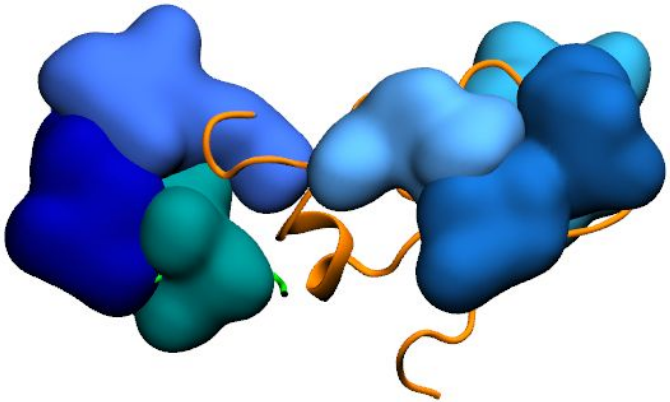
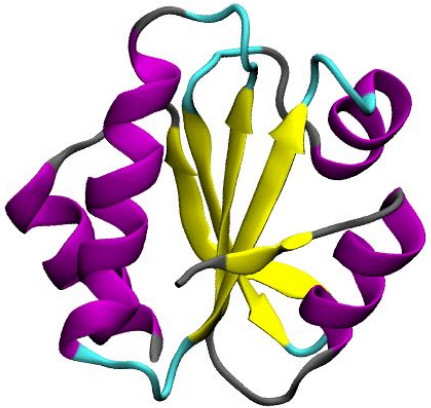
Tertiary interactions of contiguous hydrophobic residues in peptides.

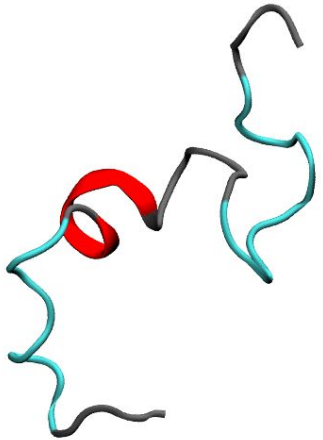


Theme: Intra Protein Interactions

Not all intra-protein interactions involve structural motifs



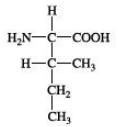
Structured Protein



Peptides and IDPs

Modules are traditionally assigned by secondary structure

Residue

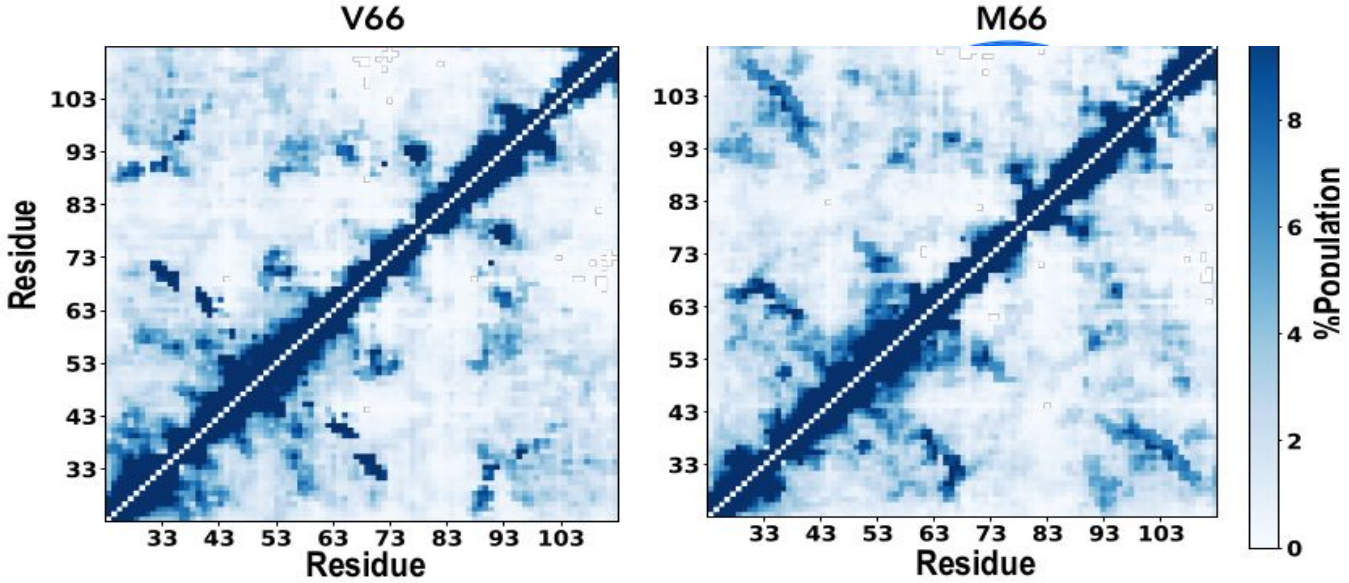


isoleucine
(Ile, I)

Secondary Structure


β - pleated sheets

Contacts between residues are hard to interpret



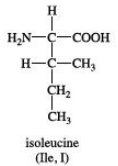
Residue by residue contacts for val66 BDNF

Residue by residue contacts for met66 BDNF

Lohia et al., PLoS Comput Biol. 2019

Modules are traditionally assigned by secondary structure

Residue

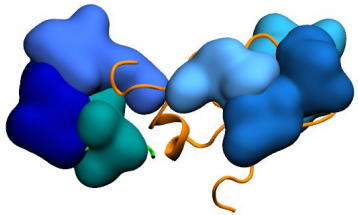


isoleucine
(Ile, I)

Secondary Structure

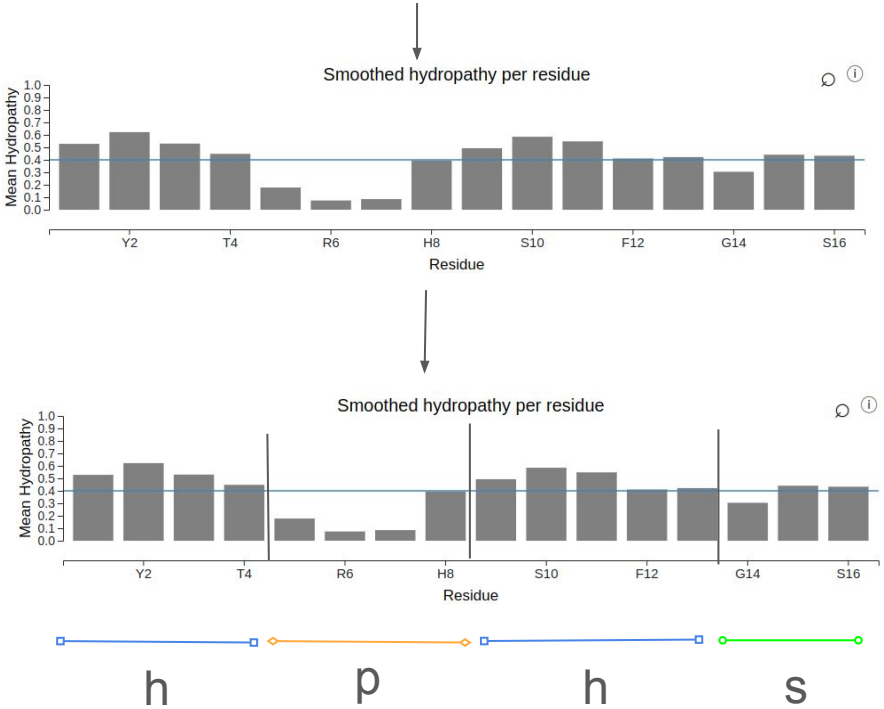

β - pleated sheets

Contiguous Hydrophobic clusters



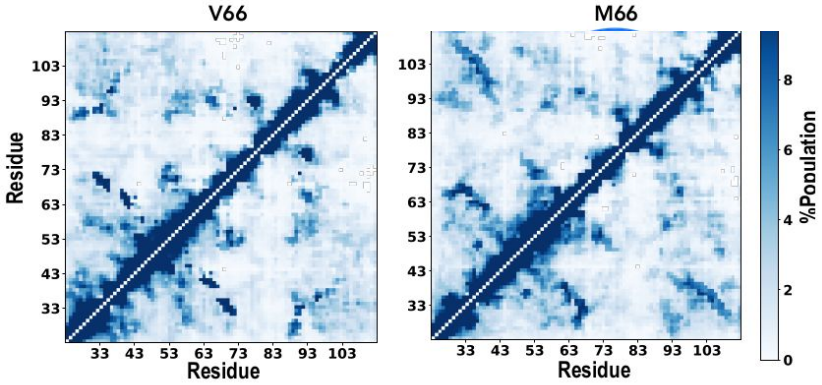
Blobulation defines clusters by hydrophobicity

