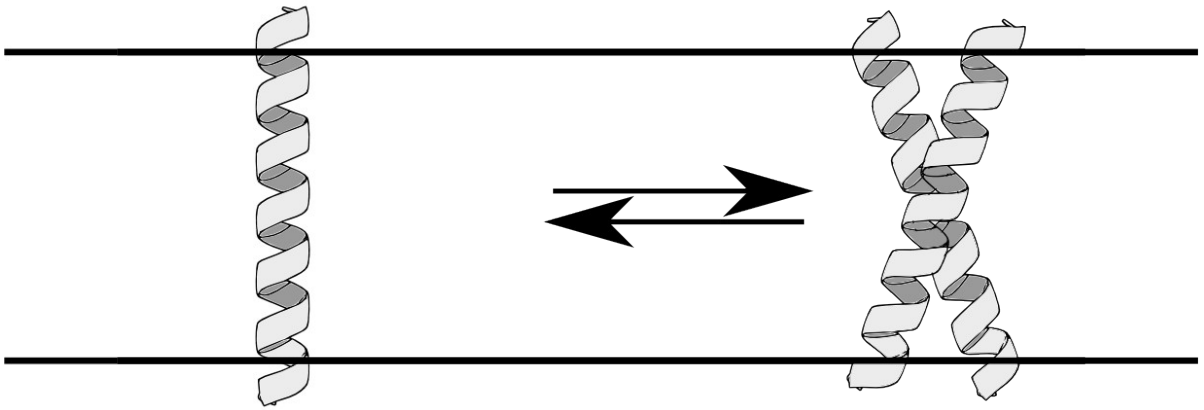
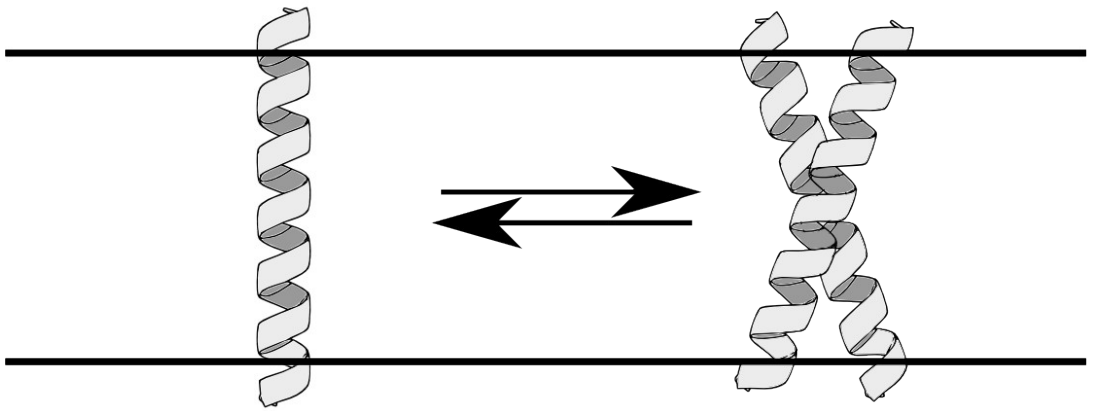
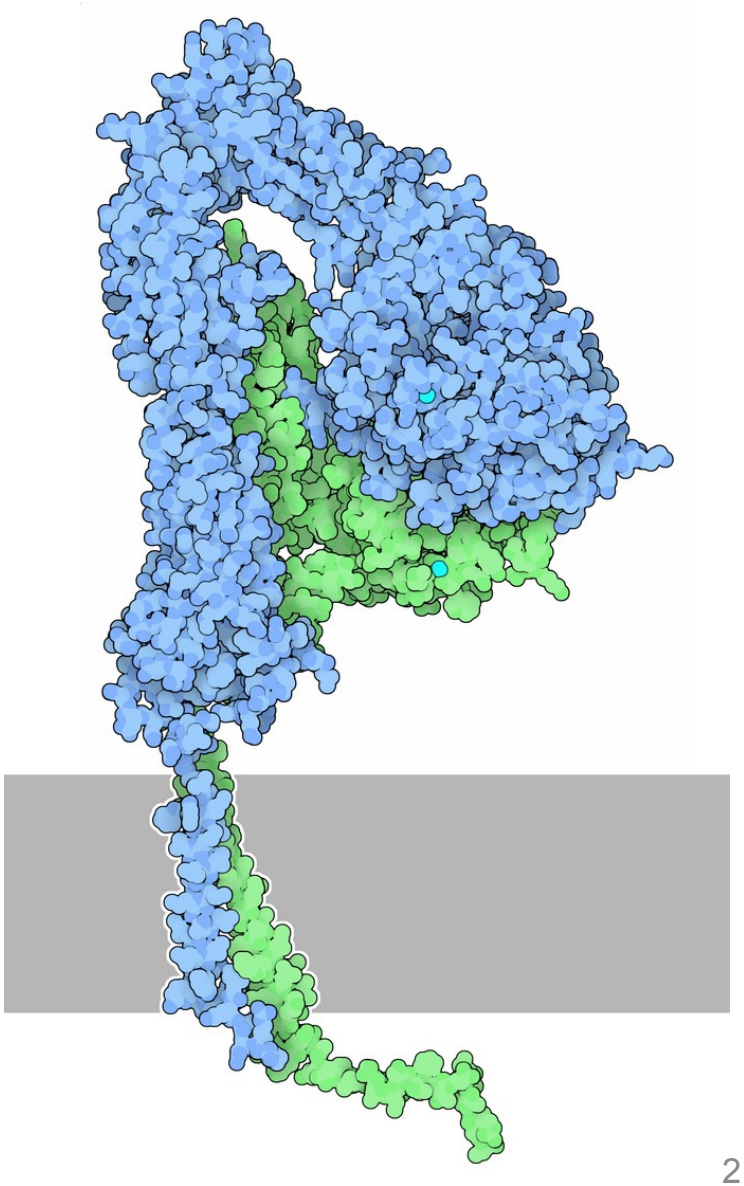
Structure prediction of transmembrane helix association



Sabareesh Subramaniam
Senes Lab
University of Wisconsin-Madison

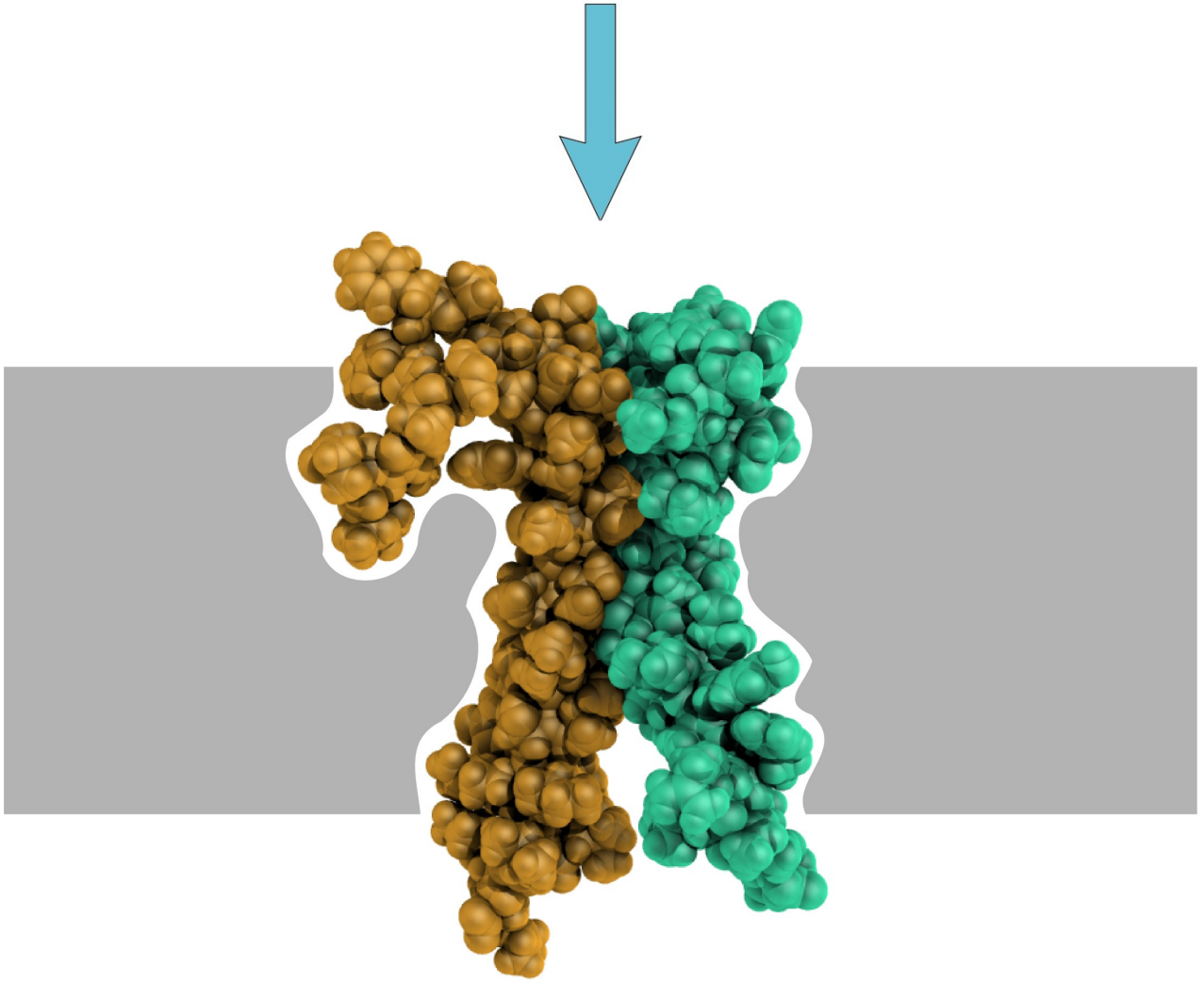
TM assocation is biologically important



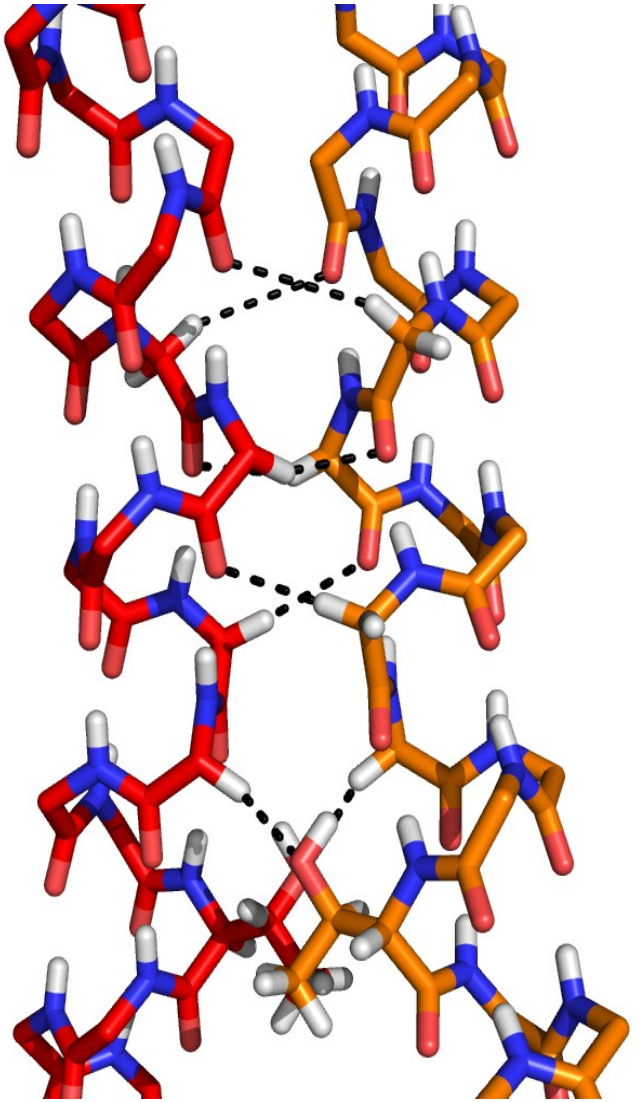


Can we predict dimer structure from sequence?

