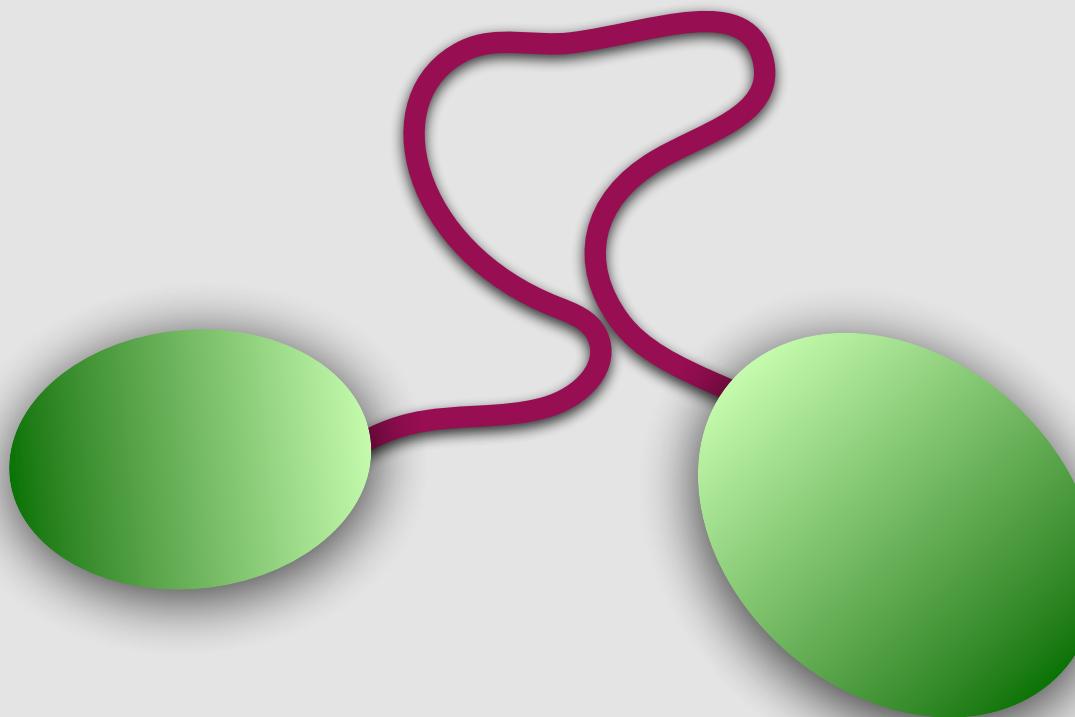


Blob-based clustering of an intrinsically disordered protein reveals distinguishable conformations