AYFTQRNHLSTFRGGS



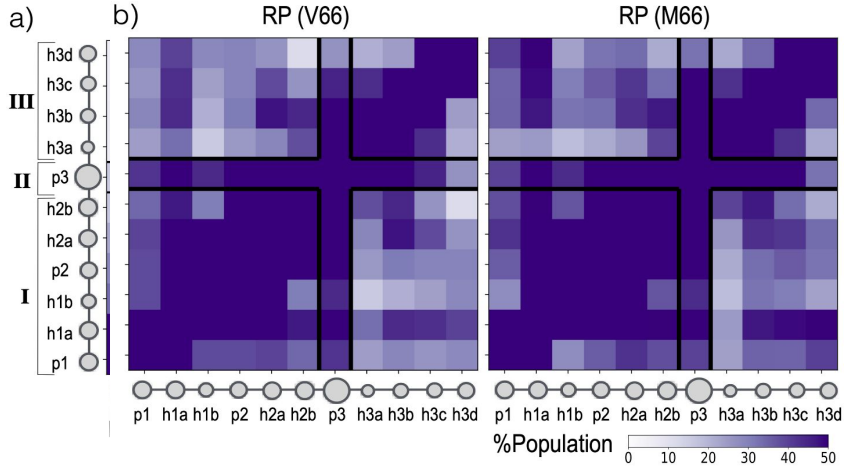
Blobulated Sequence

Blobulation offers a cleaner method to observe tertiary interactions



Residue by residue contacts for val66 BDNF

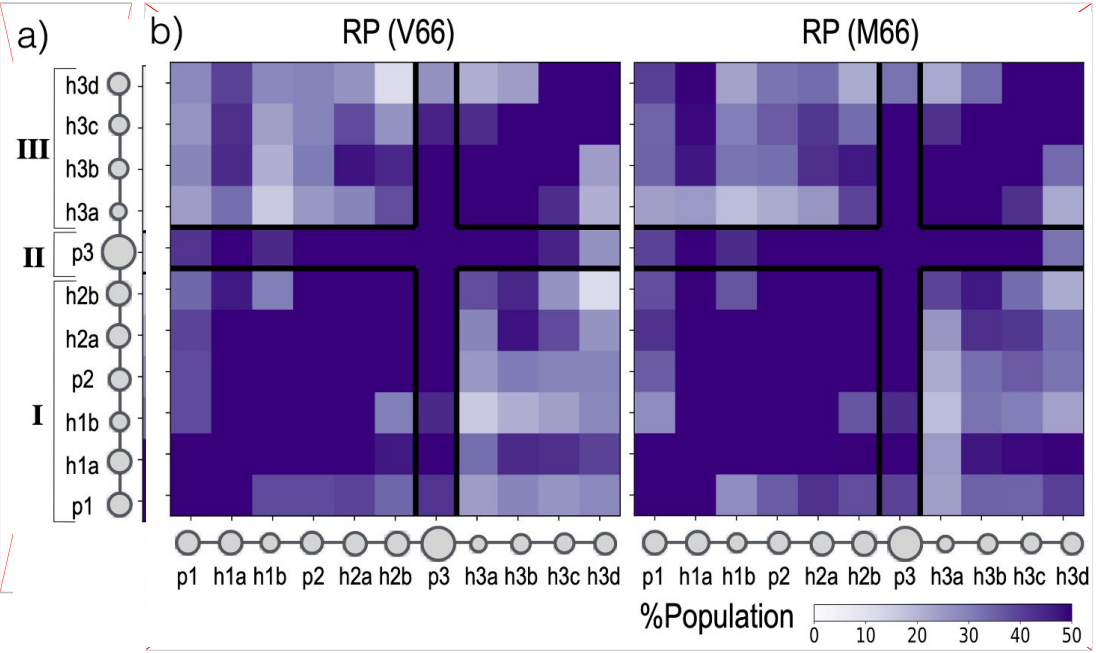
Residue by residue contacts for met66 BDNF



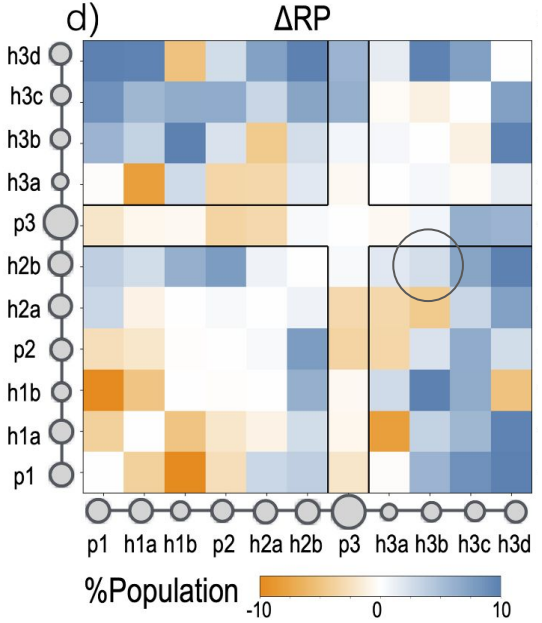
Blob-blob contact population map of BDNF with val66 and met66 respectively. Population is the ratio of frames blobs were in contact over total frames of the simulation

Lohia et al., PLoS Comput Biol. 2019

Blobulation gives insight into cluster contacts



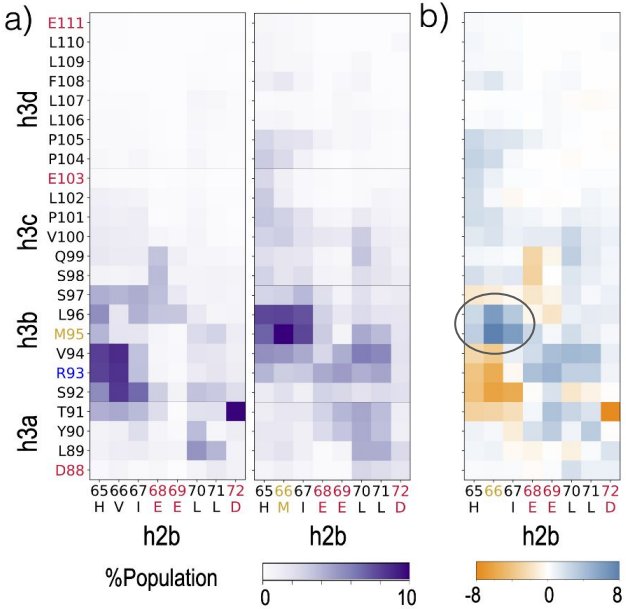
Blob-blob contact population map of BDNF with val66 and met66 respectively. Population is the ratio of frames blobs were in contact over total frames of the simulation



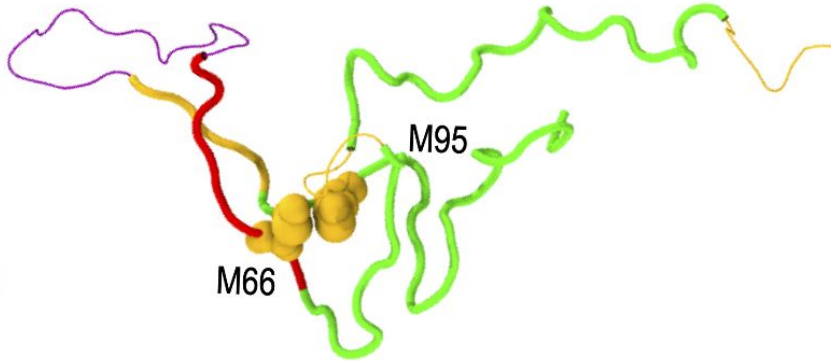
Difference between contact frequency of V66 to M66