…EAEITLIIFGVMAGVIGTILLISYGIRRL…

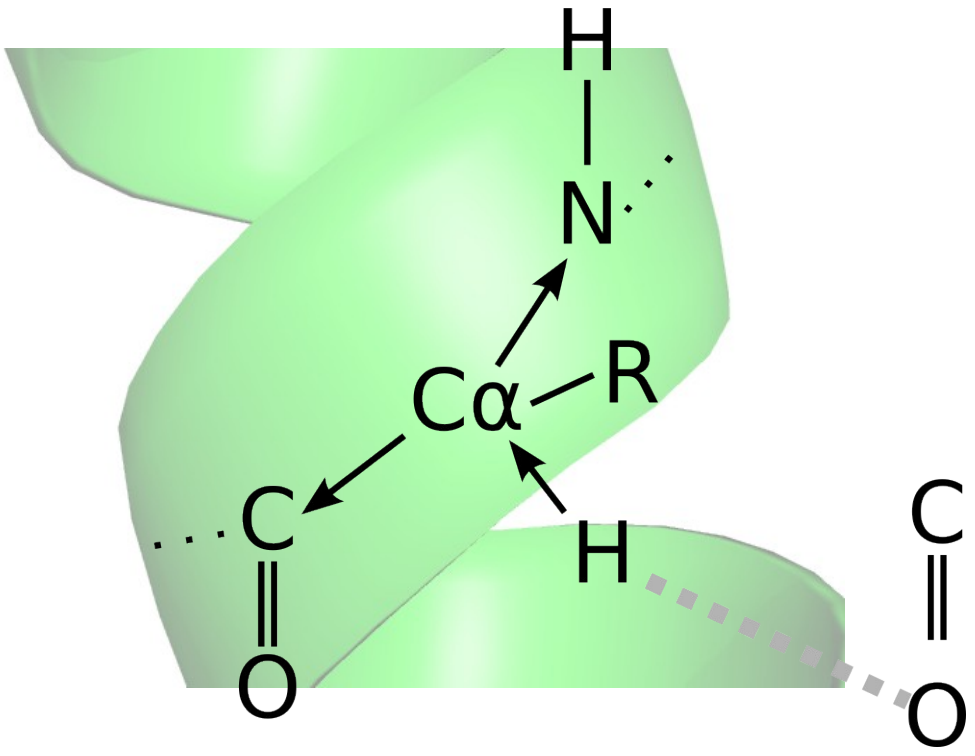


Cα-H·····O bonds mediate dimerization



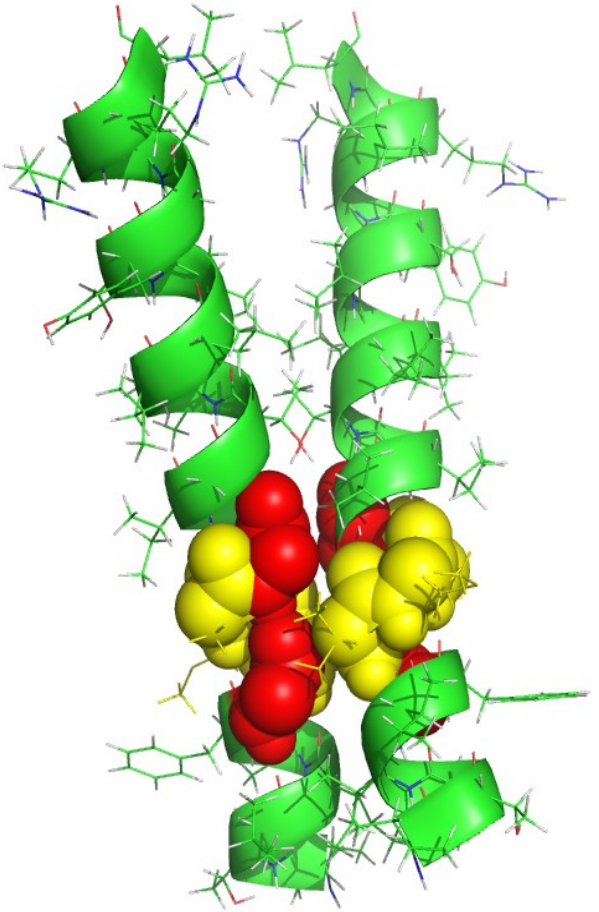
Senes A, Ubarretxena-Belandia I, Engelman DM. "The Calpha ---H...O hydrogen bond: a determinant of stability and specificity in transmembrane helix interactions." Proc Natl Acad Sci U S A. 2001 98(16), 9056-61

Cα-H·····O hydrogen bonds



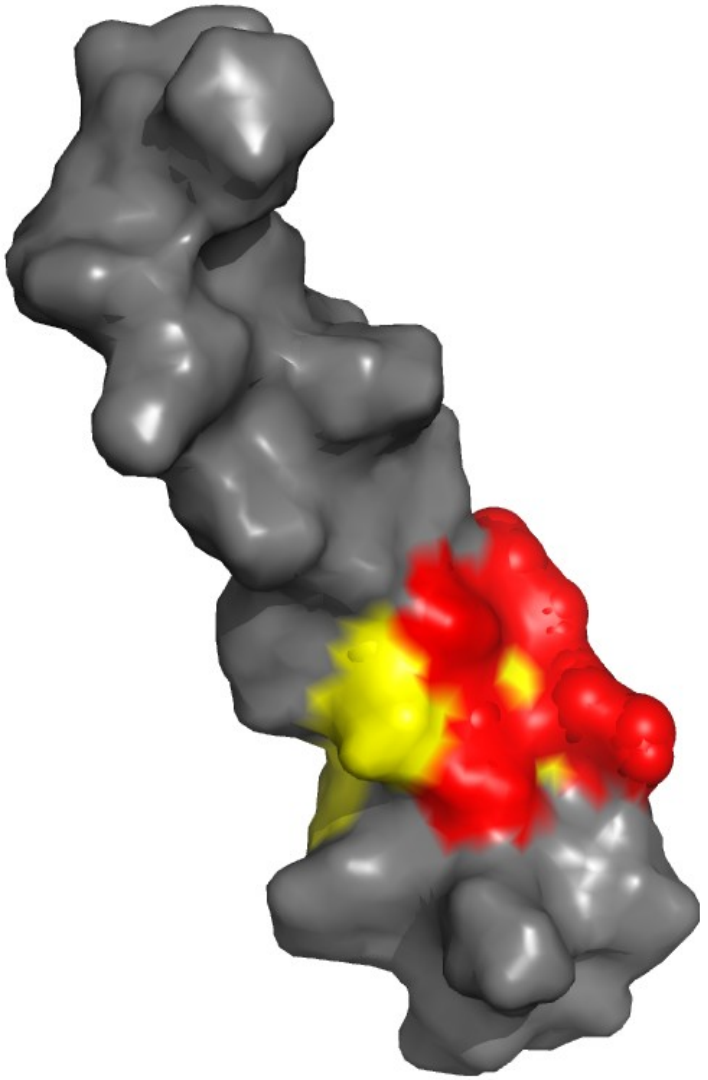
Not as strong as a canonical hydrogen bonds but still significant