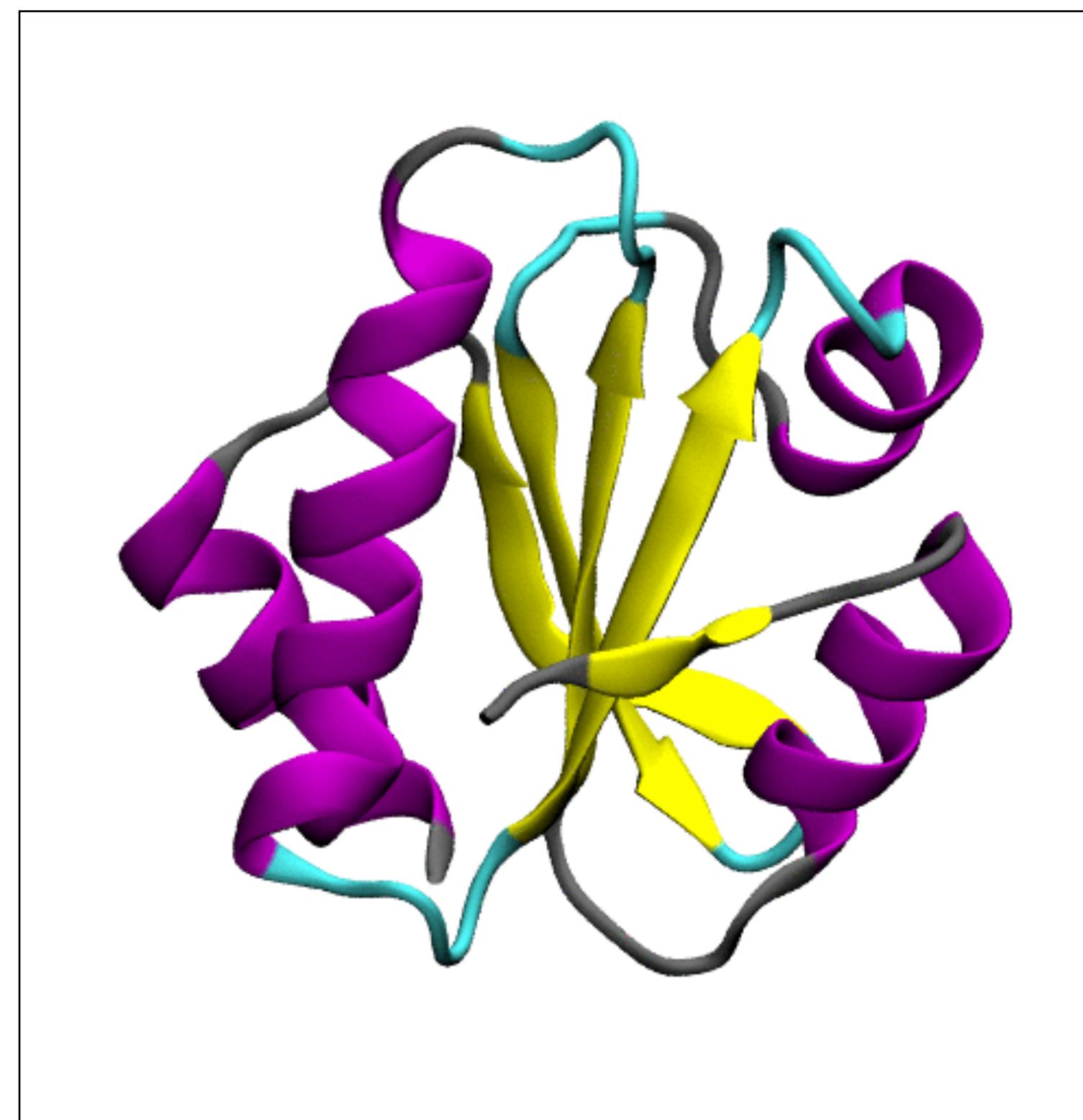


Lindsey Riggs, Seminar Fall 2025



Intrinsically disordered proteins adopt many conformations

Structured



Yeast Thioredoxin

Unstructured



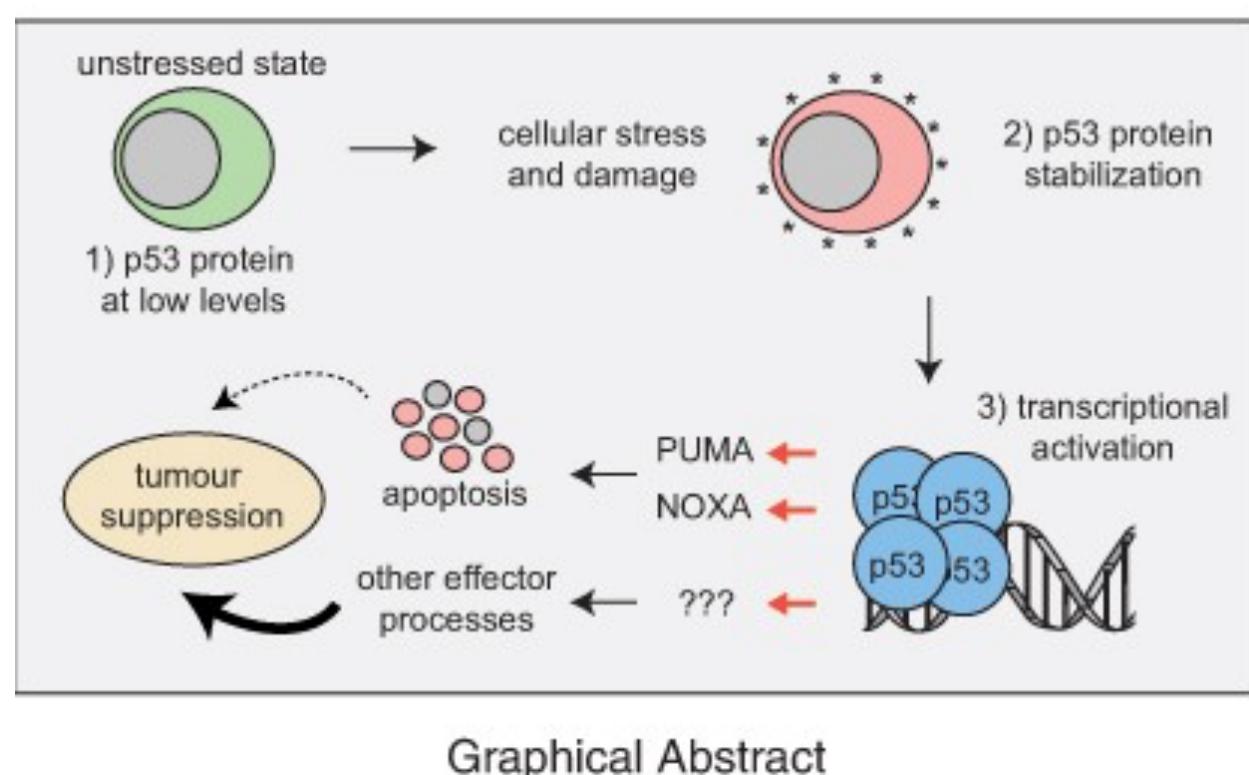
BDNF prodomain

Movie courtesy of Dr. Ruchi Lohia

Some physiological roles of IDPs in humans

Cellular death

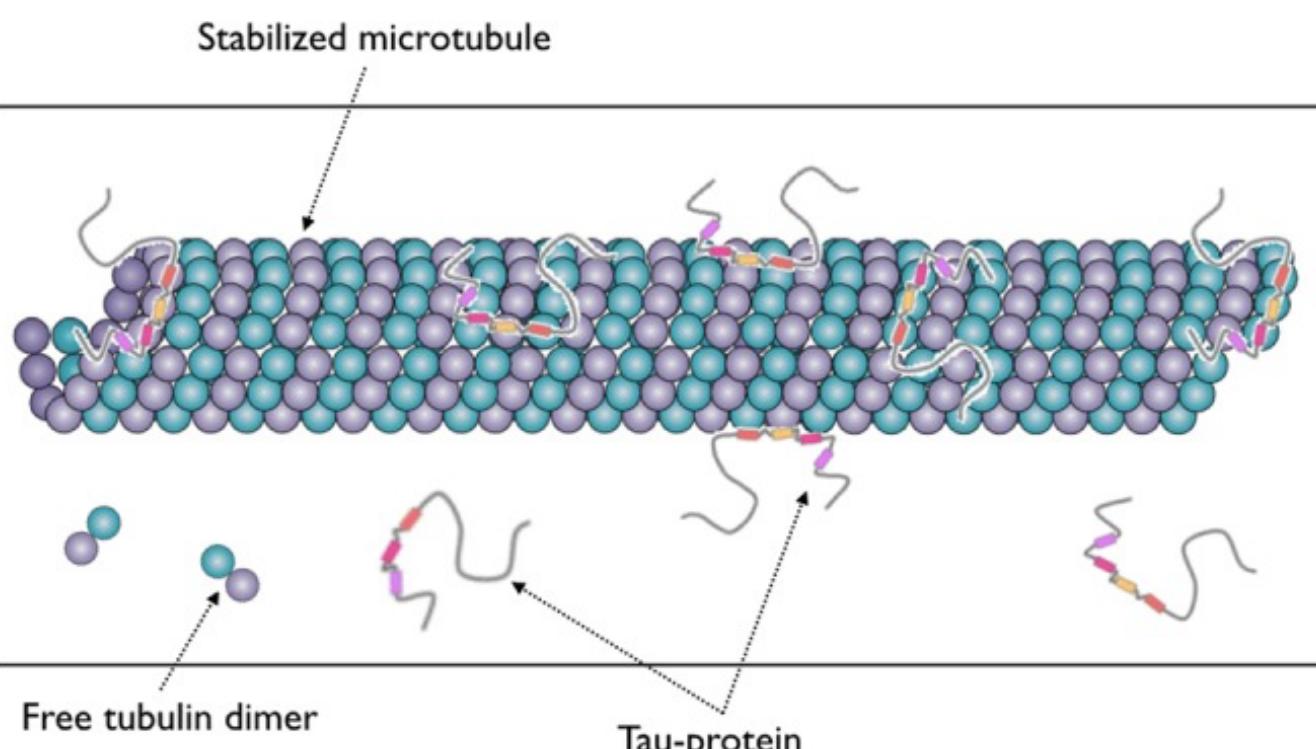
- p53 is activated by cellular damage, leading to apoptosis



Aubrey, Brandon J et al. *Cell death and differentiation*, 2018

Microtubule stabilization

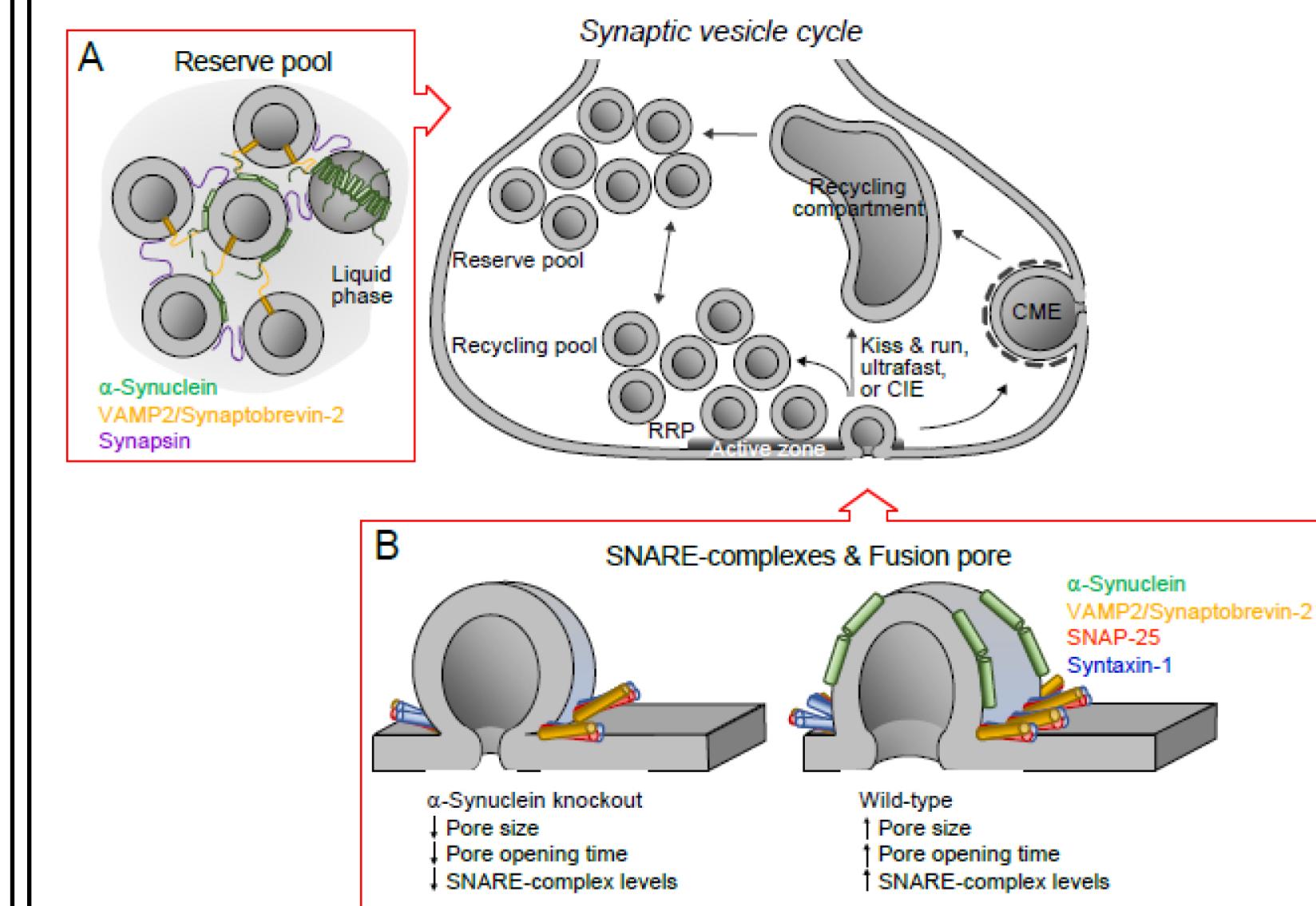
- Tau wraps around microtubules to stabilize them



Hervy, Jordan, and Dominique J Bicout. *Scientific reports*, 2019

Neurotransmitter Release

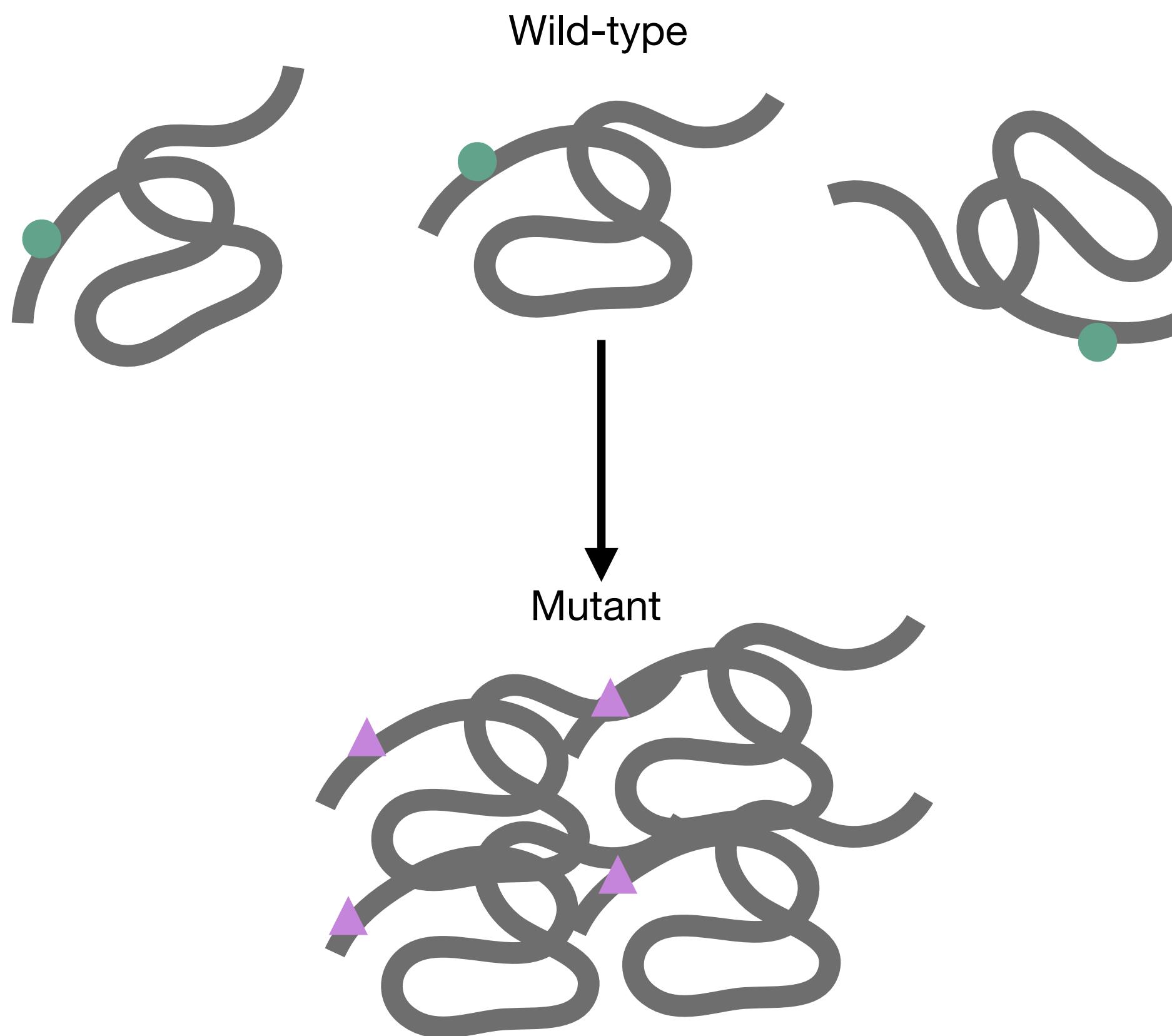
- α -synuclein inhibits neurotransmitter release by binding to synaptic membranes