Lohia et al., PLoS Comput Biol. 2019

Zooming into blob reveals key residue contact



Residue contacts within blob h2b. For val66 met66 and difference between each respectively



Met-met interaction in BDNF between residue 66 and residue 95

Lohia et al., PLoS Comput Biol. 2019

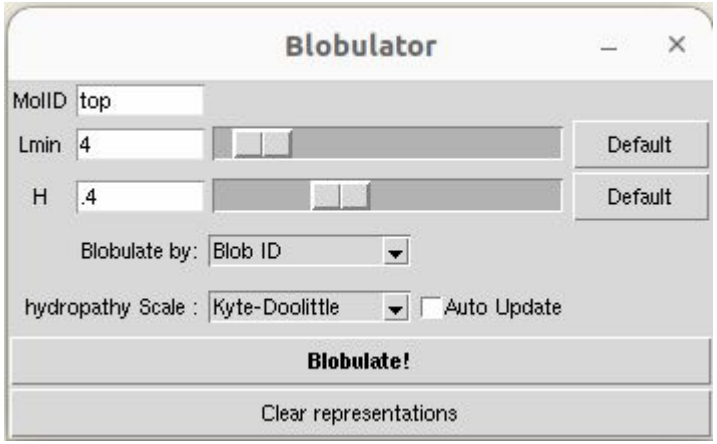
Main Question: How do h-blobs influence tertiary interactions in peptides?

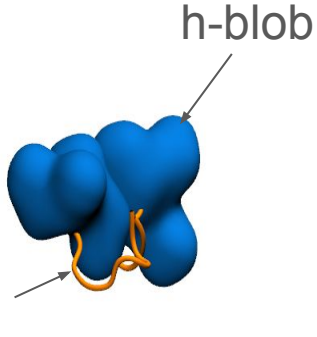
Preliminary Work

VMD Blobulation GUI

- Prior to joining the lab, blobulating a protein in VMD was an arduous task
  1. Blobulate sequence on webapp
  2. Download data table
  3. Load protein into VMD
  4. Run a script to set values
  5. Create Graphical representation for each blob

- New tool is a convenient alternative
  1. Load protein into VMD
  2. Blobulated with the GUI





Model peptide in new cartoon

Model peptide blobulated

Simulation(s)

Model peptide in new cartoon

Model peptide blobulated

Main Question: How do h-blobs influence tertiary interactions in peptides?

Aims: Overview

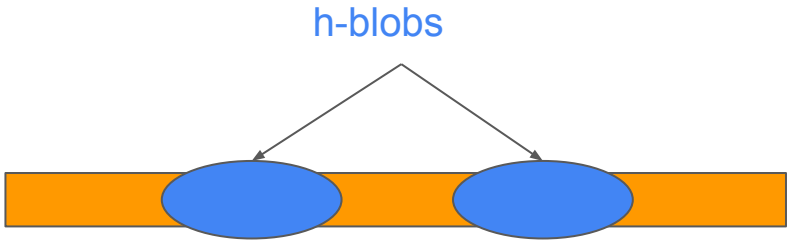
Aim 1

Main Question: How do hydrophobic properties such as length, and residue composition affect tertiary interactions in peptides?

Run 5 simulations with the following sequences

NNNNLLLLLNNNNLLLLLNNNN
NNNNLLLLLLLLLLNNNNLLLLLLLLLLNNNN
NNNNLLLLNNNNLLLLNNNNLLLLNNNNLLLLNNNN
NNNNIIIIINNNNIIIIINNNN
NNNNAAAAANNNNAAAAANNNN

Analyses: Radius of gyration, blob-blob contacts, and create network diagrams



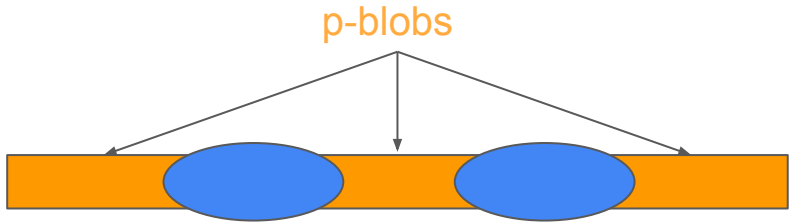
Aim 2

Main Question: Do polar blobs influence h-blobs interactions?

Run 4 simulations with the following sequences

NNRNLLLLLLLLLNNNNLLLLLLLLLNRNN
NNRNLLLLLNNNNLLLLLNNNNLLLLLNNNNLLLLLNRNN
NNNNNNLLLLLNNNNNNNLLLLLNNNNNNLLLLLNNNNNNLLLLLNNNNNN
NNNNNNLLLLLLLLLLNNNNNNLLLLLLLLLLNNNNNN

Analyses: Radius of gyration, blob-blob contacts, and create network diagrams



Aim 1: How do h-blob properties affect tertiary interactions

- Run 5 simulations of different peptide sequences each with a control sequence
- Simulation time of 1 us each
- Measure radius of gyration, and blob - blob contacts through every frame of simulation
- Create network diagrams off blob - blob contact data
- Measure change in compactness between the experimental sequence and the respective control sequence

Aim 1: Sequences are made off threshold parameters

We will run these sequences in atomistic MD simulations

h-blobs

p-blobs

NNNNLLLLLNNNNLLLLLNNNN

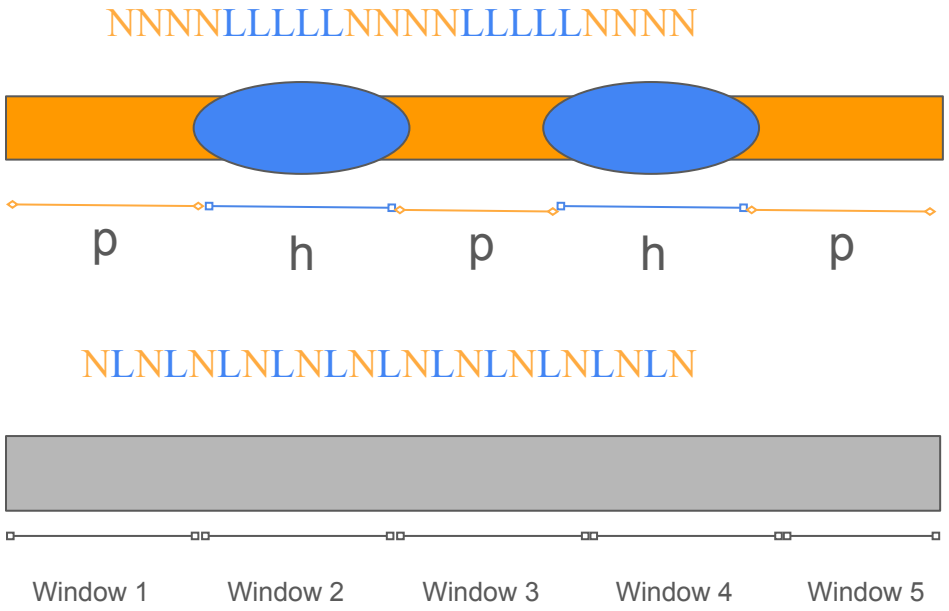
NNNNLLLLLLLLLNNNNLLLLLLLLLNNNN

NNNNLLLLLNNNNLLLLLNNNNLLLLLNNNNLLLLLNNNN

NNNNI I I I INNNNI I I I INNNN

NNNNAAAAANNNNAAAAANNNN

Negative Control: Testing if the order of amino acids influence tertiary interactions