GXXXG motif facilitates Cα-H·····O

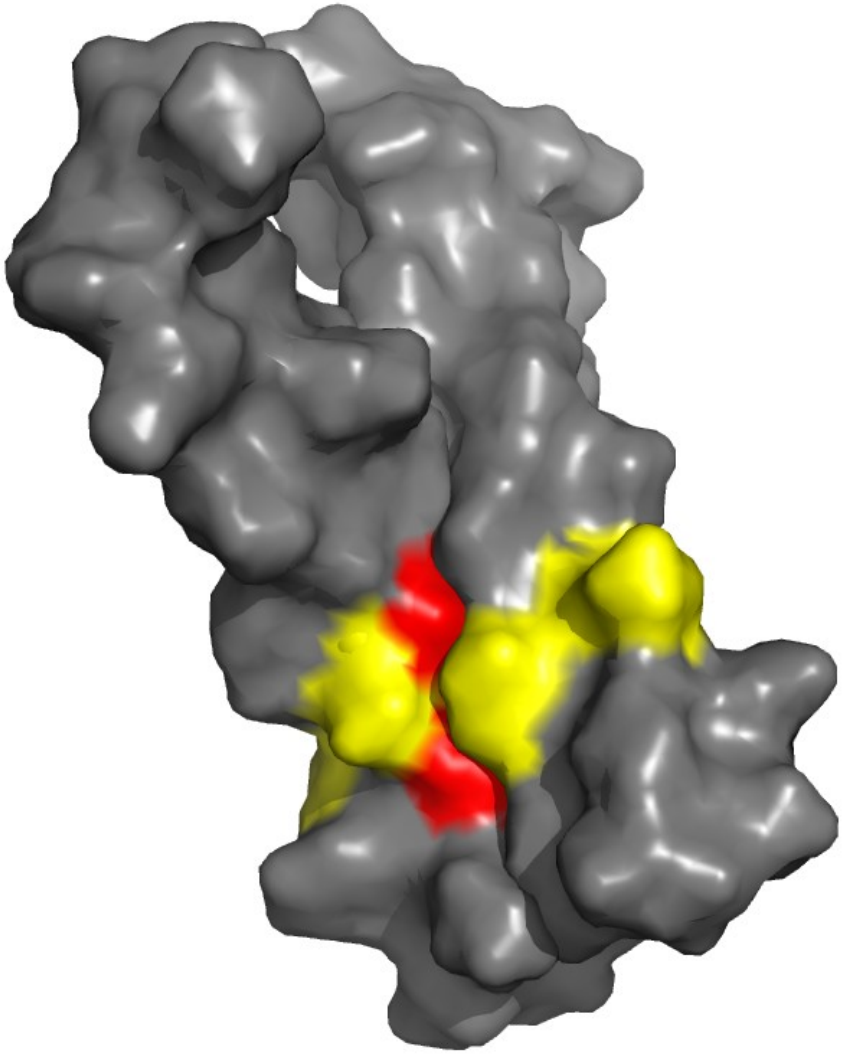


ITLIIFGVMAGVIGTILLISYGIRRL

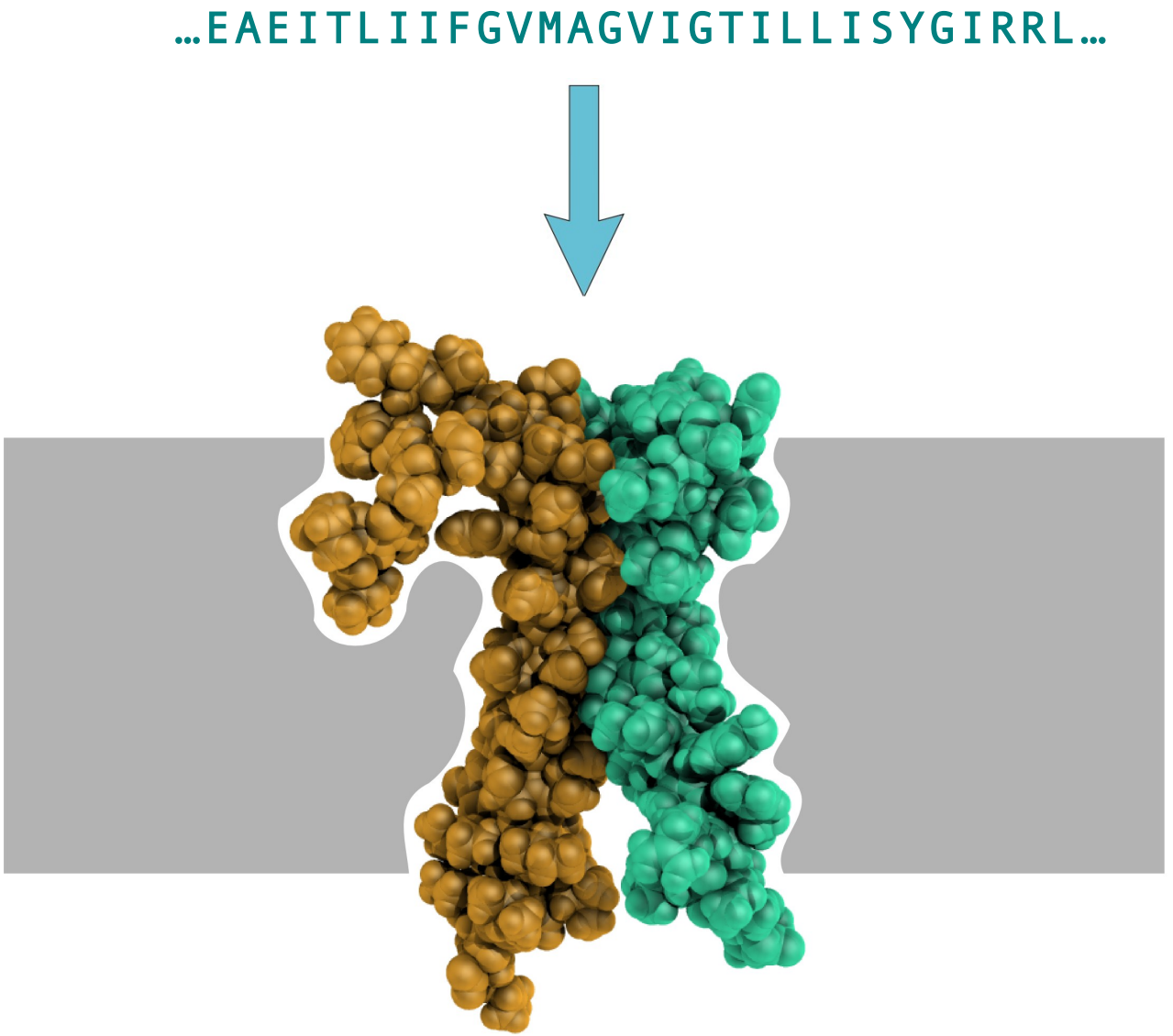
GXXXG motif facilitates Cα-H·····O



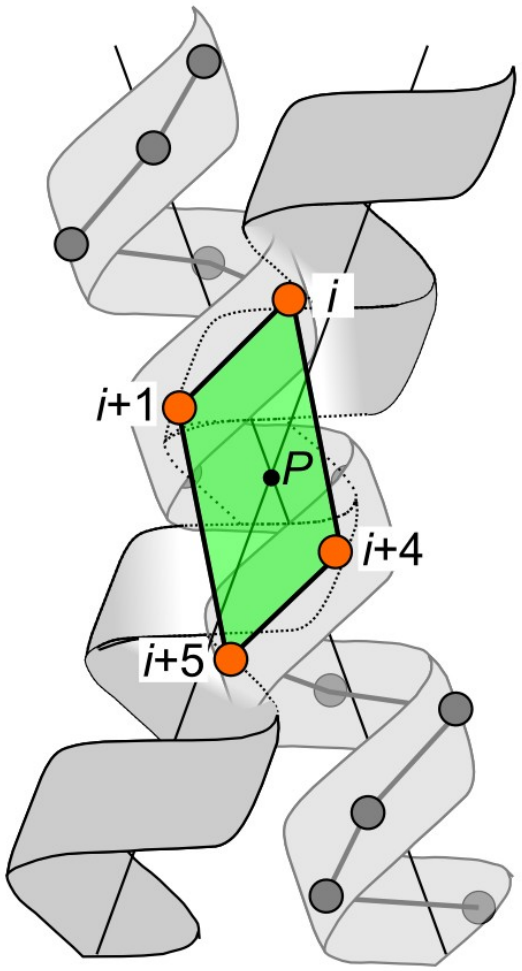
GXXXG motif facilitates Cα-H·····O

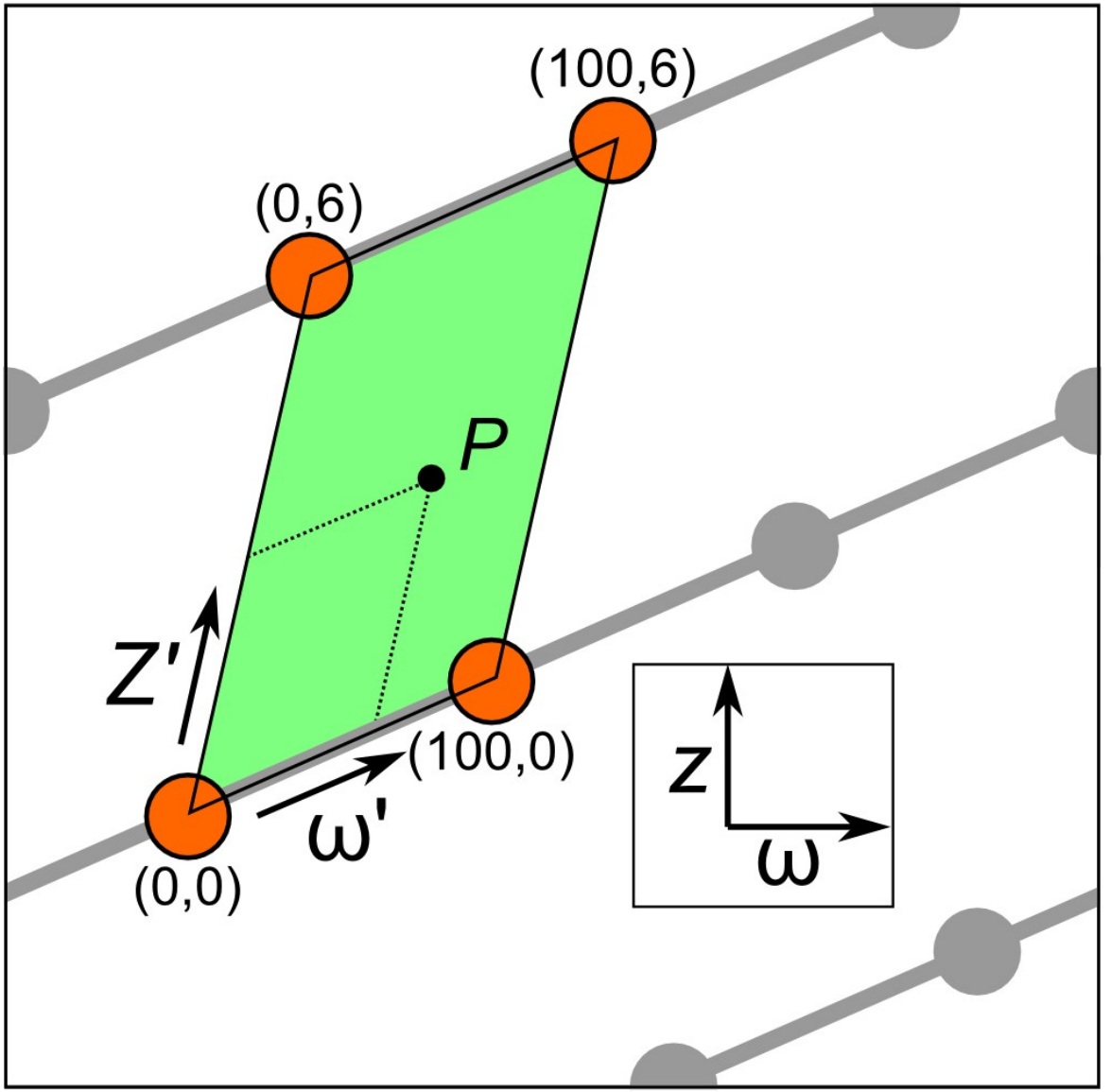


What is the Cα-H·····O mediated dimer structure?