Sharma, Manu, and Jacqueline Burré. *Trends in neurosciences*, 2023

Missense mutations can affect IDPs

Oligomerization

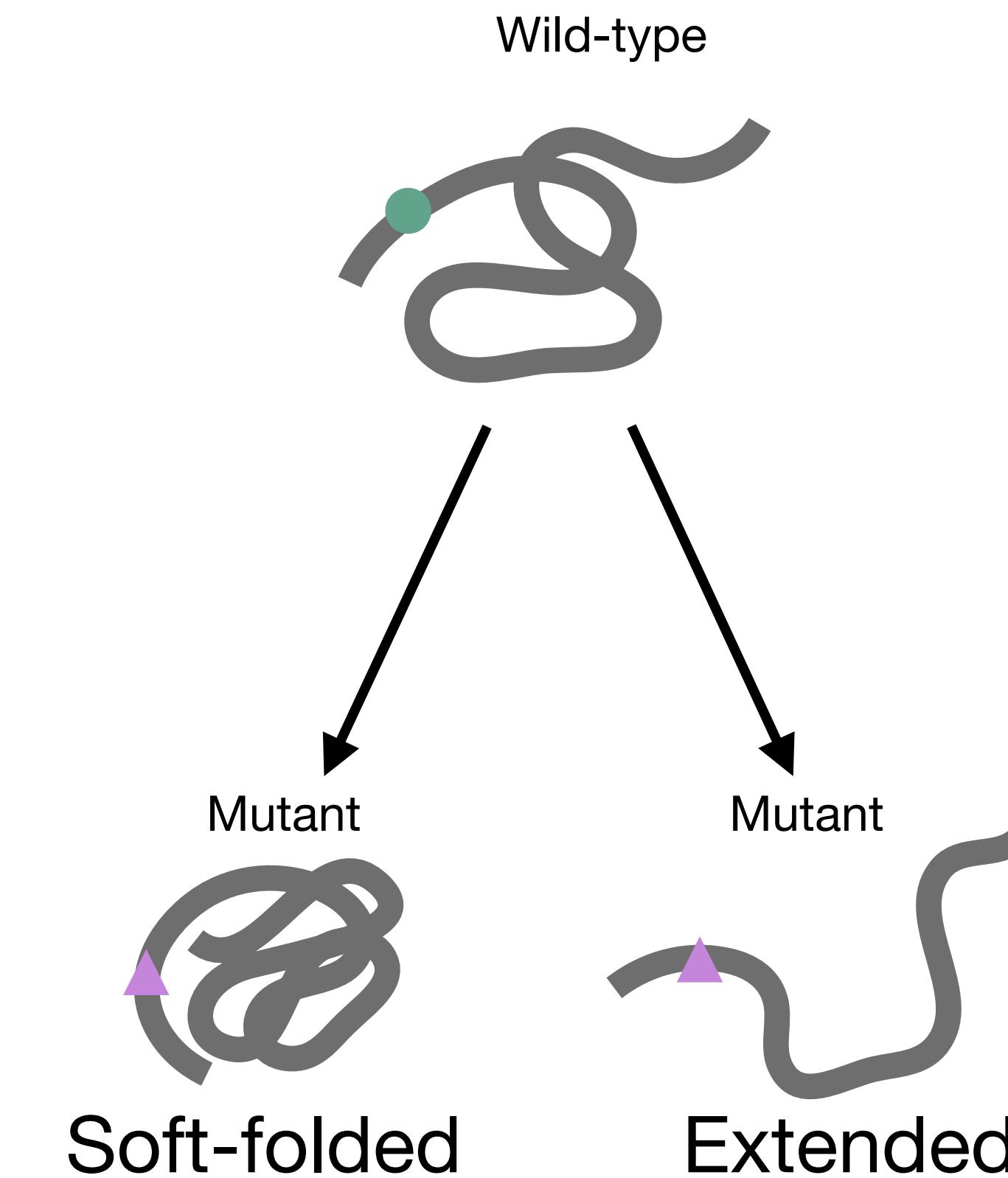


► Sci Rep. 2016 Mar 31;6:23928. doi: [10.1038/srep23928](https://doi.org/10.1038/srep23928)

Two mutations G335D and Q343R within the amyloidogenic core region of TDP-43 influence its aggregation and inclusion formation

Lei-Lei Jiang¹, Jian Zhao¹, Xiao-Fang Yin¹, Wen-Tian He¹, Hui Yang¹, Mei-Xia Che¹, Hong-Yu Hu^{1,a}

Compaction



► JACS Au. 2023 Dec 19;4(1):92–100. doi: [10.1021/jacsau.3c00550](https://doi.org/10.1021/jacsau.3c00550)

Mutations in Tau Protein Promote Aggregation by Favoring Extended Conformations

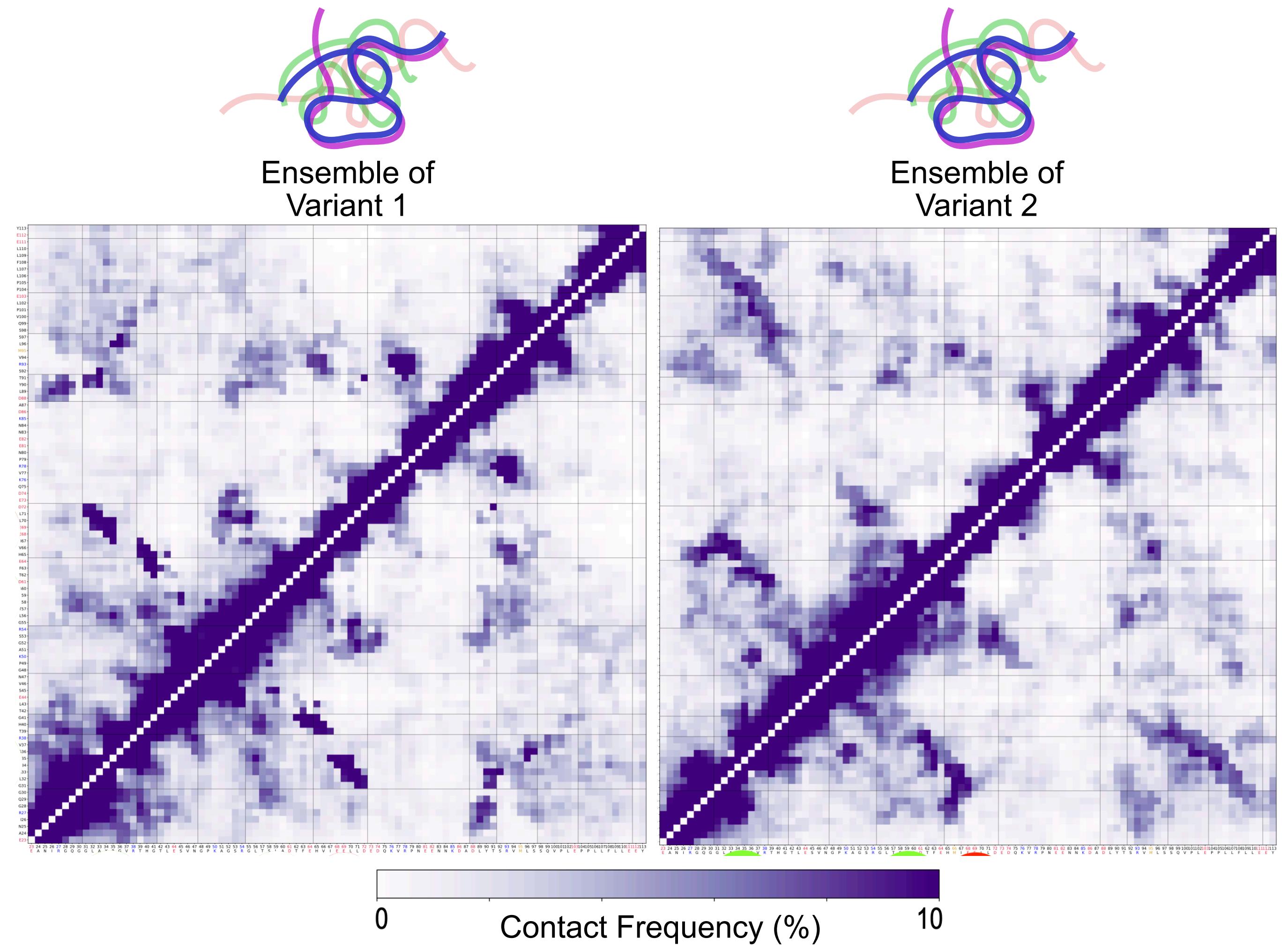
Kevin Pounot[†], Clara Pierson[‡], Andrew K Goring[§], Frédéric Rosu[¶], Valérie Gabelica^{¶,||}, Martin Weik[†], Songi Han^{#,¶}, Yann Fichou^{‡,*}

Hypothesis:

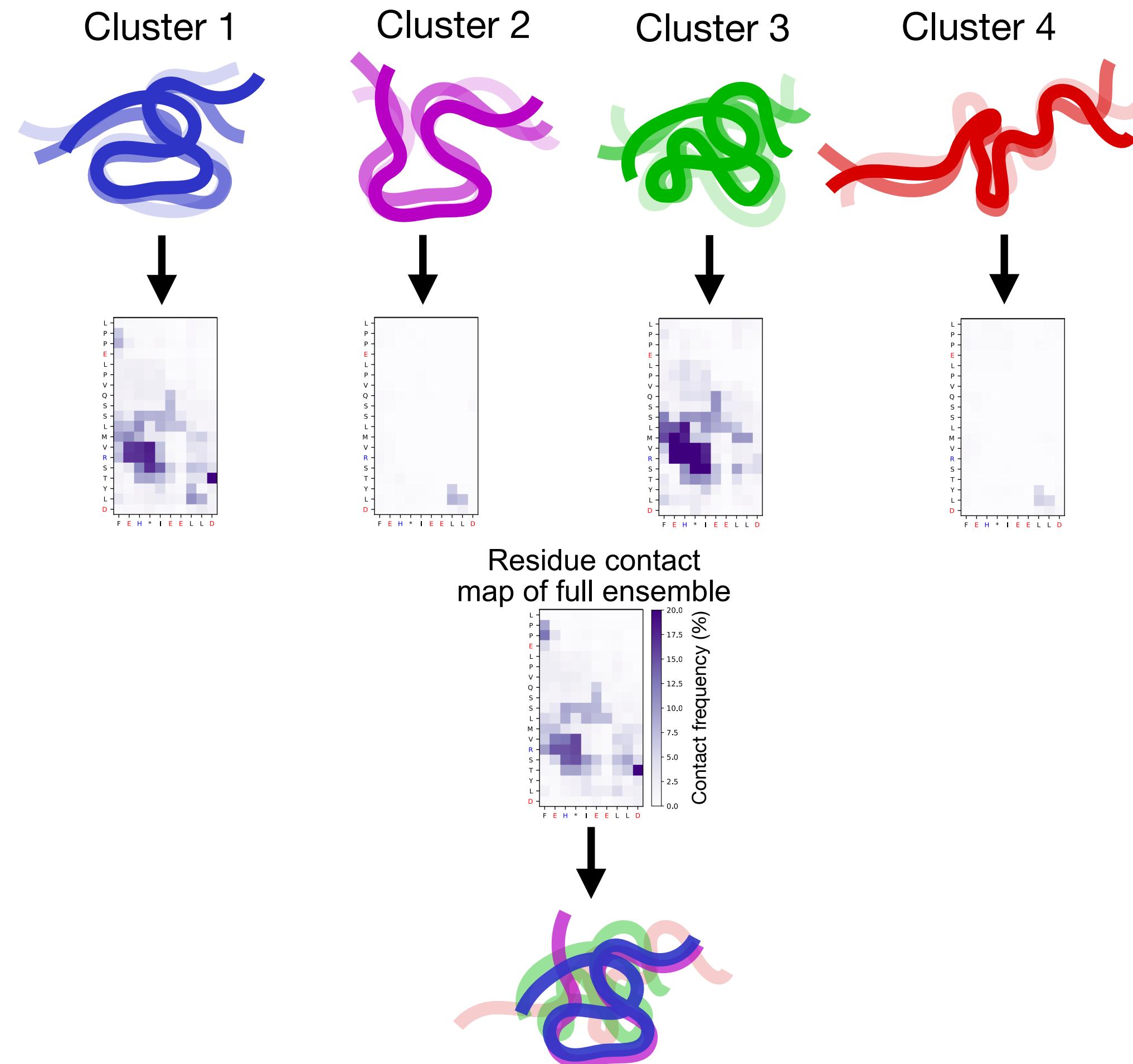
A missense mutation of an IDP can shift the population of soft-folded states in the ensemble without creating new states.