For each simulation

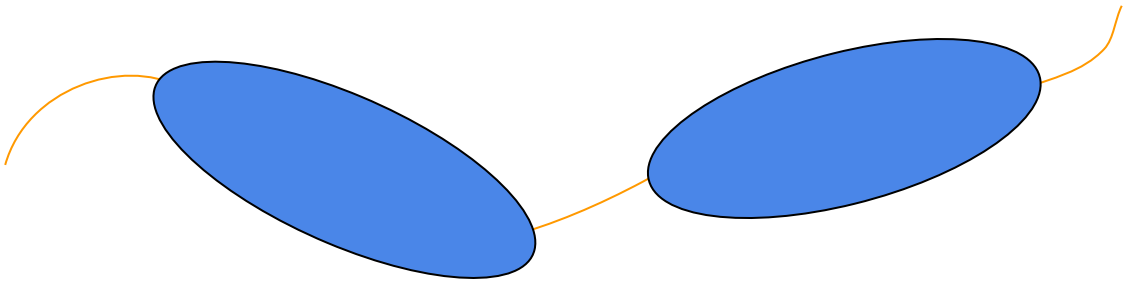
Aim 1: Long stretches of hydrophobicity

We will run these sequences in atomistic MD simulations

NNNNLLLLLLLLLLNNNNLLLLLLLLLLNNNN

h-blobs

p-blobs



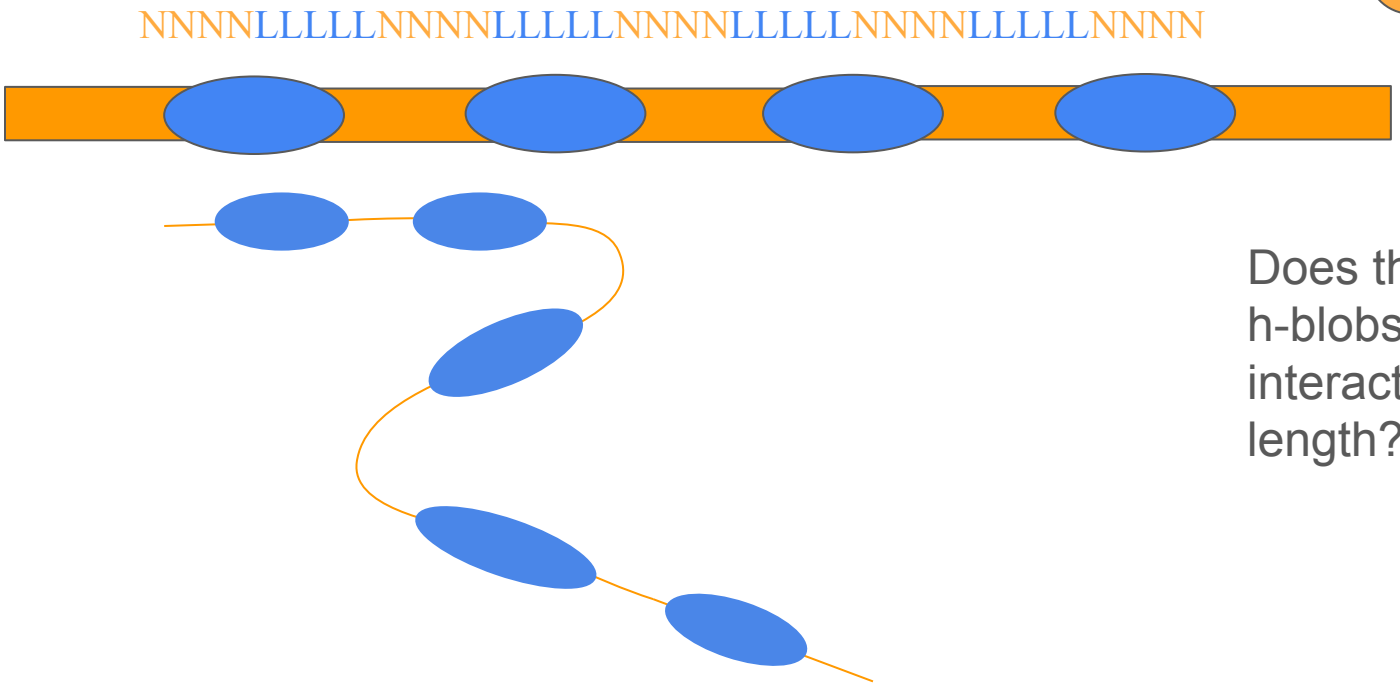
Long contiguous hydrophobic residues produce exponentially stronger tertiary interactions between h-blobs

Aim 1: Could h-blob quantity match length?

We will run these sequences in atomistic MD simulations

h-blobs

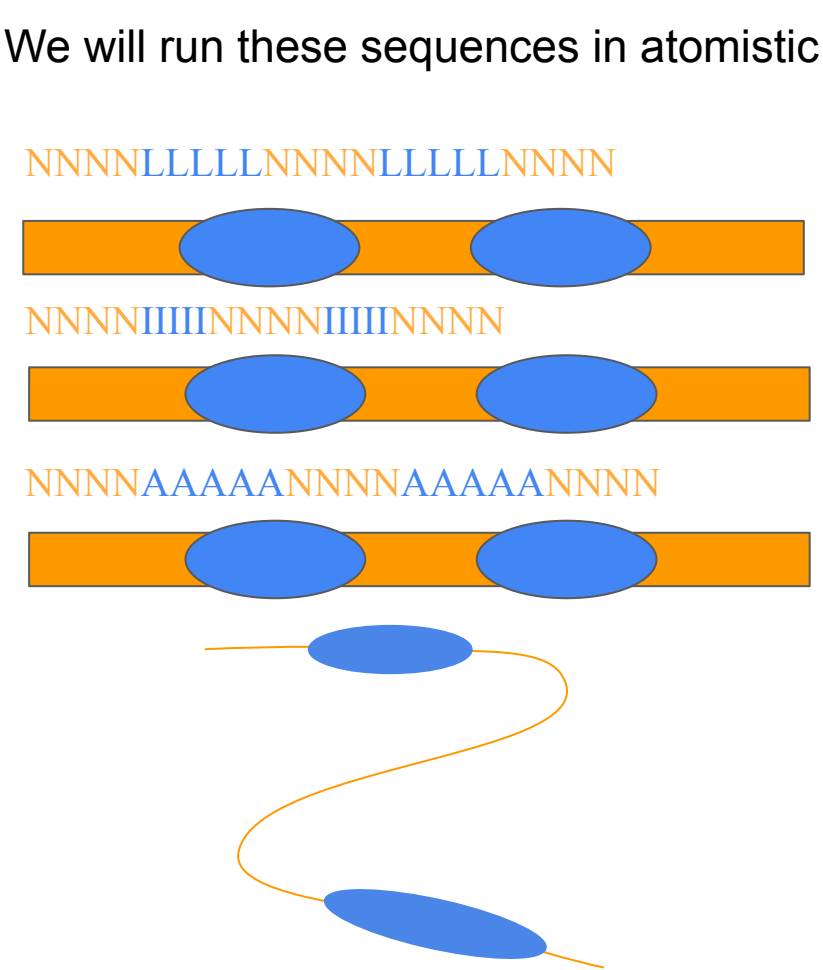
p-blobs



Does the quantity of h-blobs influence tertiary interactions more than length?

Aim 1: Do stronger hydrophobic residues influence tertiary interactions?

We will run these sequences in atomistic MD simulations



h-blobs

p-blobs

These sequences focus more on the residue composition.