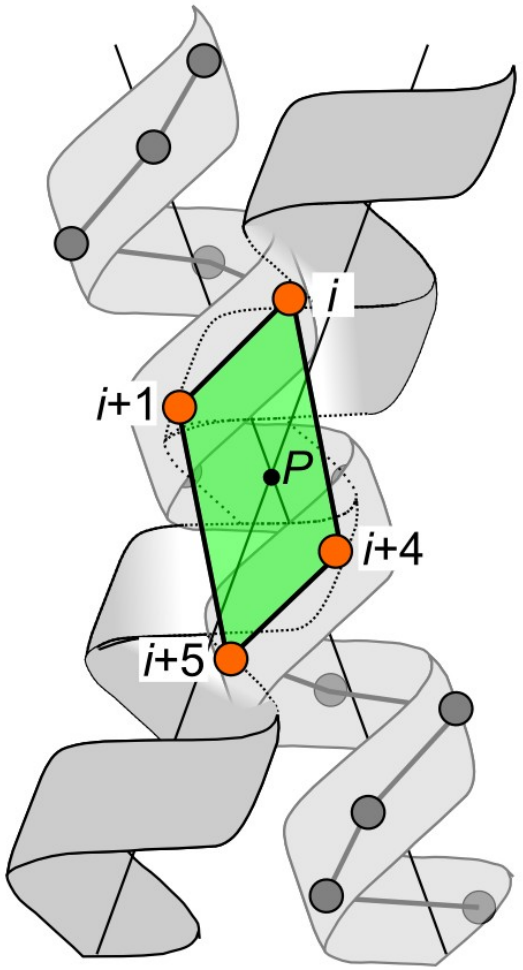


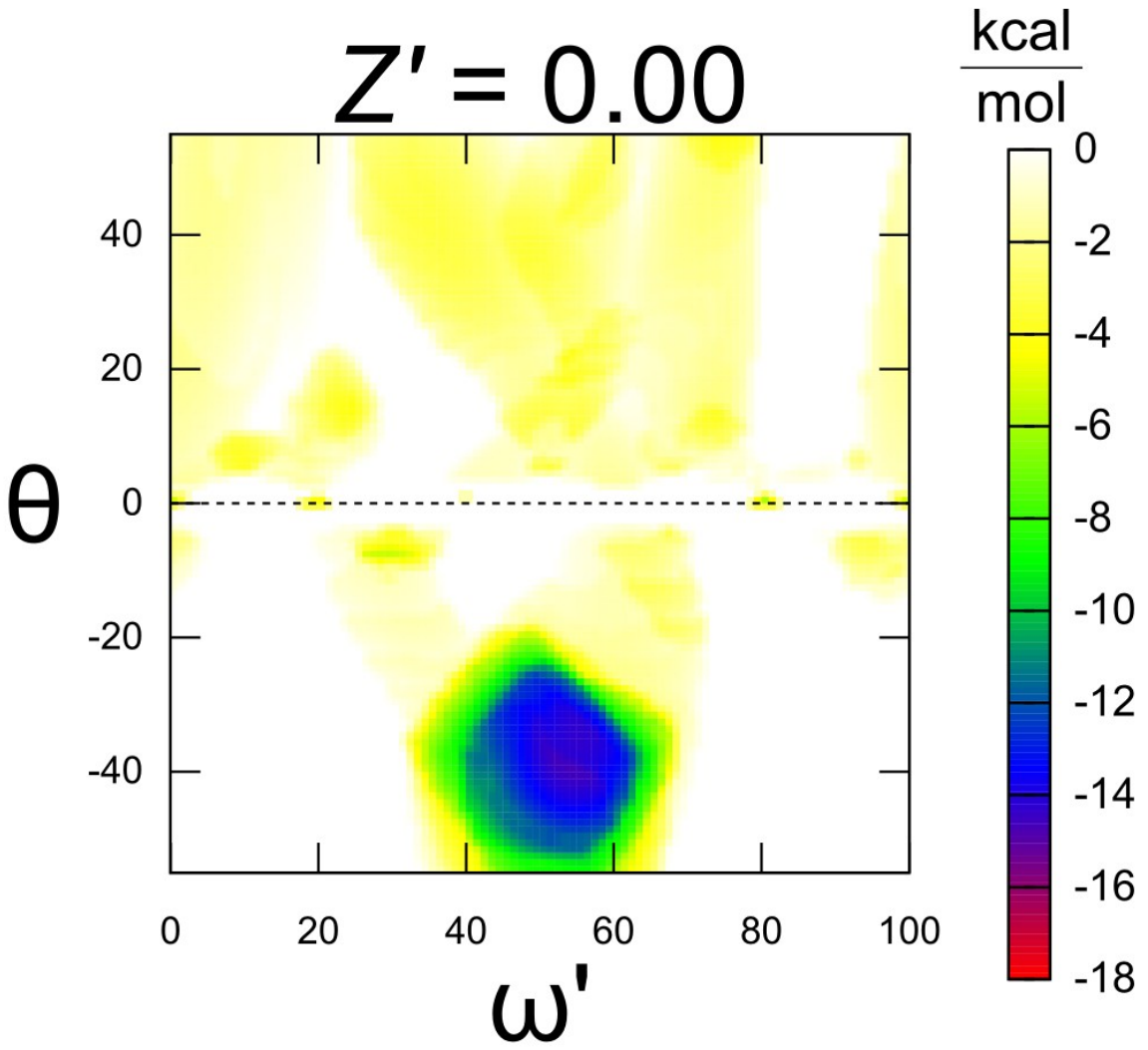
The homodimer space



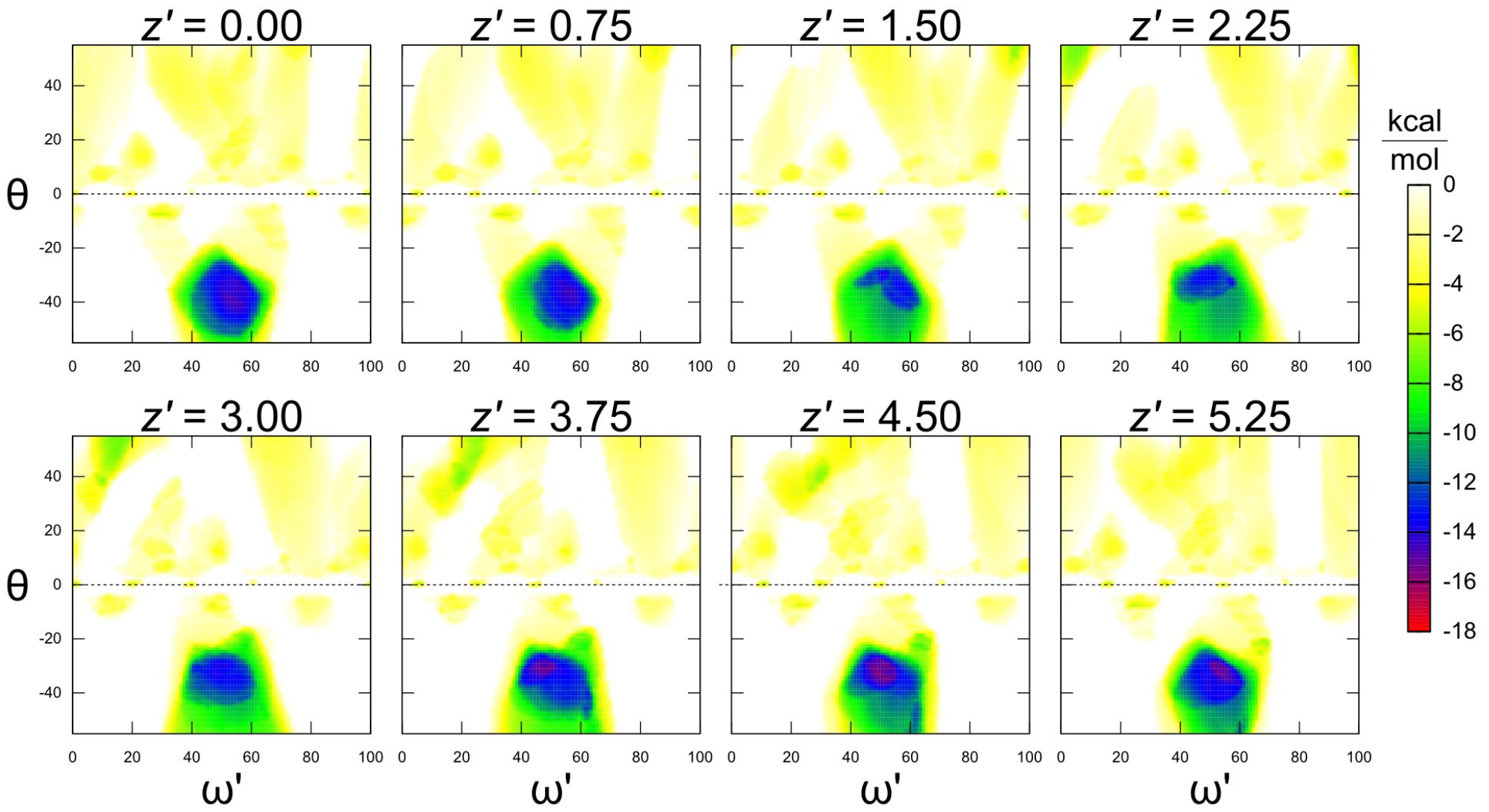


Bias in the dimer space

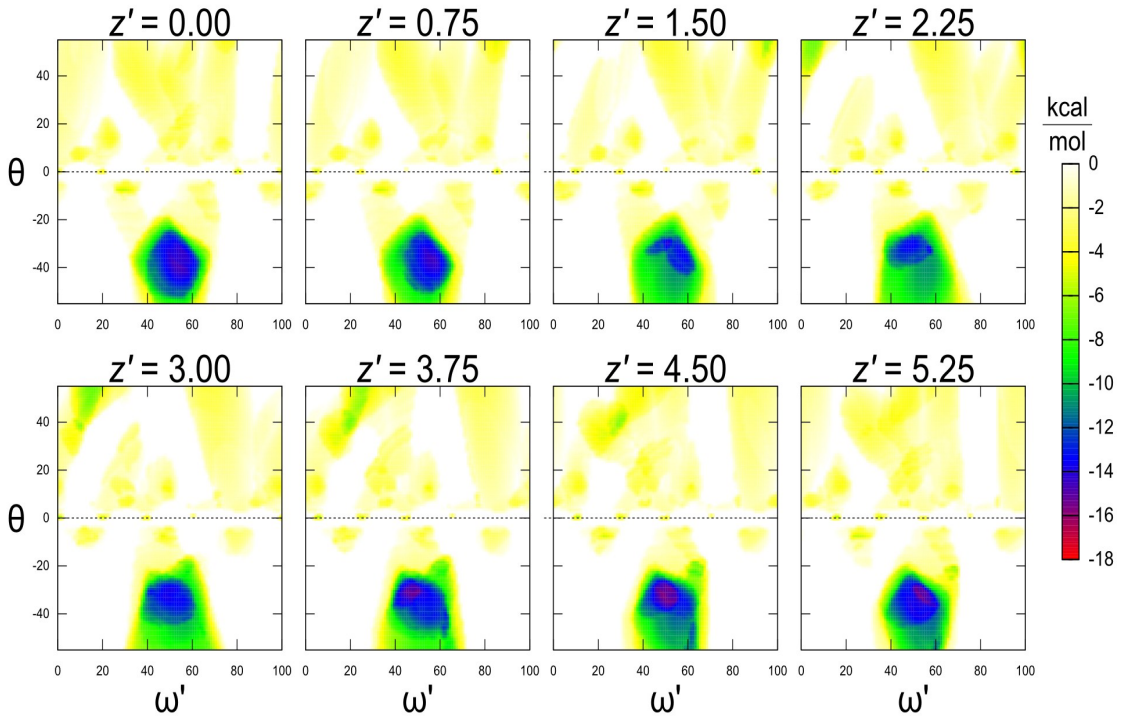


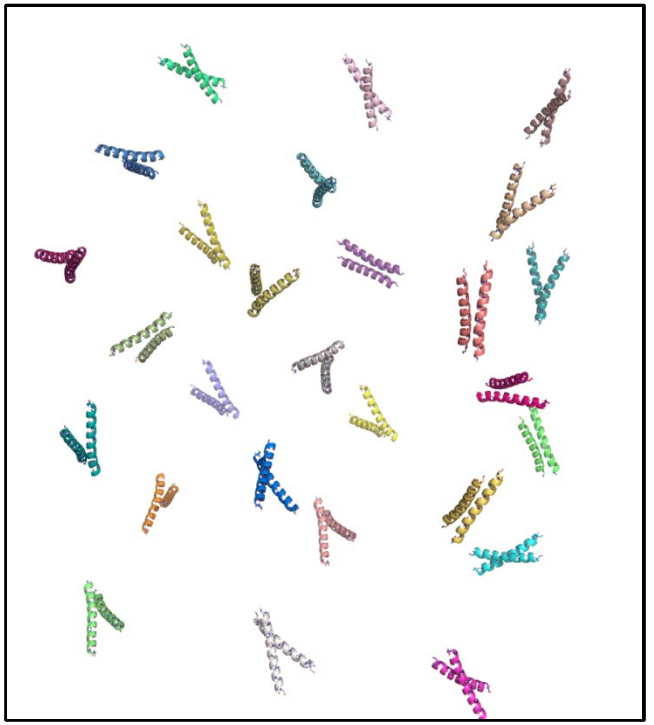


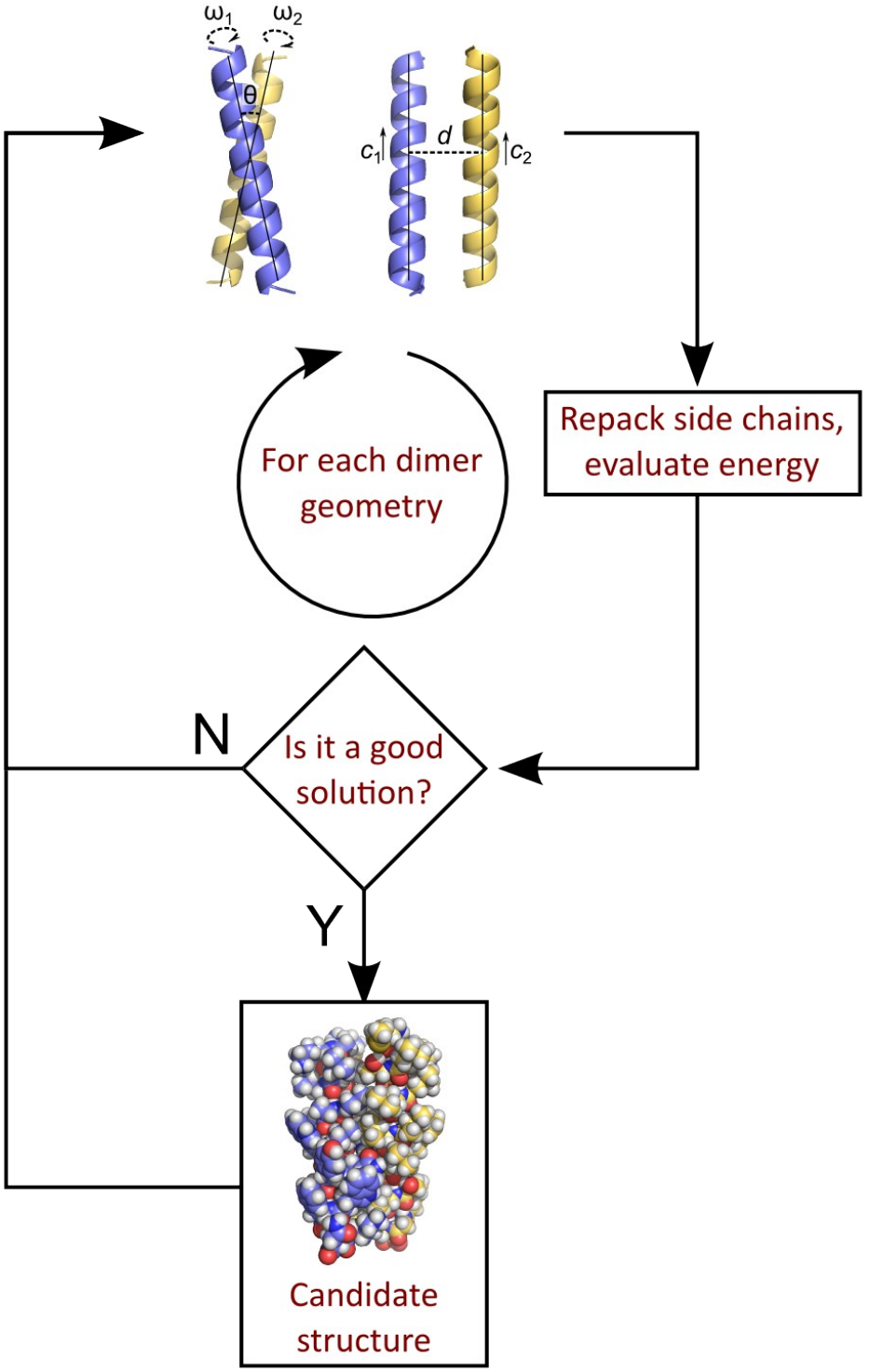
Bias in the dimer space



Precompute favorable dimer geometries






ω1
ω2
θ
c1
d
c2
For each dimer geometry
Repack side chains, evaluate energy
N
Is it a good solution?
Y
Candidate structure

Glycophorin-A

UniProt data

UniProt name: GLPA_HUMAN (P02724)

Gene name: GYPA; Synonyms=GPA

Organism: Homo sapiens (Human).

Function: Glycophorin A is the major intrinsic membrane protein of the erythrocyte. The N-terminal glycosylated segment, which lies outside the erythrocyte membrane, has MN blood group receptors. Appears to be important for the function of SLC4A1 and is required for high activity of SLC4A1. May be involved in translocation of SLC4A1 to the plasma membrane. Is a receptor for influenza virus. Is a receptor for Plasmodium falciparum erythrocyte-binding antigen 175 (EBA-175); binding of EBA-175 is dependent on sialic acid residues of the O-linked glycans. Appears to be a receptor for Hepatitis A virus (HAV).