Challenges of interpreting the effects of a mutation

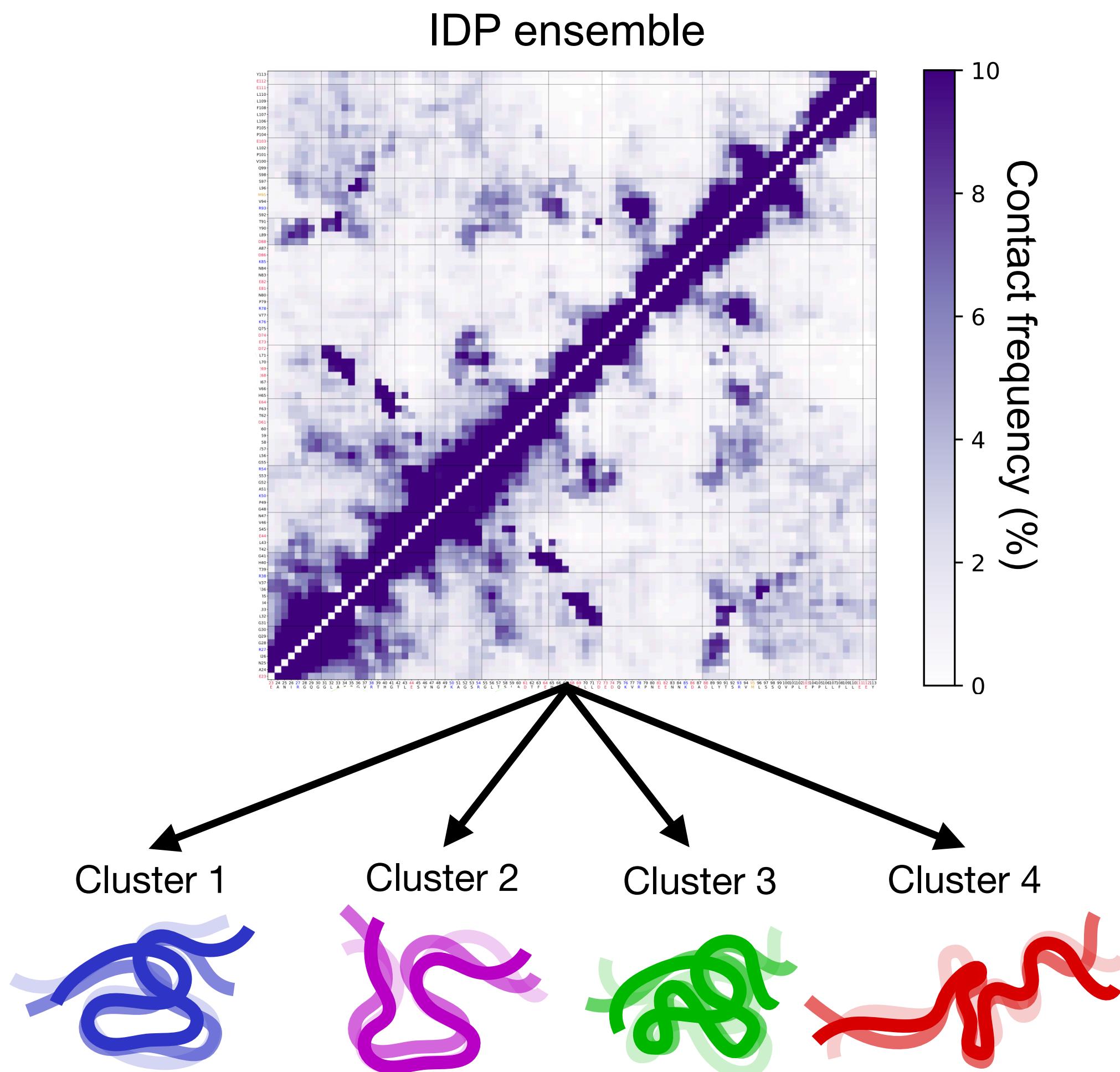
- The effects of a mutation can be **subtle**.
- Residue-residue Contact Maps are **noisy**.
- Represents an overlay of **multiple conformations** of the ensemble.



Ensembles are composed of clusters of conformations

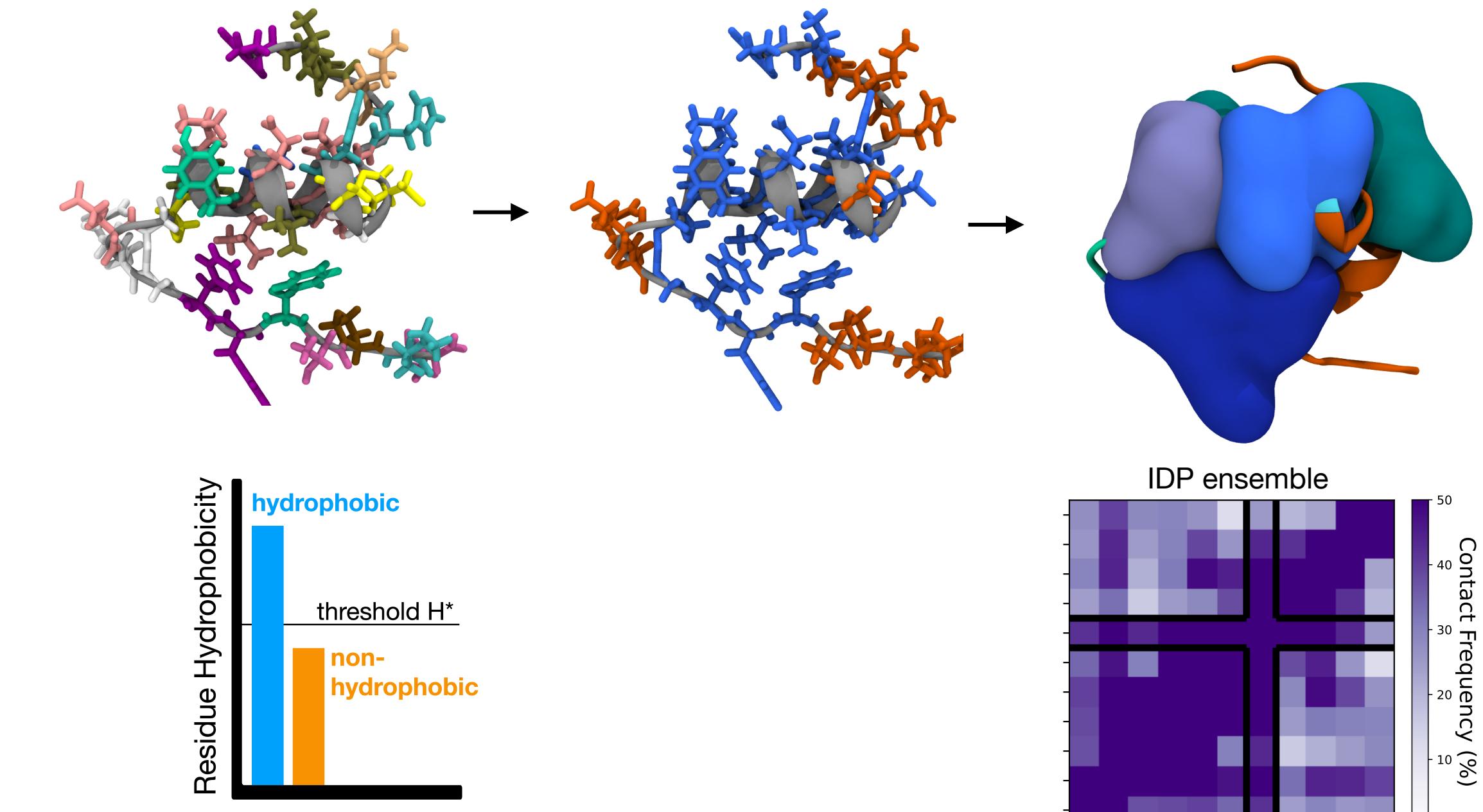


Goal: Determine clusters of conformational states that make up the ensemble



How can we define clusters?