Aim 1: Expectation of contacts from more contacts to less

1. NNNNLLLLLLLLLLNNNNLLLLLLLLLLNNNN

2. NNNNLLLLLNNNNLLLLLNNNNLLLLLNNNNLLLLLNNNN

3. NNNNLLLLLNNNNLLLLLNNNN

4. NNNN I I I I I NNNN I I I I INNNN

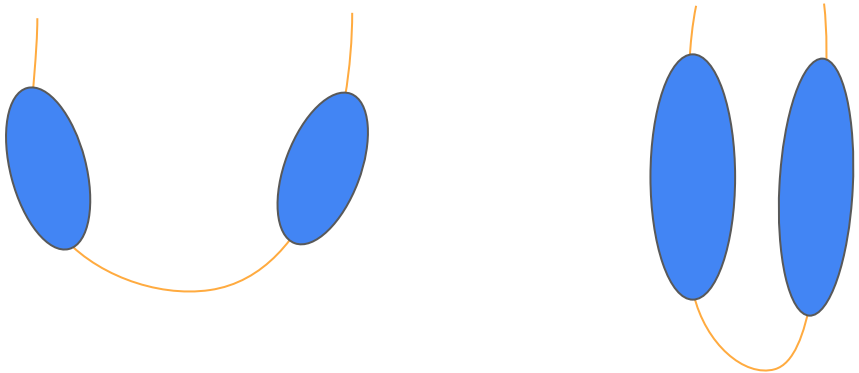
5. NNNNAAAAANNNNAAAAANNNN

More Contacts

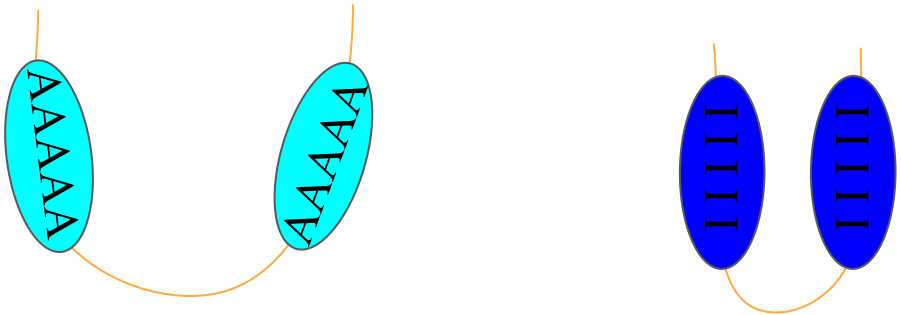
Less Contacts

Hypothesis: Blob properties correlate to tertiary interactions

Length of blobs



Residue composition of blobs



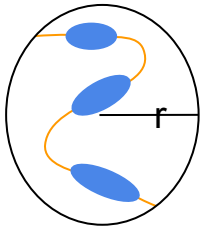
How will we characterize tertiary interactions