Subcellular location: Cell membrane; Single-pass type I membrane protein. Note=Appears to be colocalized with SLC4A1.

Transmembrane domains:

1. 92-114, 23 AA, Helical.

Sequence: 150 AA

```
        10         20         30         40         50         60
MYGKIIFVLL LSEIVSISAS STTGVAMHTS TSSSVTKSYI SSQTNDTHKR DTYAATPRAH
        70         80         90        100        110        120
EVSEISVRTV YPPEEETGER VQLAHHFSEP EITLIIFGVM AGVIGTILLI SYGIRRLIKK
       130        140
SPSDVKPLPS PDTDVPLSSV EIENPETSDQ
```

CATM prediction

The sequence of GLPA_HUMAN returned 5 potential models

The positions at the dimer's interface are highlighted in yellow. The position at the crossing point is highlighted in orange.

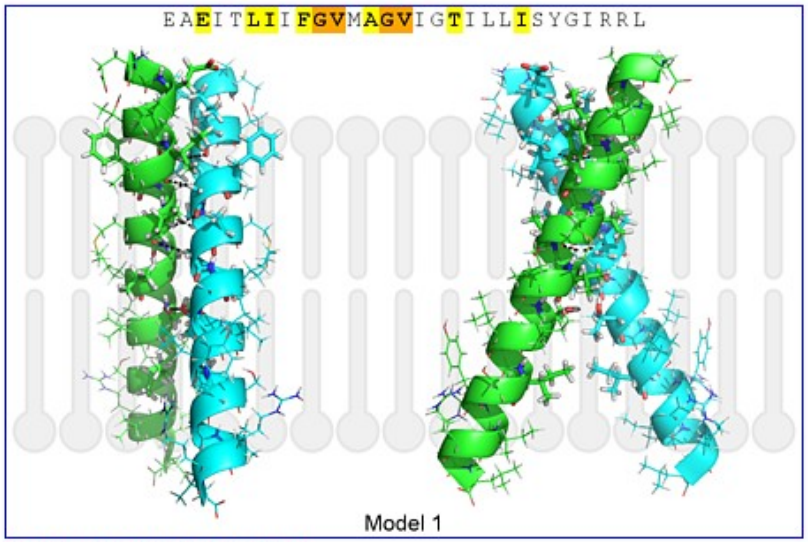
#	Sequence and interface	d	θ	Z'	ω'	Score
Model 1	EAEITLIIFGVMAGVIGTILLISYGIRRL	6.3Å	-49.1°	5.9	55.9	-46.3
Model 2	EAEITLIIFGVMAGVIGTILLISYGIRRL	7.5Å	+26.0°	1.7	25.9	-32.0
Model 3	EAEITLIIFGVMAGVIGTILLISYGIRRL	6.7Å	+54.0°	0.4	20.2	-30.2
Model 4	EAEITLIIFGVMAGVIGTILLISYGIRRL	7.4Å	-45.8°	3.0	41.3	-24.6
Model 5	EAEITLIIFGVMAGVIGTILLISYGIRRL	7.1Å	-54.0°	4.2	44.7	-23.1
Prolins*	EPEITLIIFGVMAGVIGTILLISYGIRRL					

*** Note:** this sequence contains Pro residues. CATM substituted them with Ala during the modeling. The positions are marked.

Click on the model number to visualize its details below.