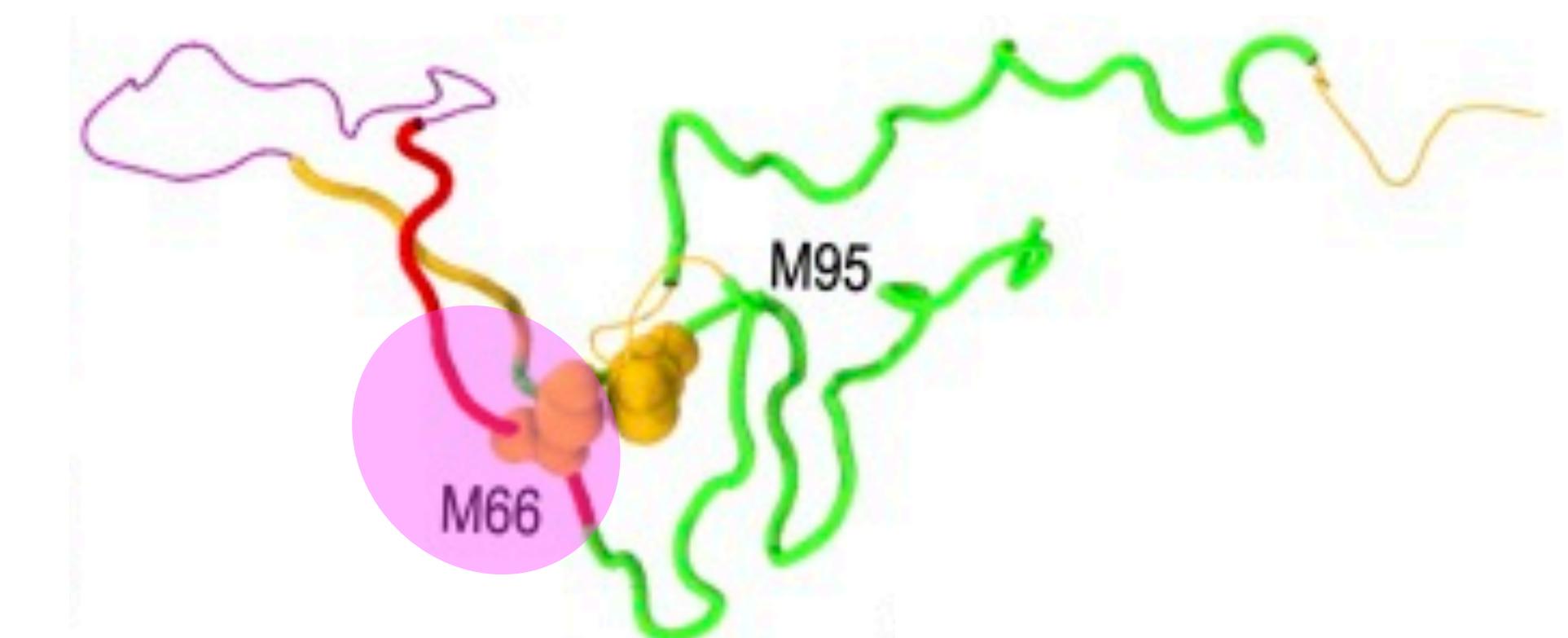
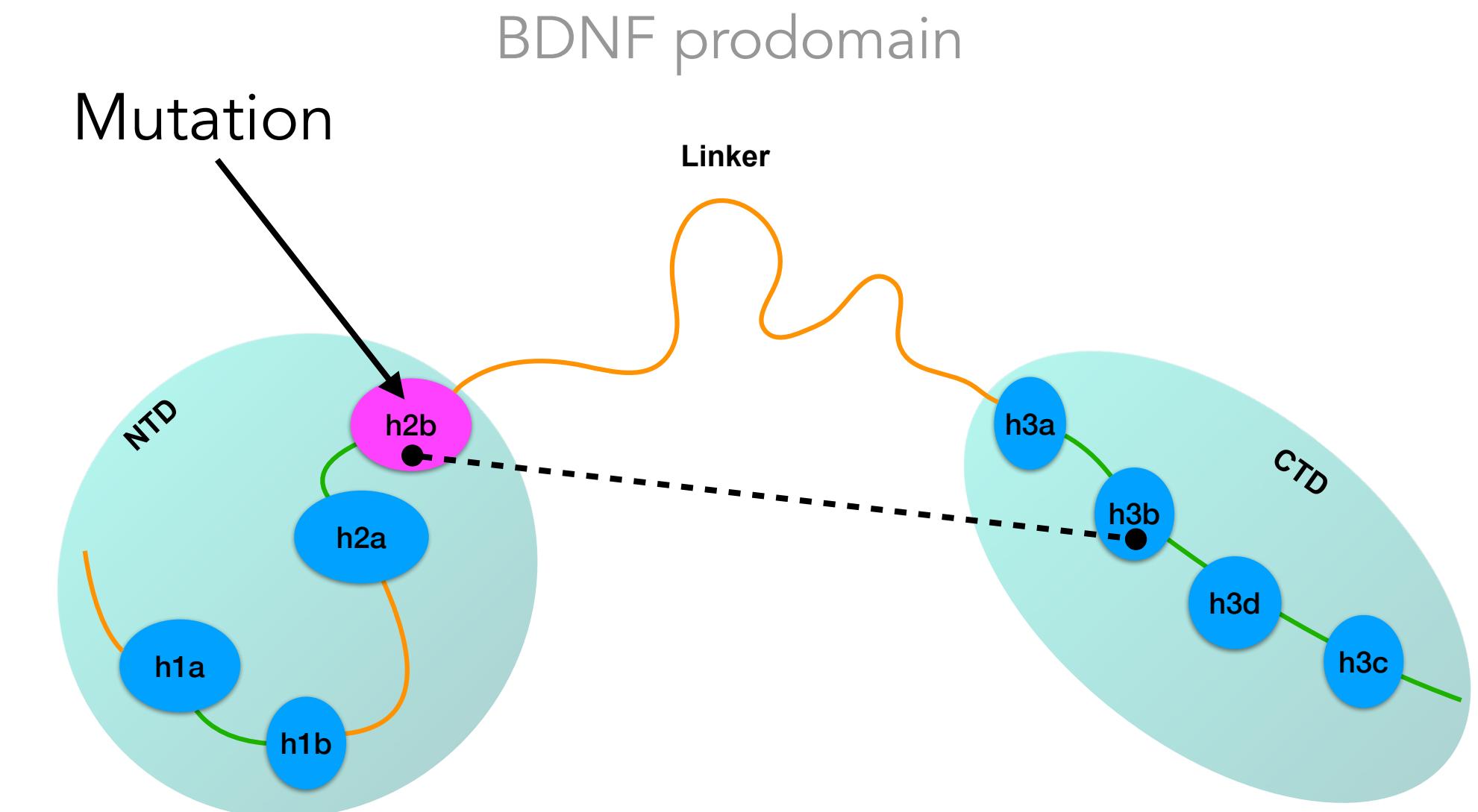
Solution: Reduce dimensionality with Blobulation



R. Lohia, M.E.B. Hansen, and G. Brannigan. PNAS, 2022.

Blobulation-based clustering revealed key interaction

- Val66Met mutation of the Brain-Derived Neurotrophic Factor (BDNF) prodomain **increases intra-domain interactions.**
- Despite the mutation being uncharged, it **alters tertiary interactions and increases compactness** of the ensemble.
- We questioned whether **any neutral-charged, hydrophobic amino acid** could shift the ensemble.