Compactness of the peptide

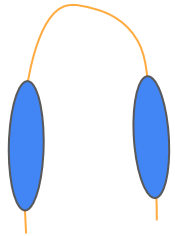
blob - blob contacts

Analyses

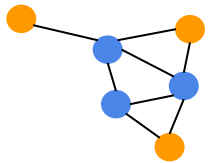
Radius of Gyration



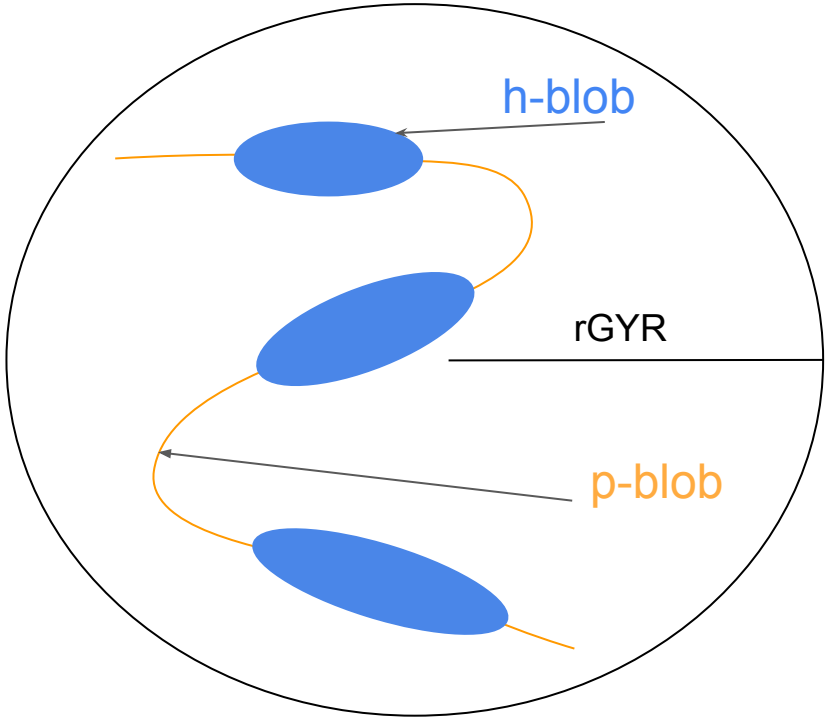
Blob-Blob Distance



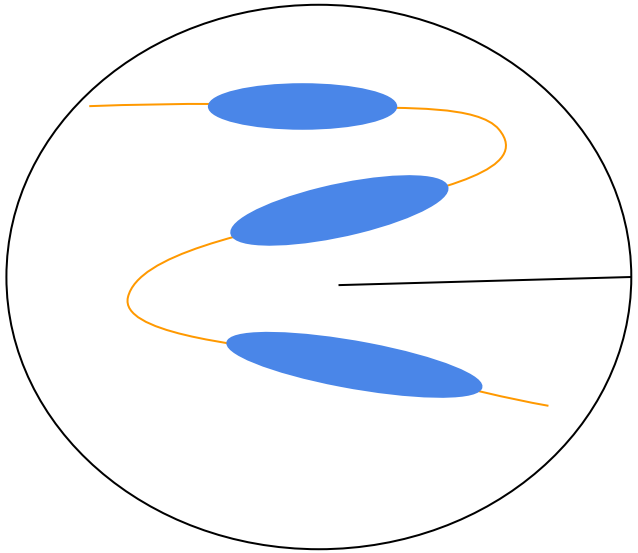
Network Diagrams



Radius of Gyration



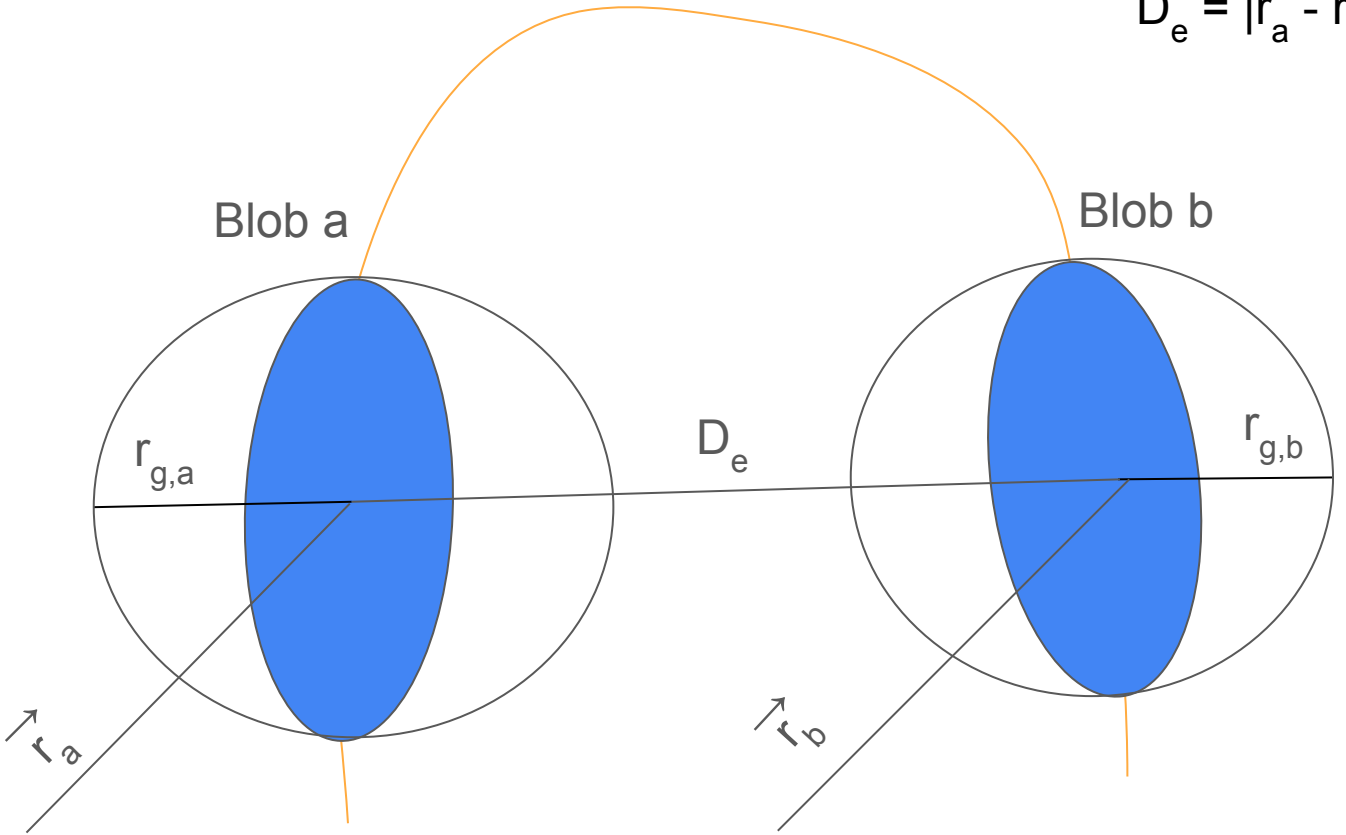
Non-Compact Peptide



Compact Peptide

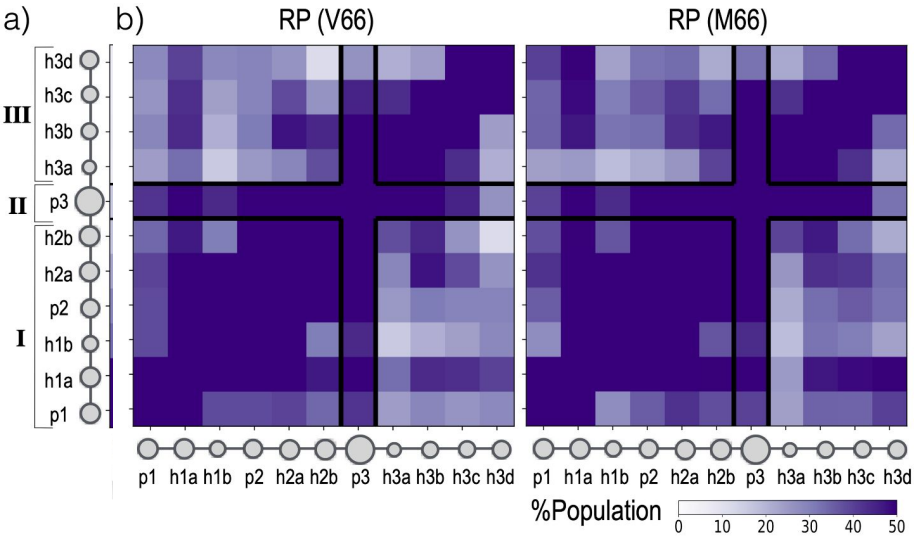
Blob-Blob Contacts

$$D_e = |\vec{r_a} - \vec{r_b}| - (R_{g,a} + R_{g,b})$$



If D_e is less than 0.55 nm the blobs are considered touching

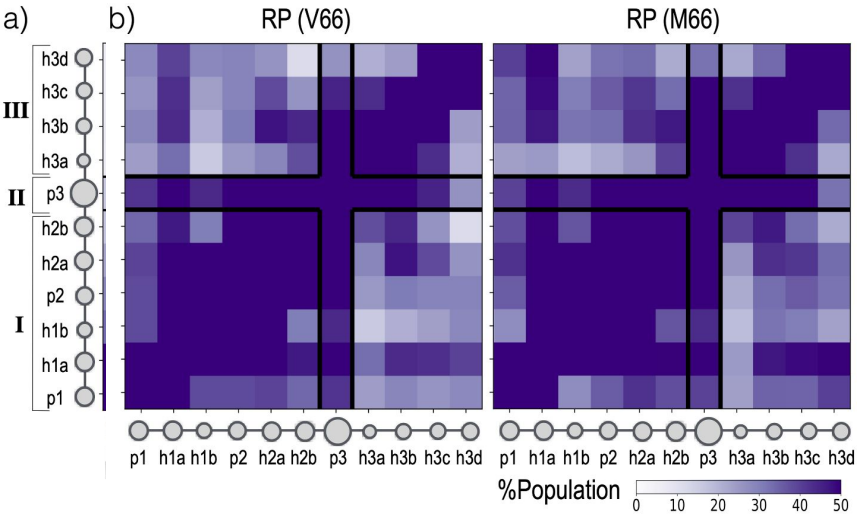
Blob-Blob Contacts: A measurement of contact over time



$$\%\text{Population} = \frac{\text{Frames of Blobs Contacts}}{\text{Total Frames}}$$

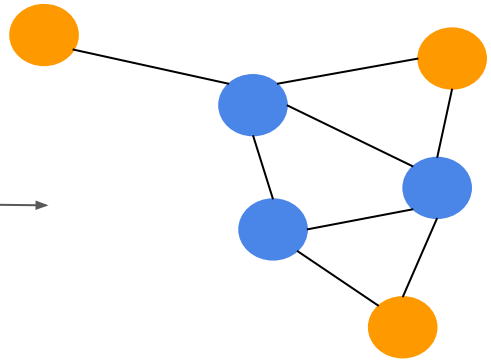
Lohia et al., PLoS Comput Biol. 2019

Network Diagrams: Examining connections for intra protein interactions



D_e

From each blob-blob contact



Lohia et al., PLoS Comput Biol. 2019

Network Diagrams: visualize blob contacts in a different context

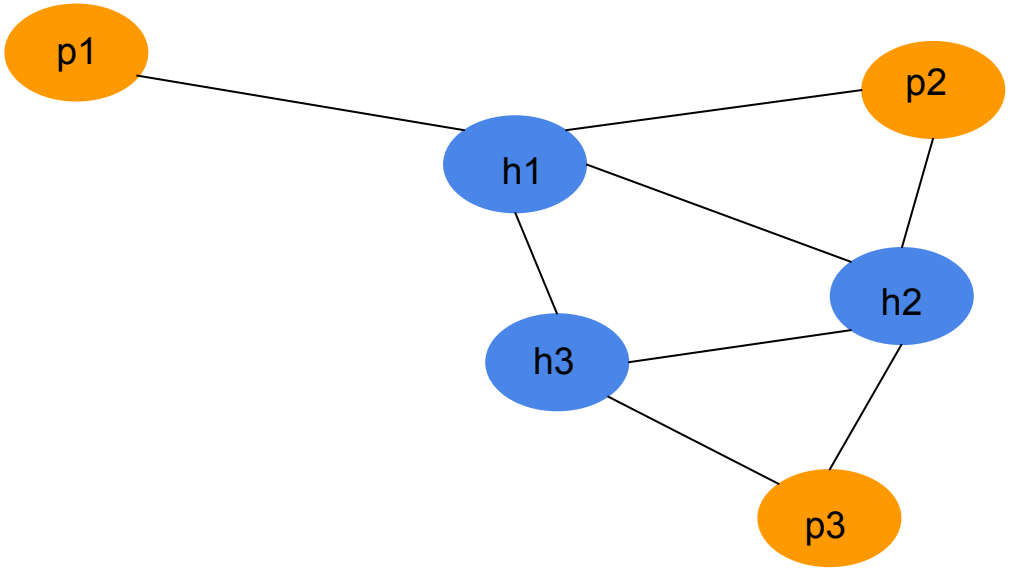


Image of a potential network diagram for our model peptide.

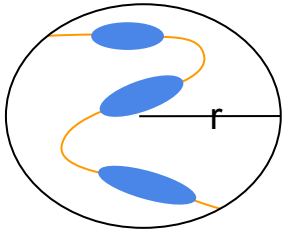
Nodes (blobs)

Edges (distance)

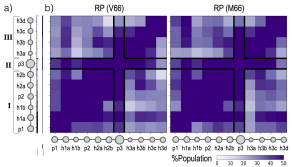
Using D_e from blob-blob contact frequency

Aim 1: How do hydrophobic properties such as length, residue composition affect tertiary interactions in peptides?