MODEL 1 DETAILS

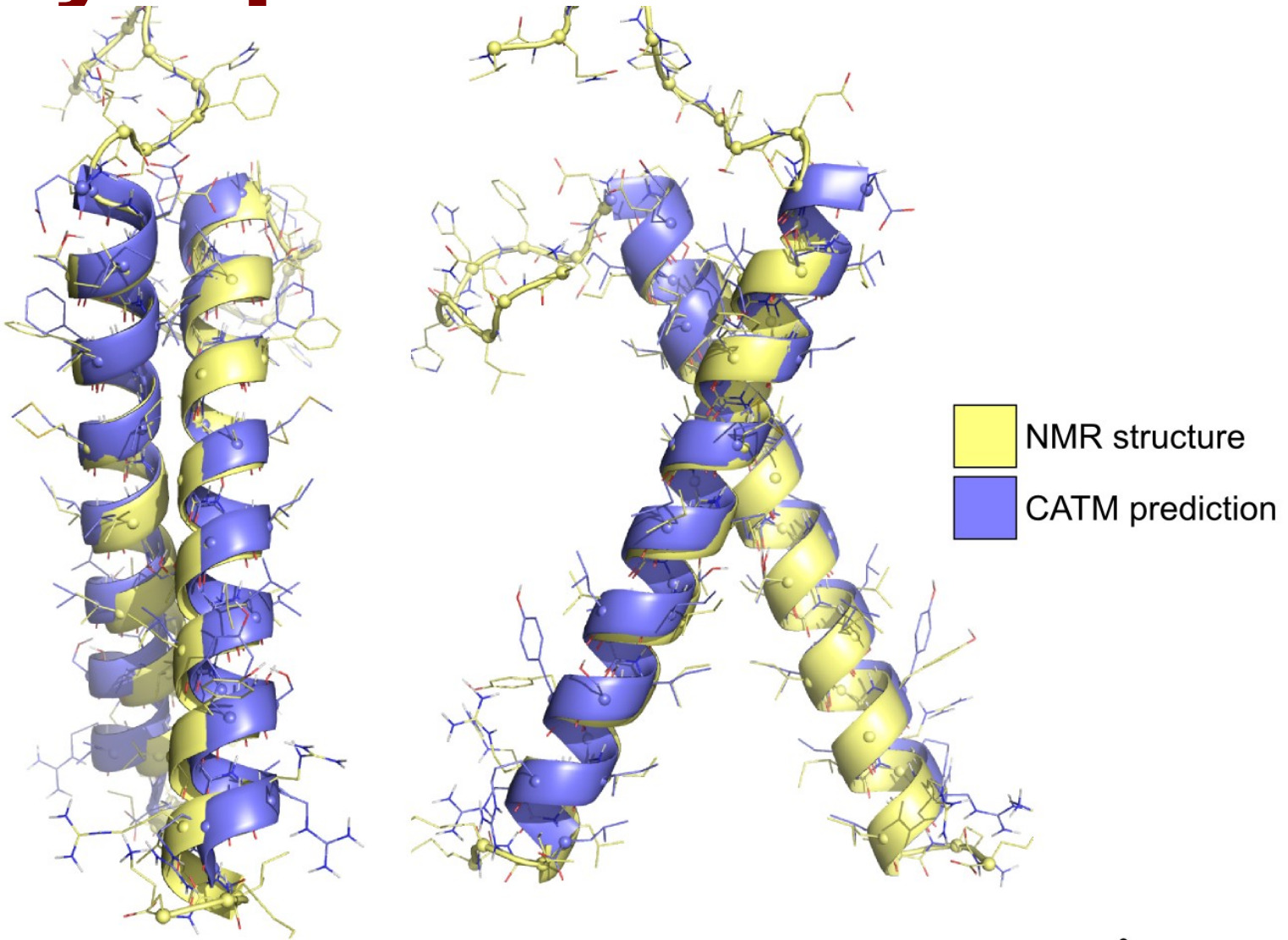
Precited structure:



Click on the image to enlarge

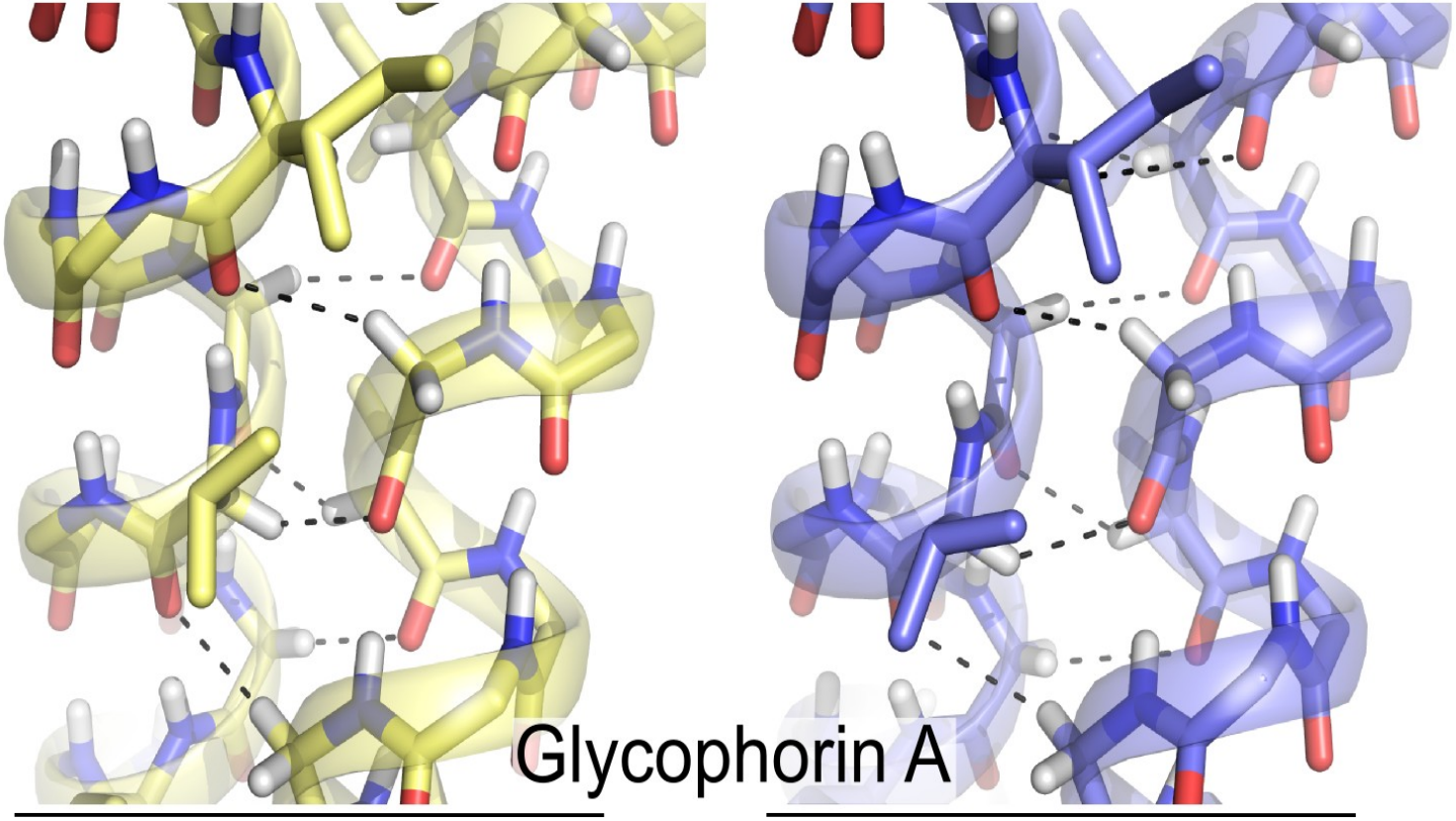
Download model 1 structure: PDB, PyMOL session

GlycophorinA backbone



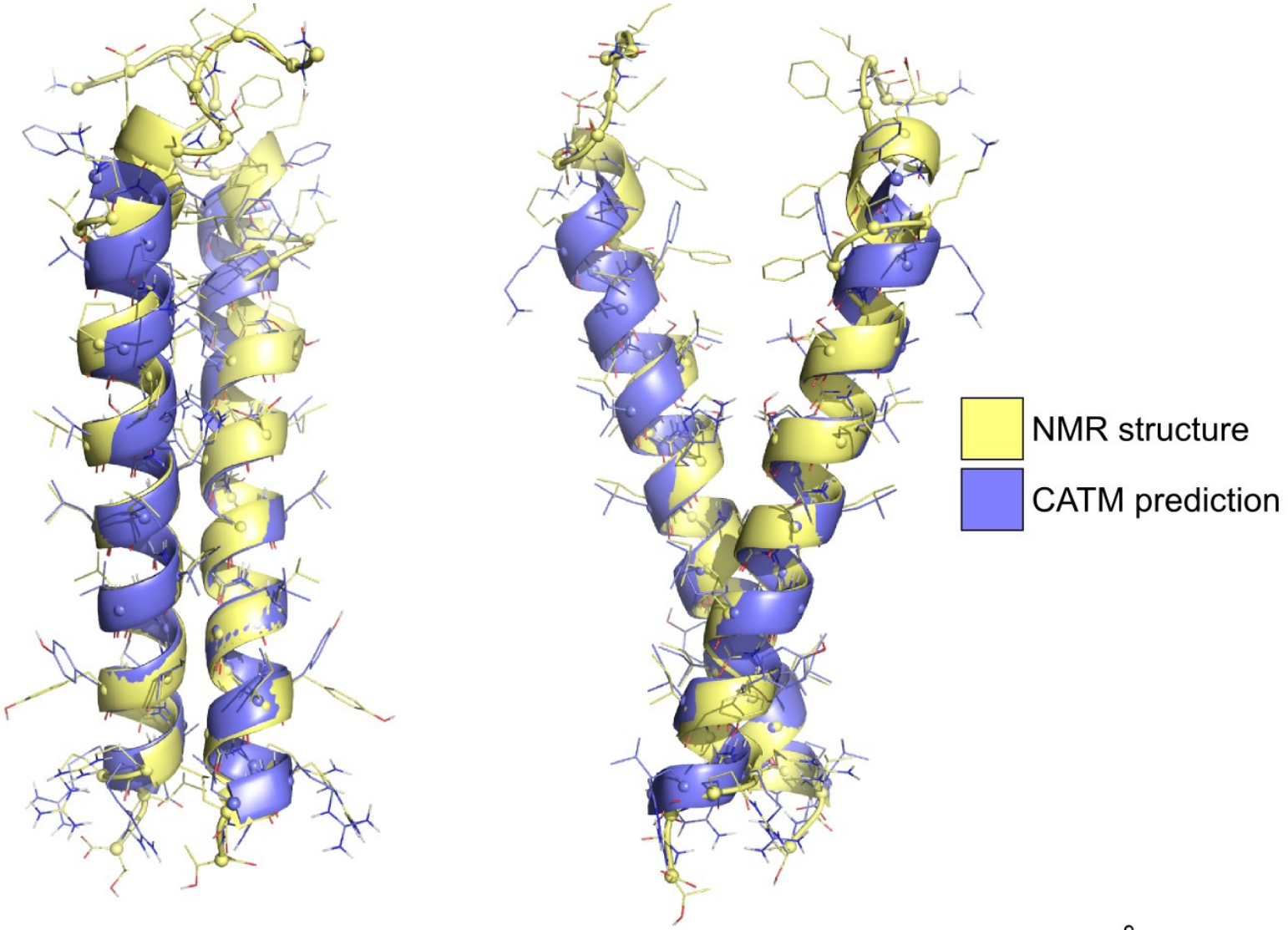
Protein	PDB	Residues	Cα RMSD (Å)
GpA	1afo	75- 87	1.11 ± 0.21