Lohia, Ruchi et al. PLOS Comp Bio, 2019

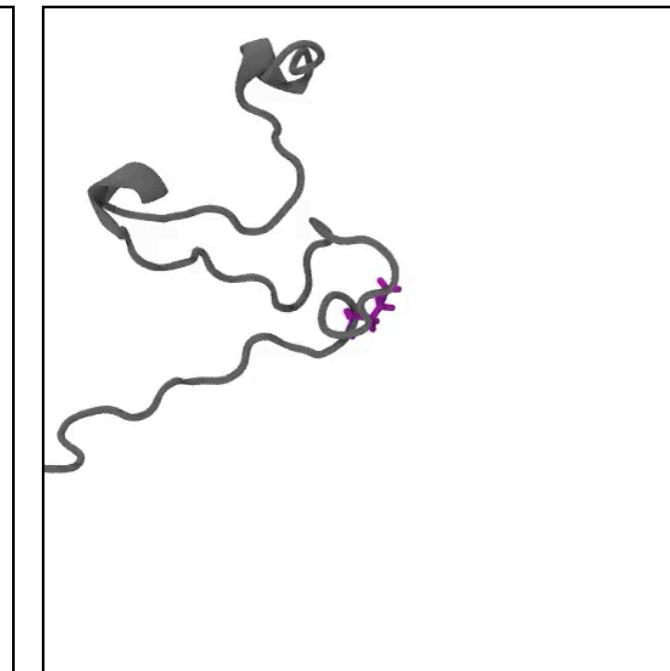
Approach

Molecular dynamics simulations

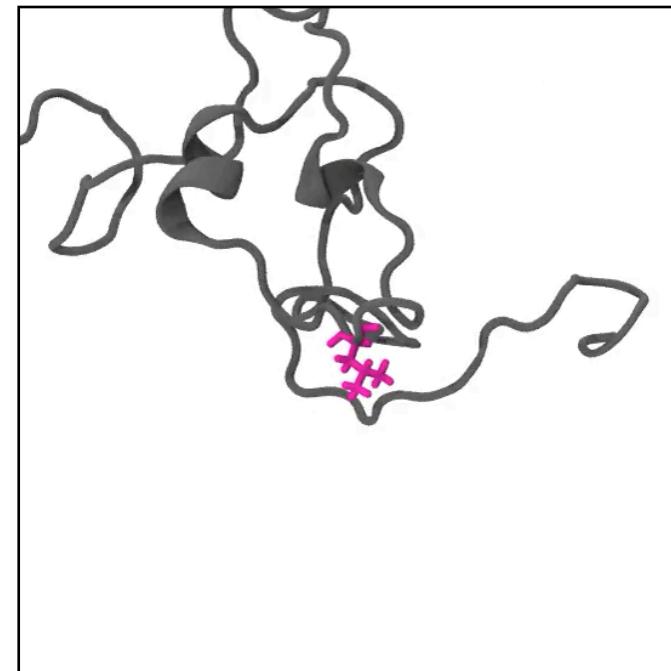
Valine



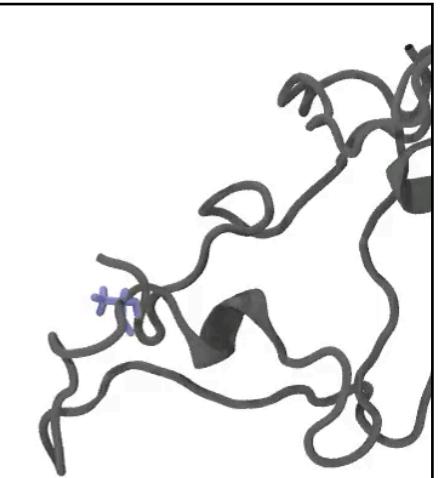
Methionine



Leucine



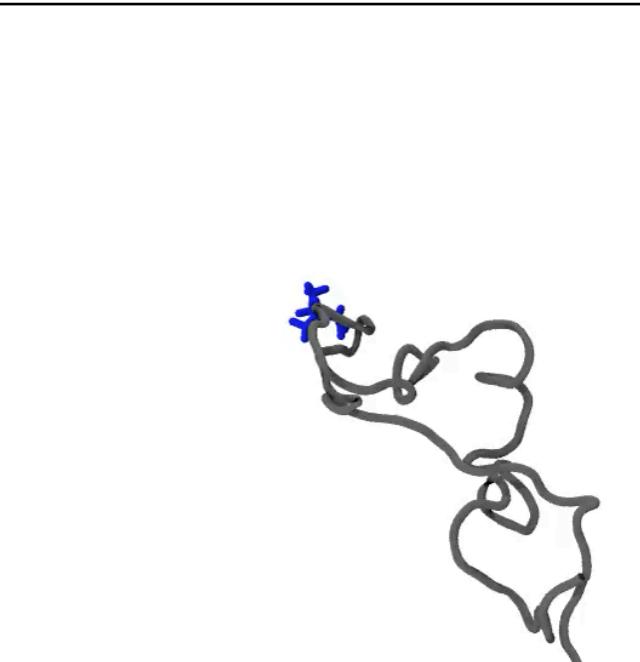
Alanine



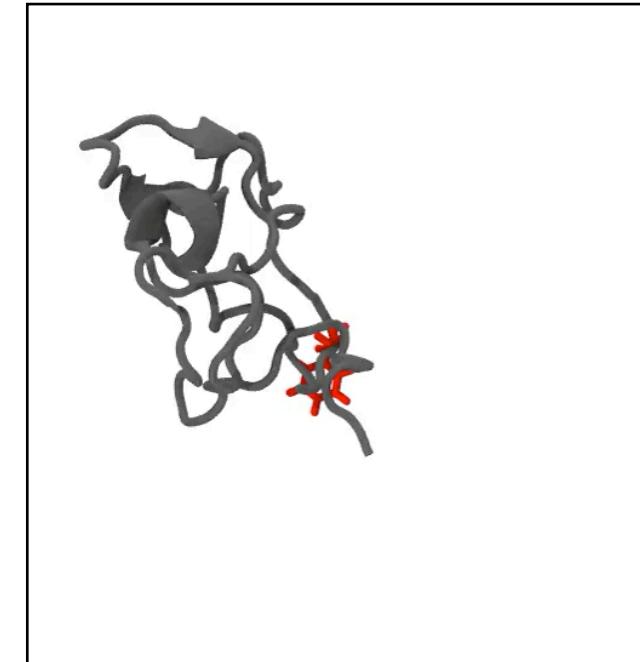
Tyrosine



Isoleucine

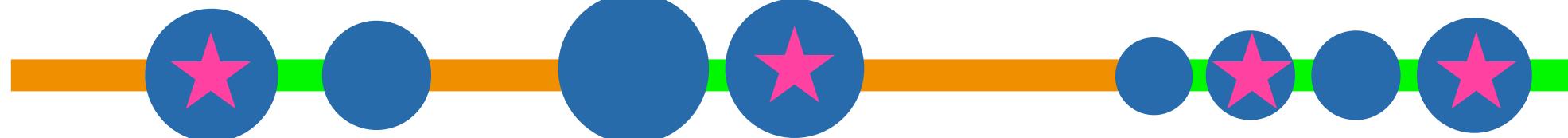
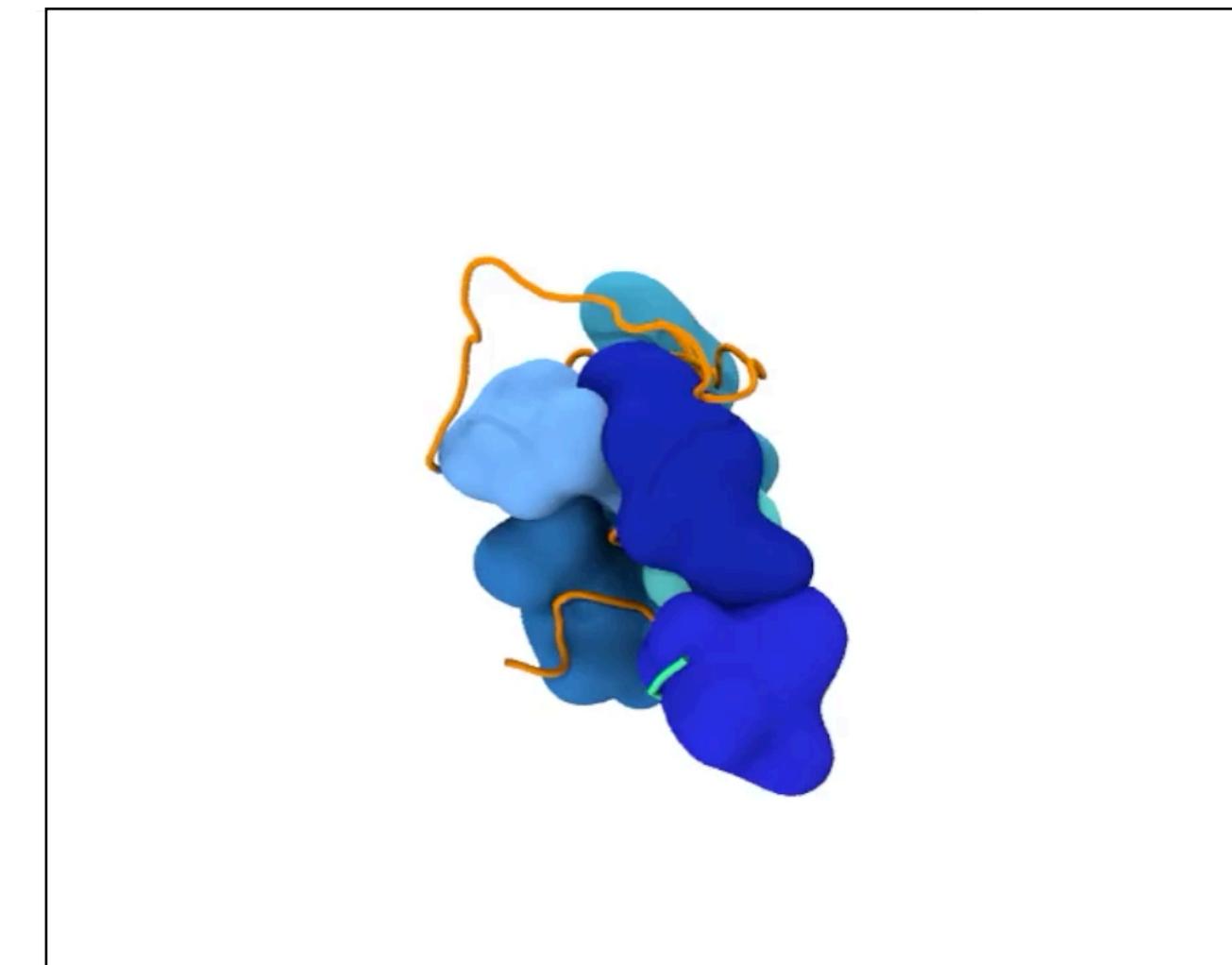