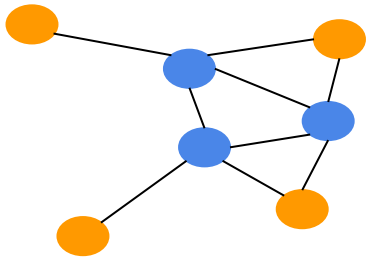
- Determine the most compact sequence by running simulations with different peptides.



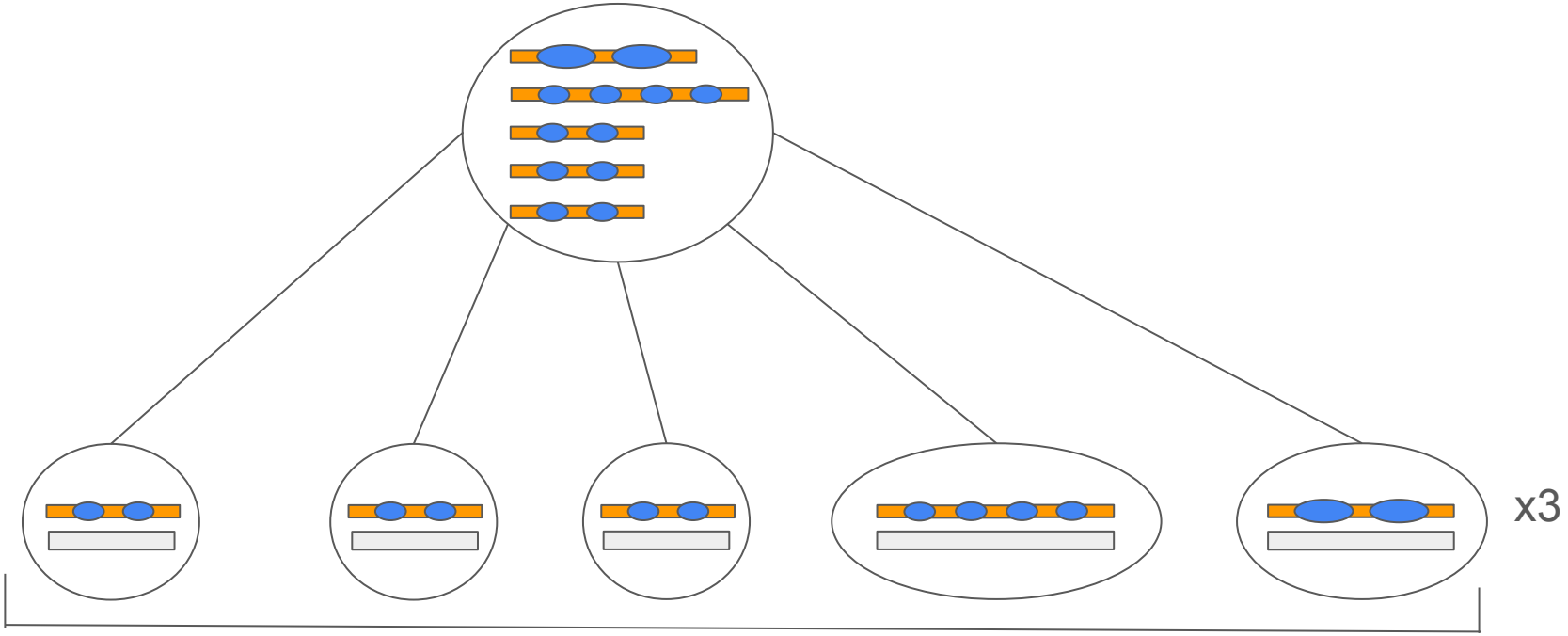
- Find the sequence with the most enriched blob-blob contacts.



- Demonstrate the average distance of blob-blob contacts in 2d space with network diagrams.



Deliverables



1 us of simulation each

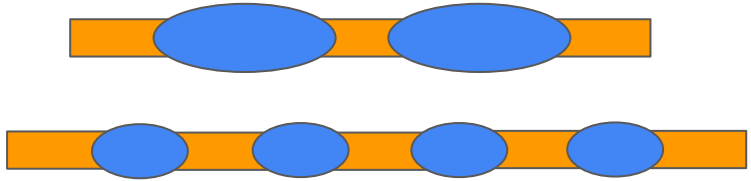
Analyzing: Radius of Gyration, Blob-Blob Contacts, Network Diagrams

Aim 2: Do polar blobs influence h-blobs interactions?

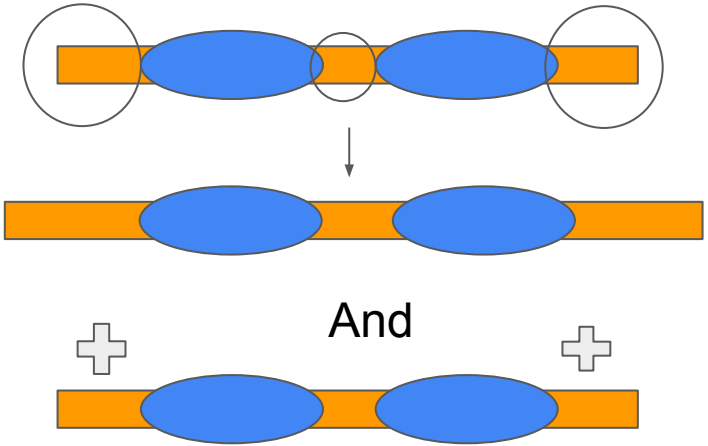
- Take 2 sequences from aim 1 that have the lowest radius of gyration from control sequence.
- Manipulate the p-blobs, increasing length of p-blobs or changing residues inside p-blobs
- Run 4 simulations of different peptide sequences each with a control sequence
- Simulation time of 1 us each
- Measure radius of gyration, and blob - blob contacts through every frame of simulation
- Create network diagrams off blob - blob contact data

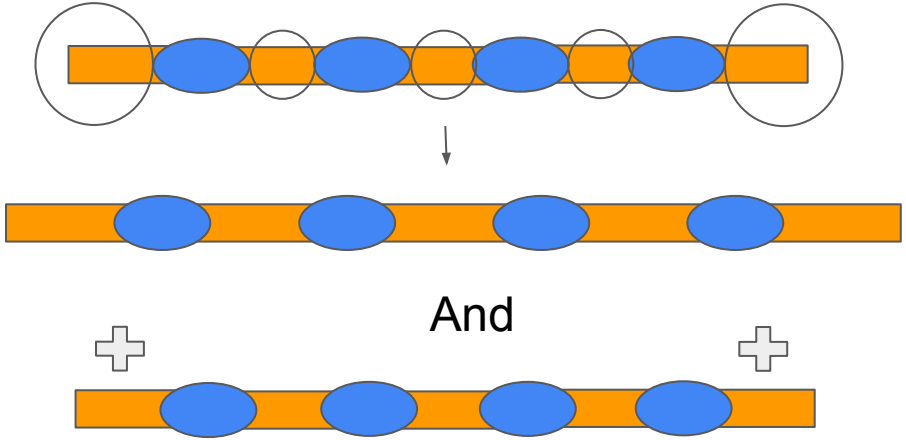
Aim 2: Breaking tertiary interactions with p-blobs