GlycophorinA hydrogen bonds



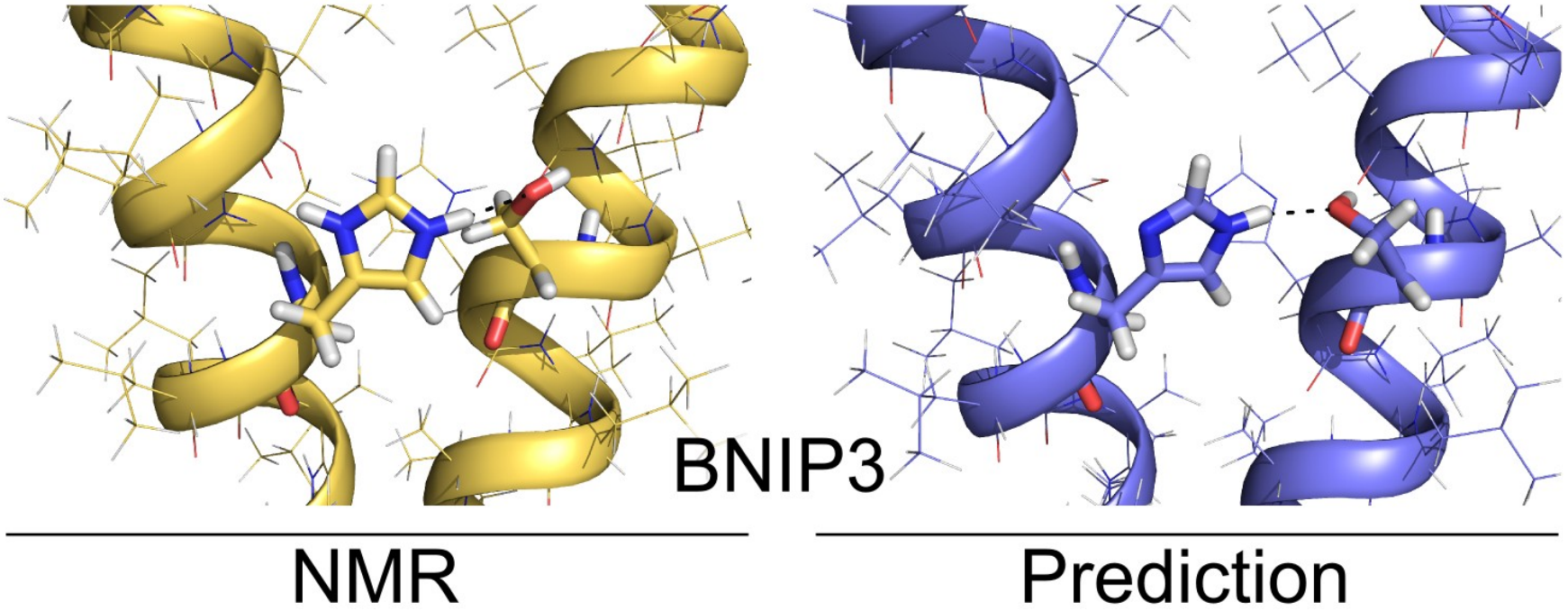
NMR **CATM**

BNIP3 backbone



Protein	PDB	Residues	Cα RMSD (Å)
BNIP3	1ka1	172-184	0.51 ± 0.10

BNIP3 hydrogen bonds

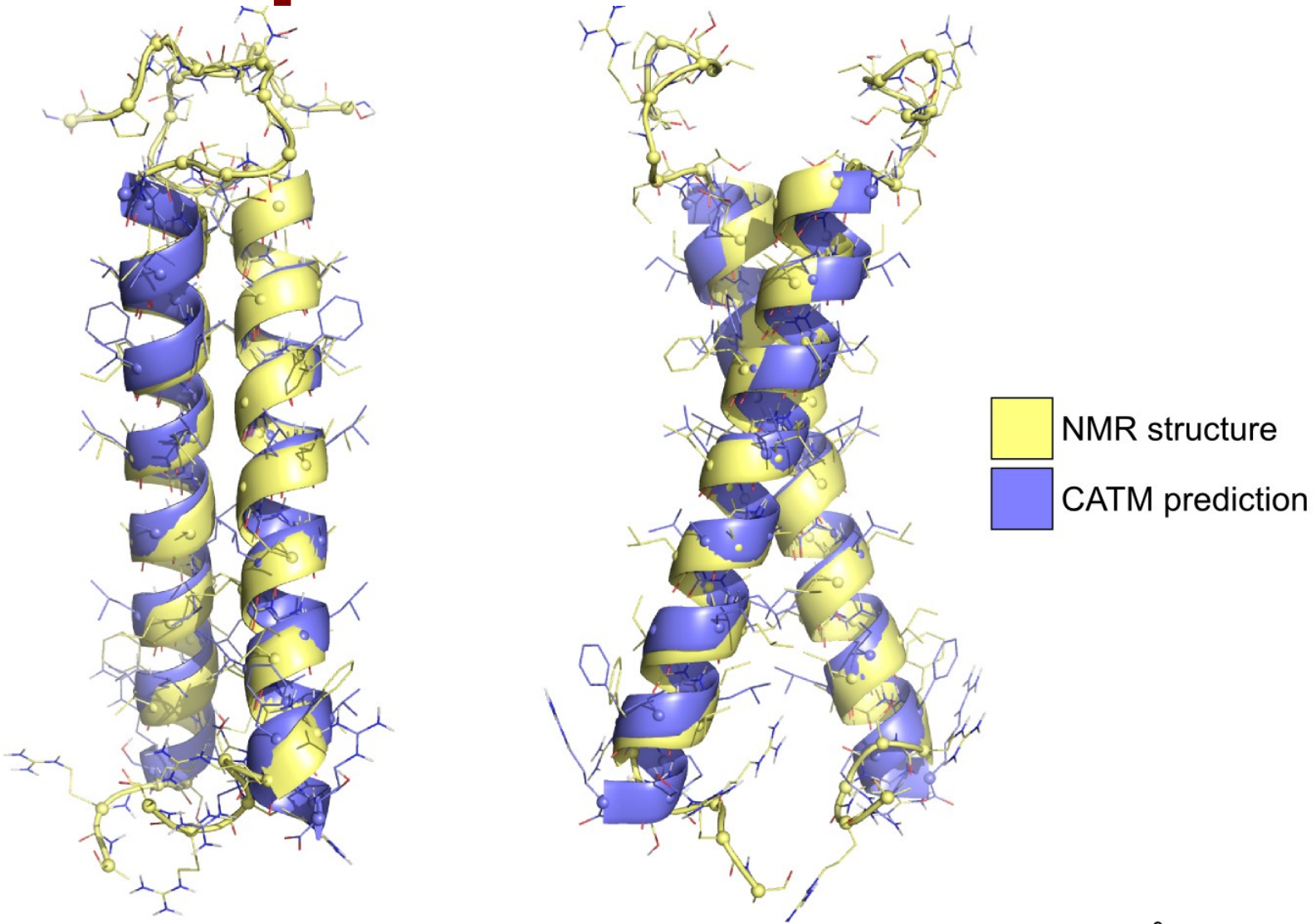


EphA1 backbone

..

GGEIV**AVIFG**LLLGAAGR

..



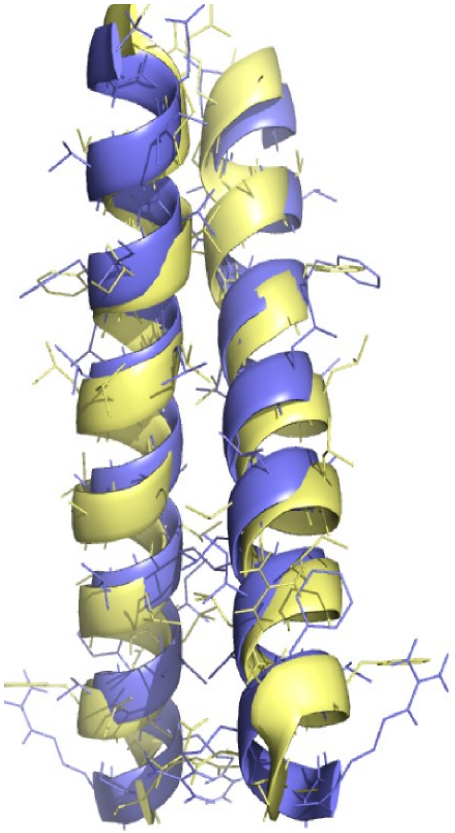
Protein	PDB	Residues	Cα RMSD (Å)
EphA1	2k1k	547-566	1.26 ± 0.01

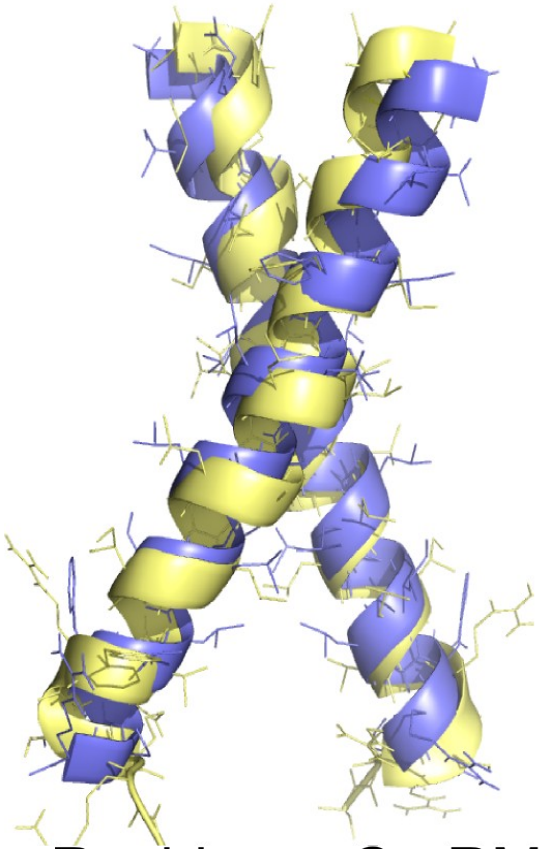
EphA1 backbone

..

GGEIV**AVIF**GLLLGAAGR

..