Phenylalanine



Blobulation & Clustering

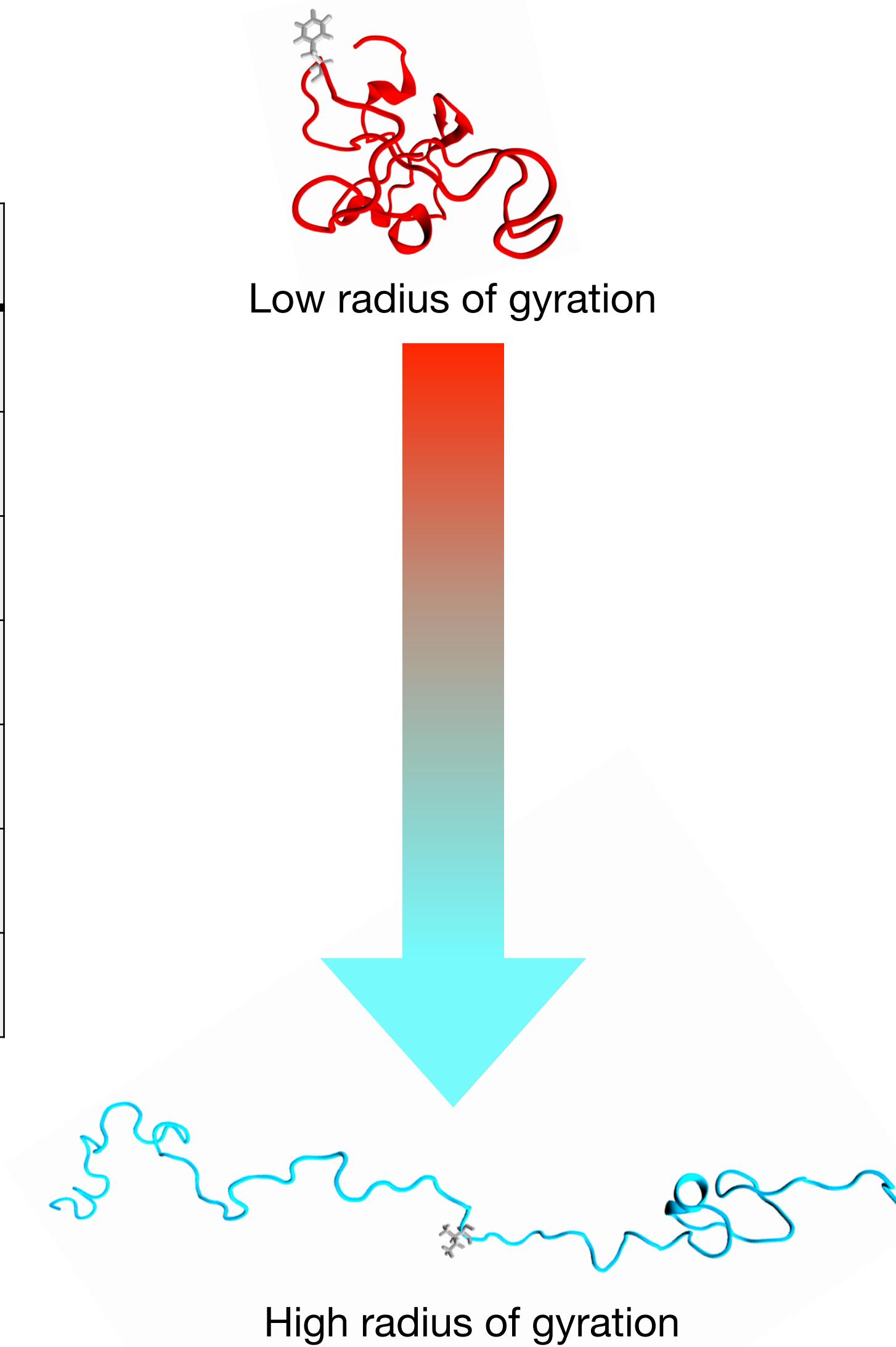
Blobulated BDNF prodomain



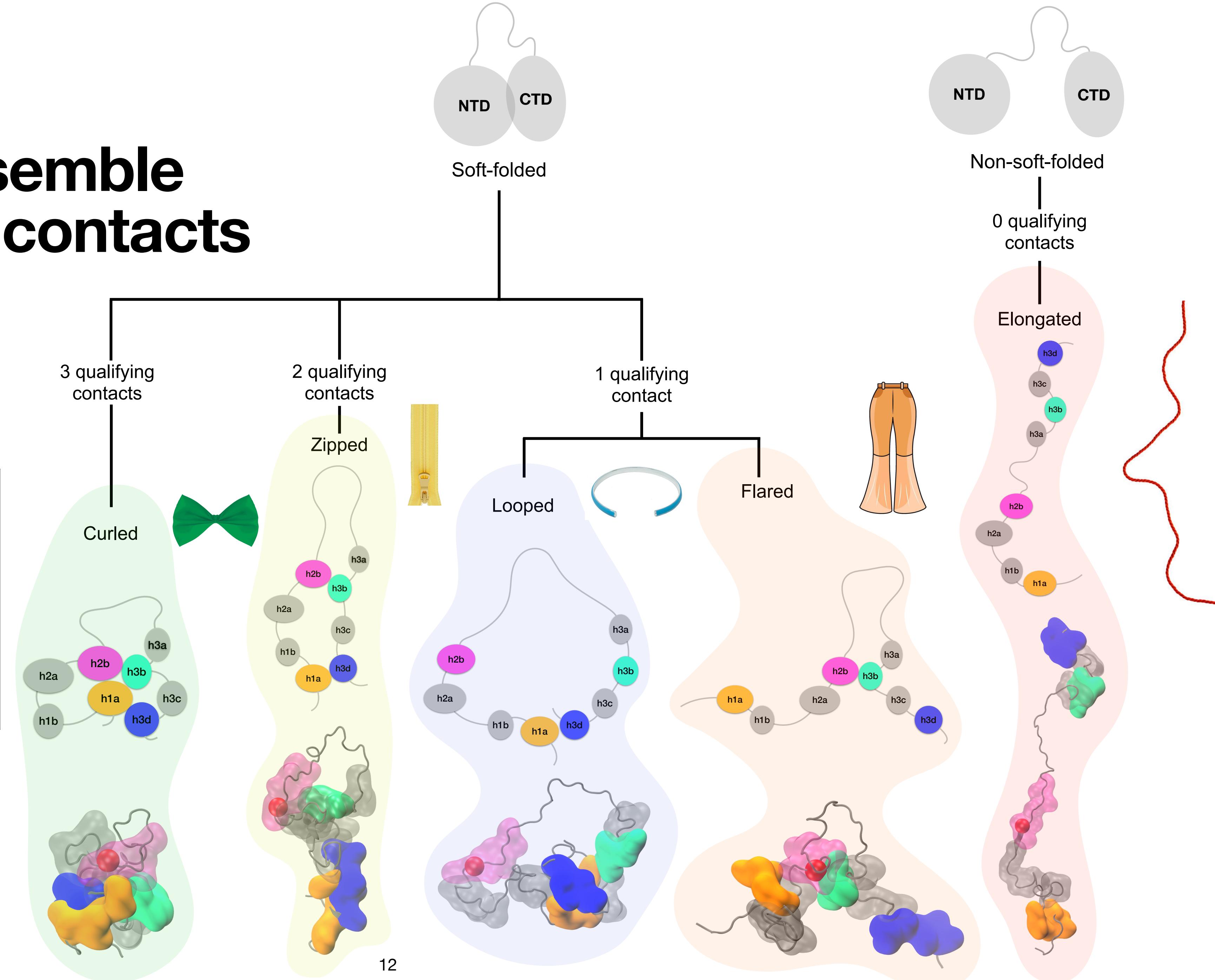
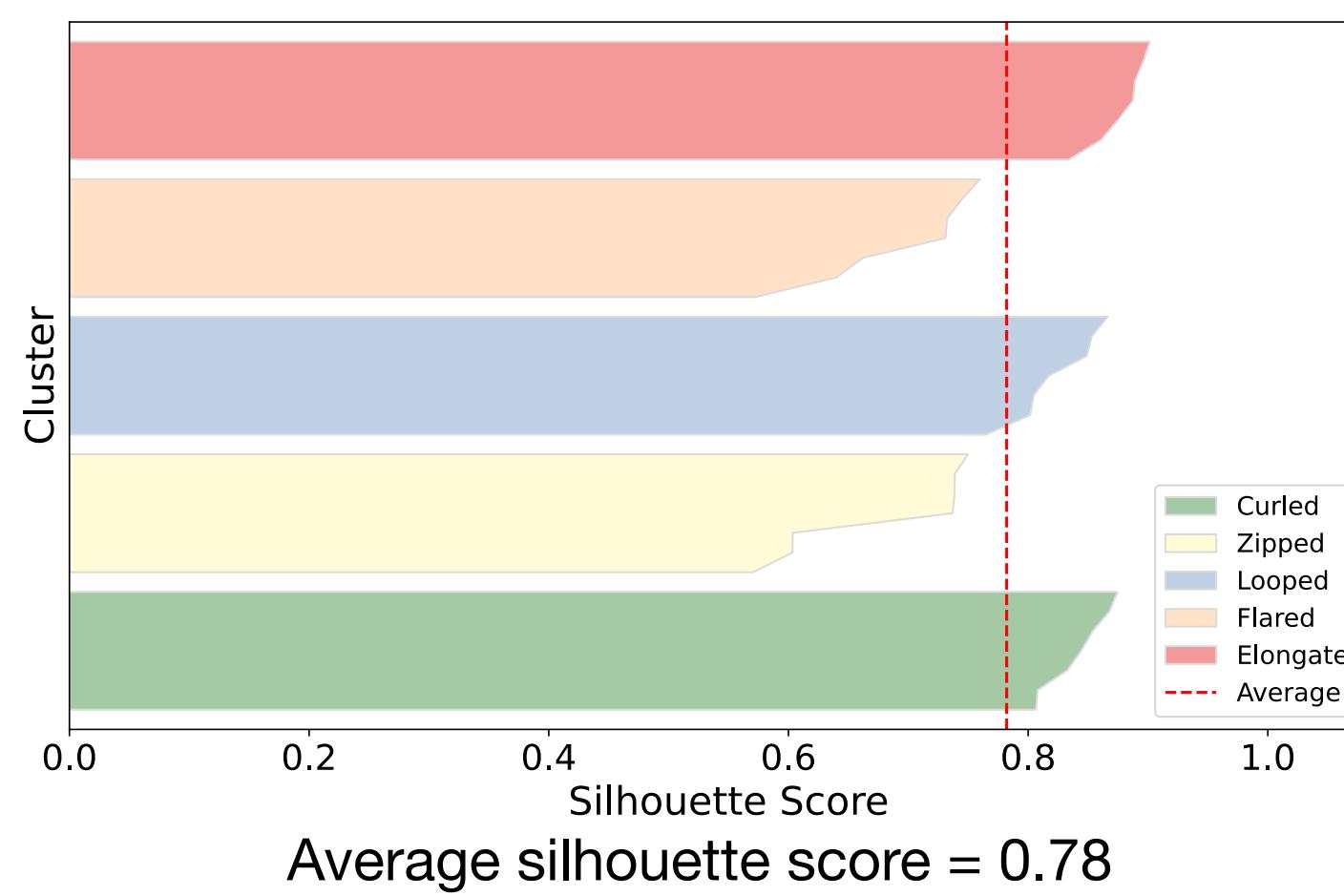
● h-blob
— p-blob
— s-blob