Most Compact Sequences

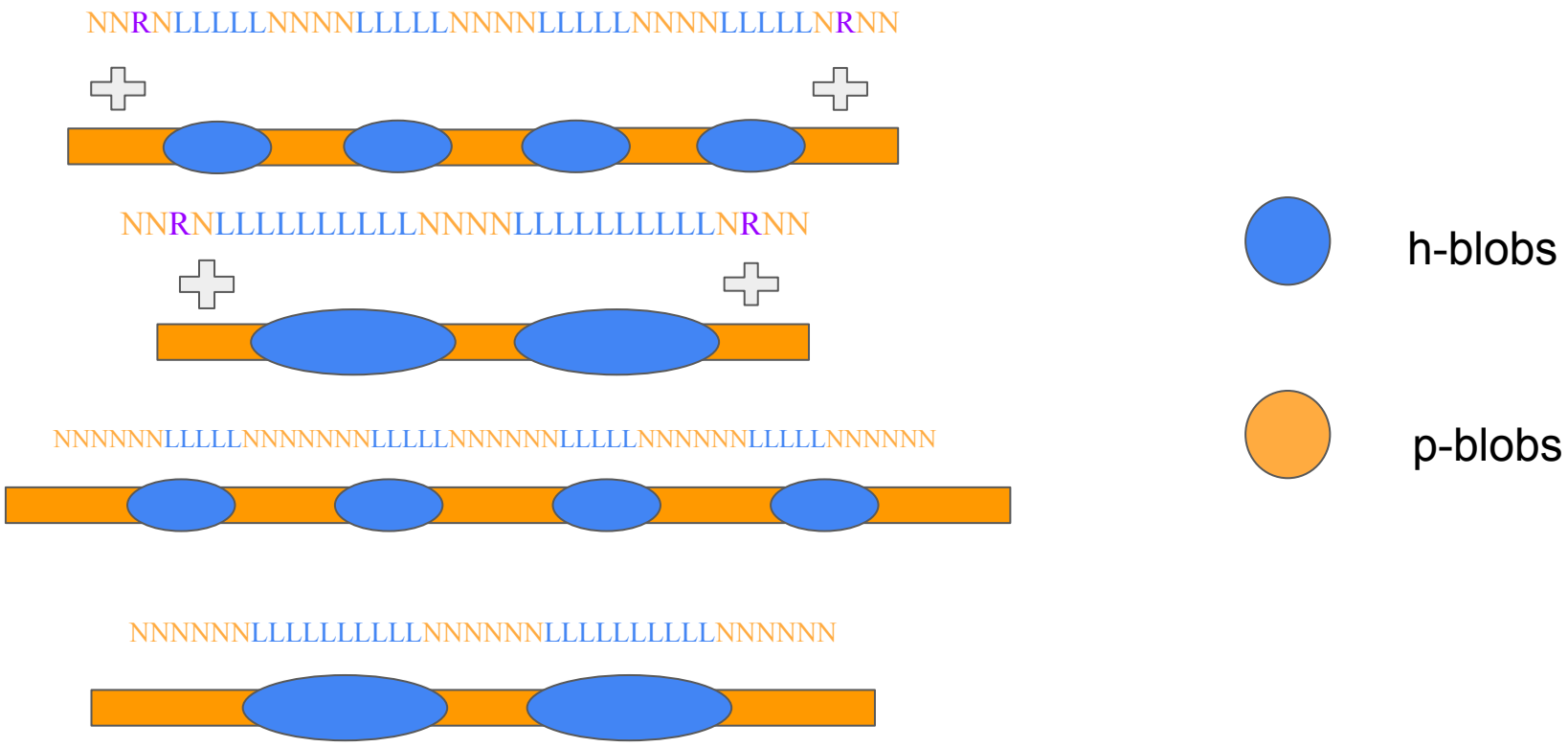


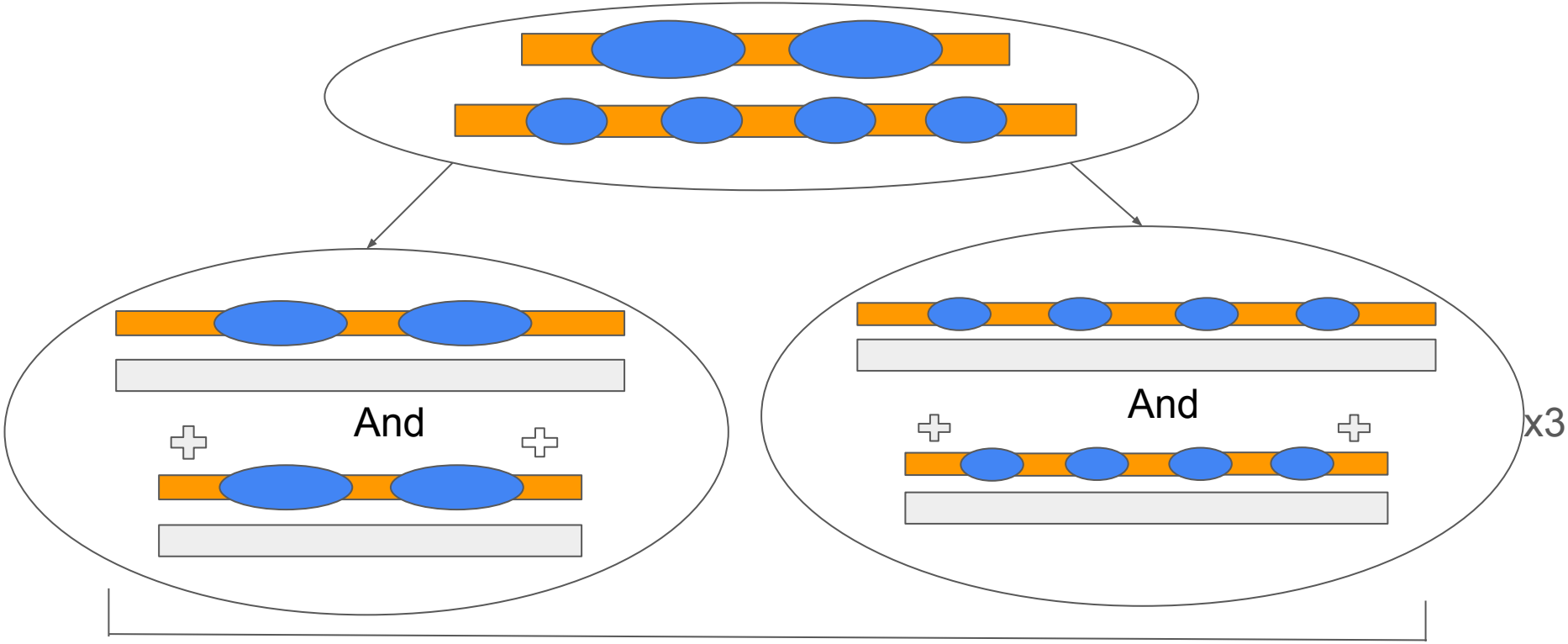
Manipulate their p-blobs





Most change from aim 1 to least changed





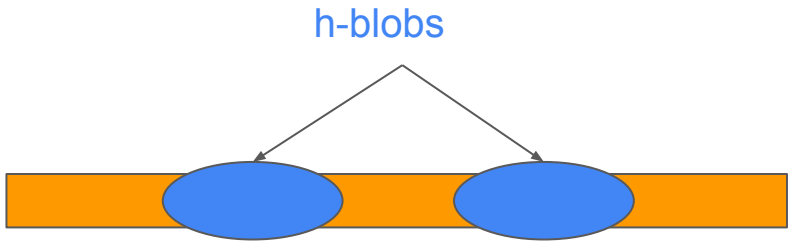
1 us of simulation each

Analyzing: Radius of Gyration, Blob-Blob Contacts, Network Diagrams

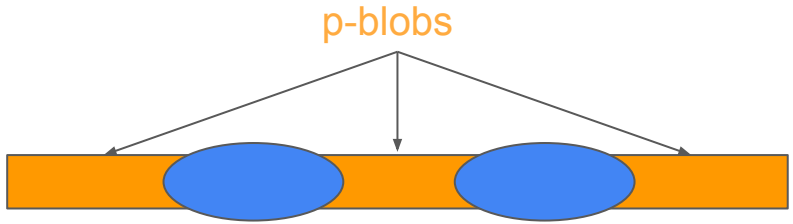
Summary

Main Question: How do h-blobs influence tertiary interactions in peptides?

Aim 1 main question: How do hydrophobic properties such as length and residue composition affect tertiary interactions in peptides?



Aim 2 main question: Do polar blobs influence h-blobs interactions?



Acknowledgements

Brannigan Lab

- Grace Brannigan
- Ezry Santiago
- Connor Pitman
- Jesse Sandberg
- Jahmal Ennis
- Regina Salzer
- Lindsey Riggs
- Alejandro Dagnino

Ruchi Lohia

Questions?