Amino acid at site 66 affects ensemble compactness

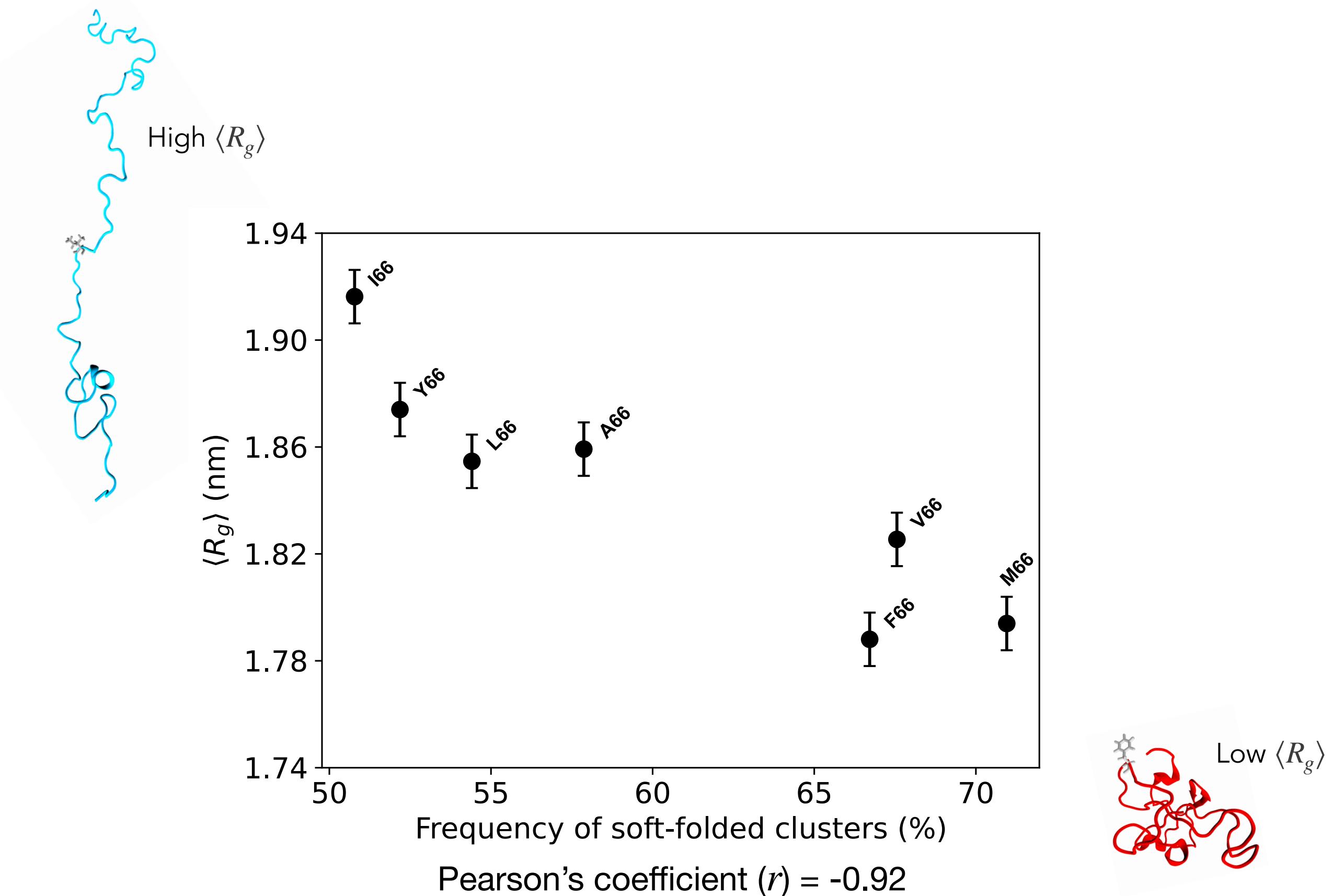
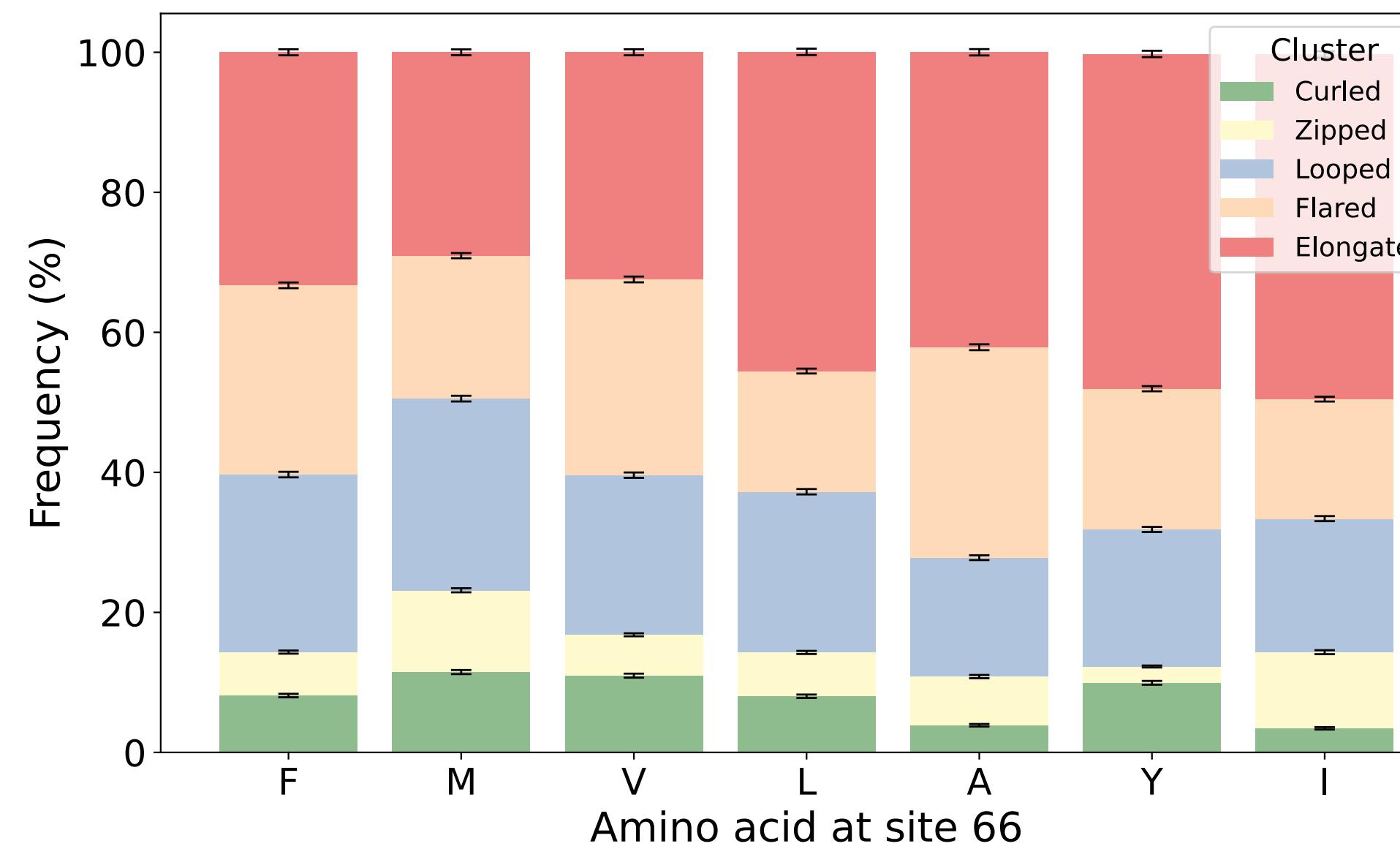
| Sequence | Radius of gyration (nm) |
|----------|-------------------------|
| F66 | 1.78 ± 0.01 |
| M66 | 1.79 ± 0.01 |
| V66 | 1.82 ± 0.01 |
| L66 | 1.85 ± 0.01 |
| A66 | 1.85 ± 0.01 |
| Y66 | 1.87 ± 0.01 |
| I66 | 1.91 ± 0.01 |



Clustering the prodomain ensemble based on blob contacts

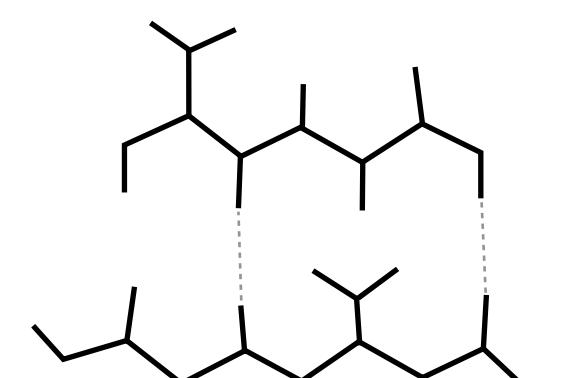
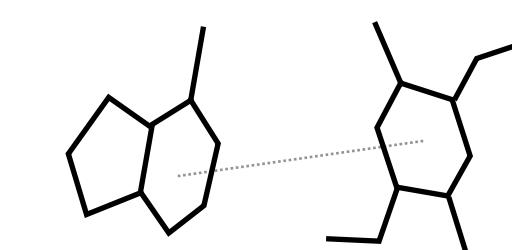
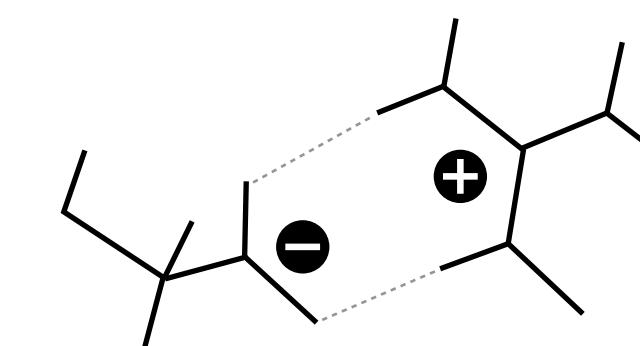
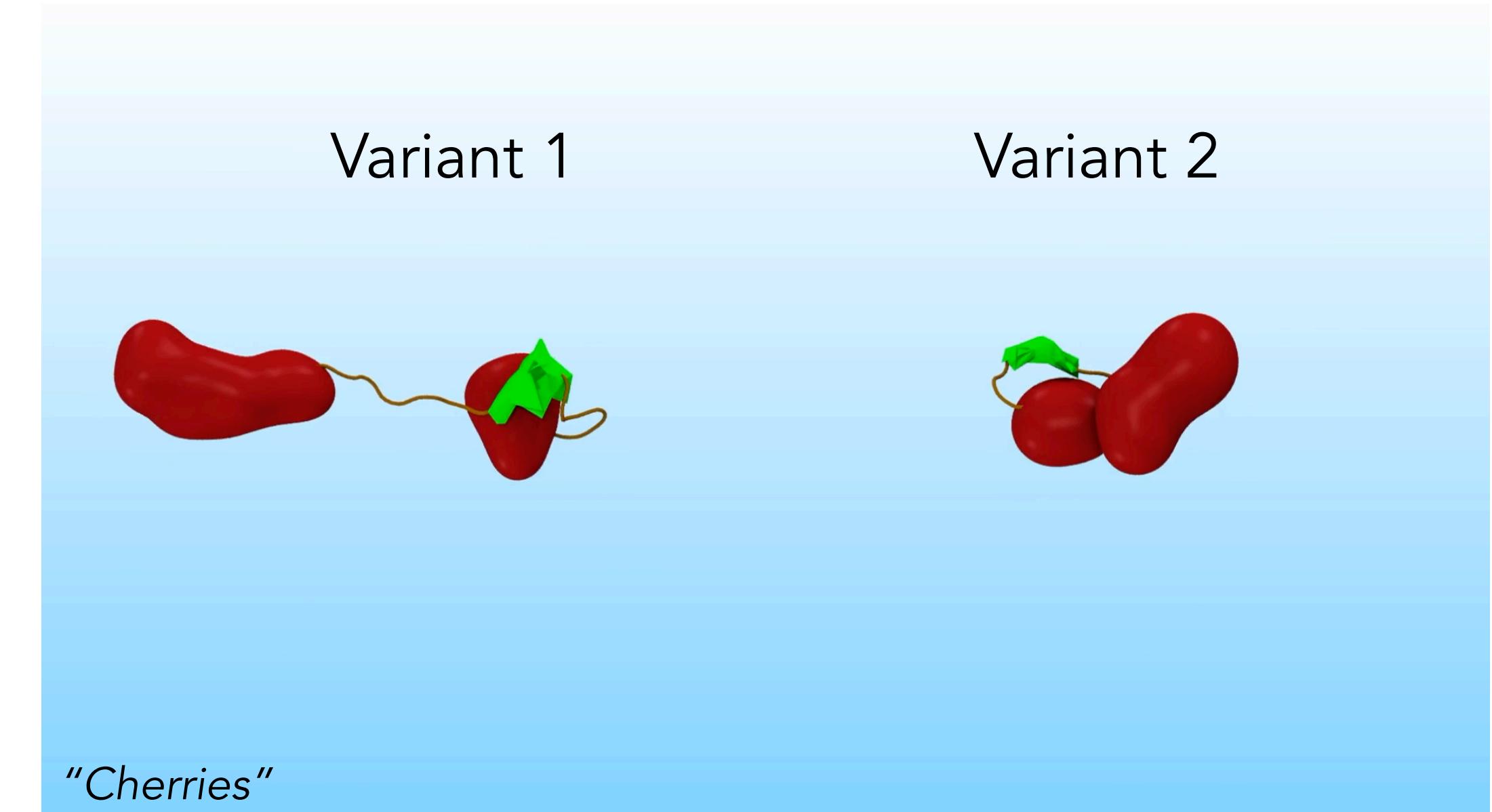


Amino acid at site 66 affects population of clusters



Summary

- **Interactions between hydrophobic blobs are sensitive** to residue 66 in the BDNF prodomain sequence.
- The amino acid at site 66 **shifts the population of soft-folded states** without creating new states.
- Population of soft-folded states is **correlated with the compactness** of the ensemble.
- Future analysis: How are clusters stabilized?



Acknowledgements

Lab members:

Dr. Grace Brannigan

Dr. Ruchi Lohia

Connor Pitman

Jahmal Ennis

Ezry Santiago-McRae

Jesse Sandberg

Ryan Lamb

Asim Dave

Regina Salzer

Resources

Rutgers Office of Advanced
Research and Computing (OARC)

NRT, NSF DGE 2152059