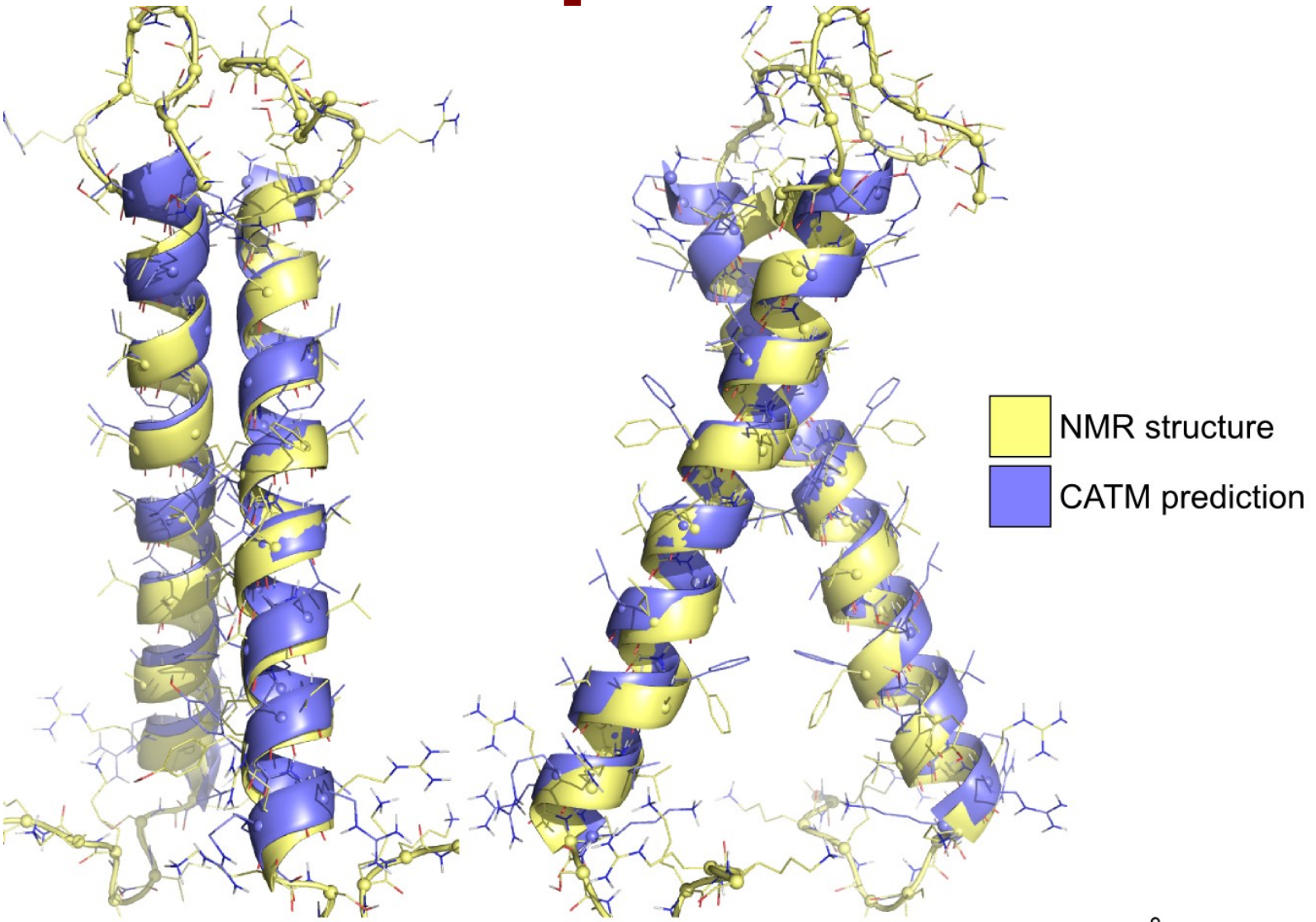


NMR structure

CATM prediction

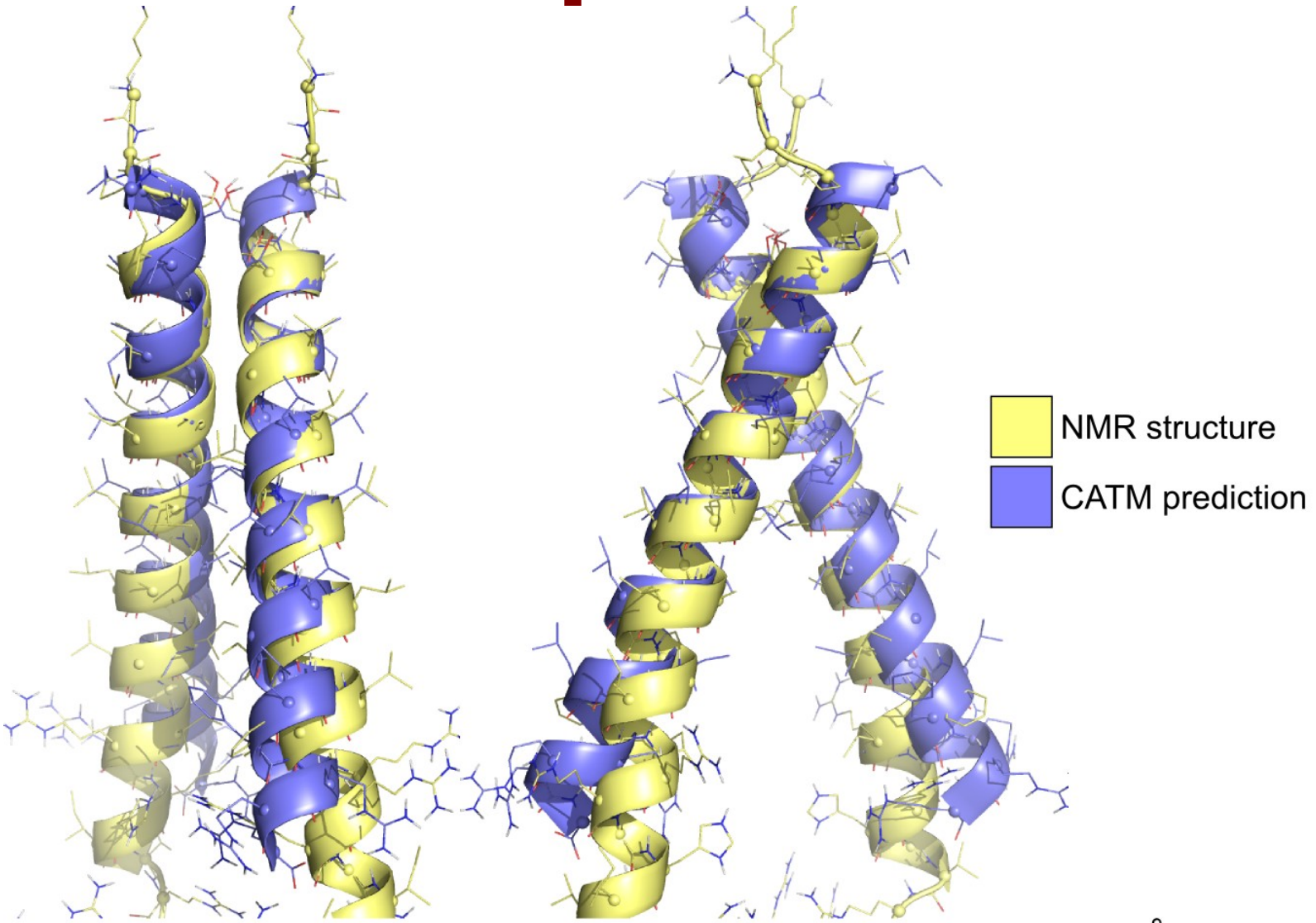
Protein	PDB	Residues	Cα RMSD (Å)
EphA1	2k1l	547-566	1.36 ± 0.02

ErbB4 prediction



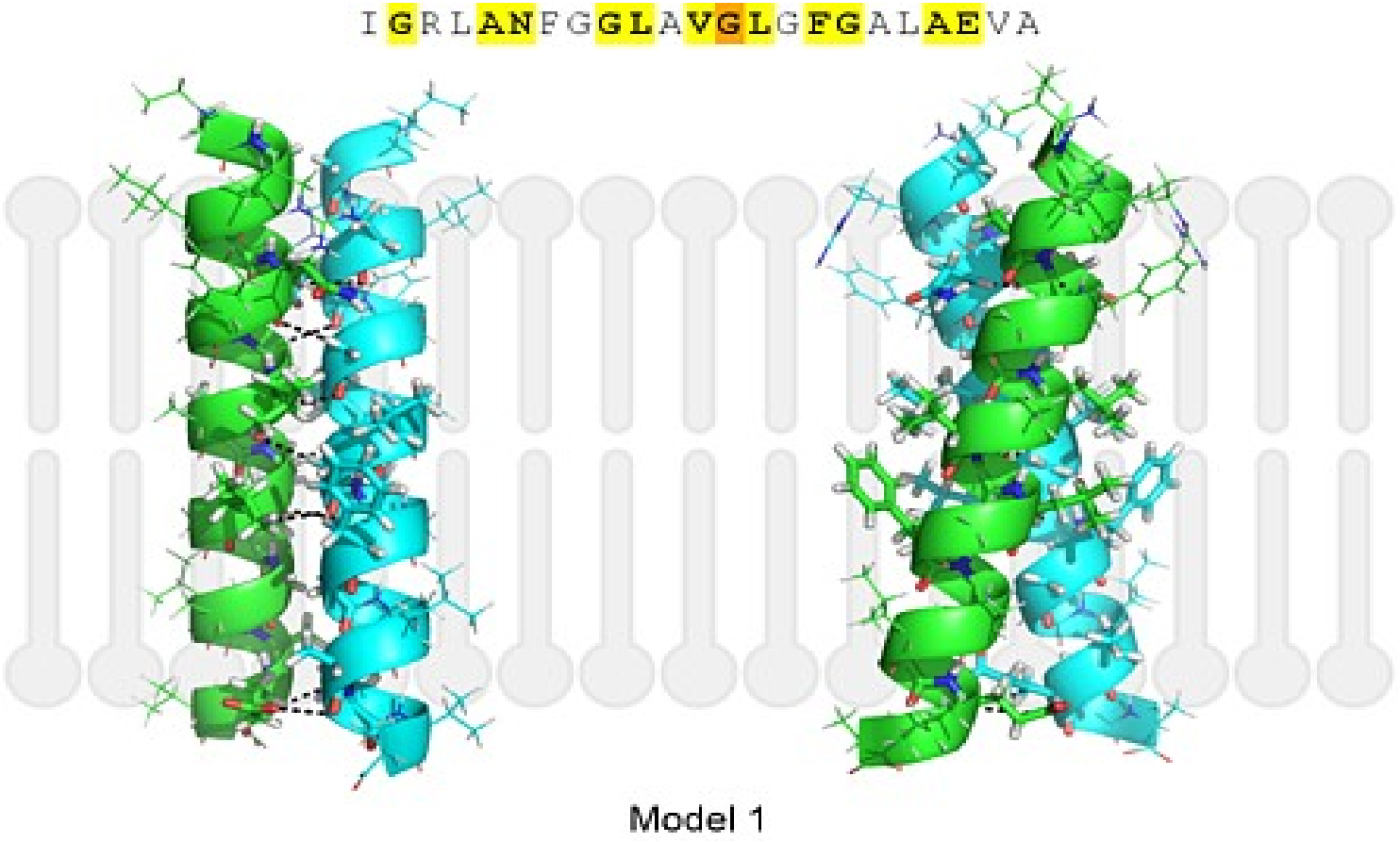
Protein	PDB	Residues	Cα RMSD (Å)
ErbB4	2l2t	652-664	0.82 ± 0.05

ErbB1 prediction



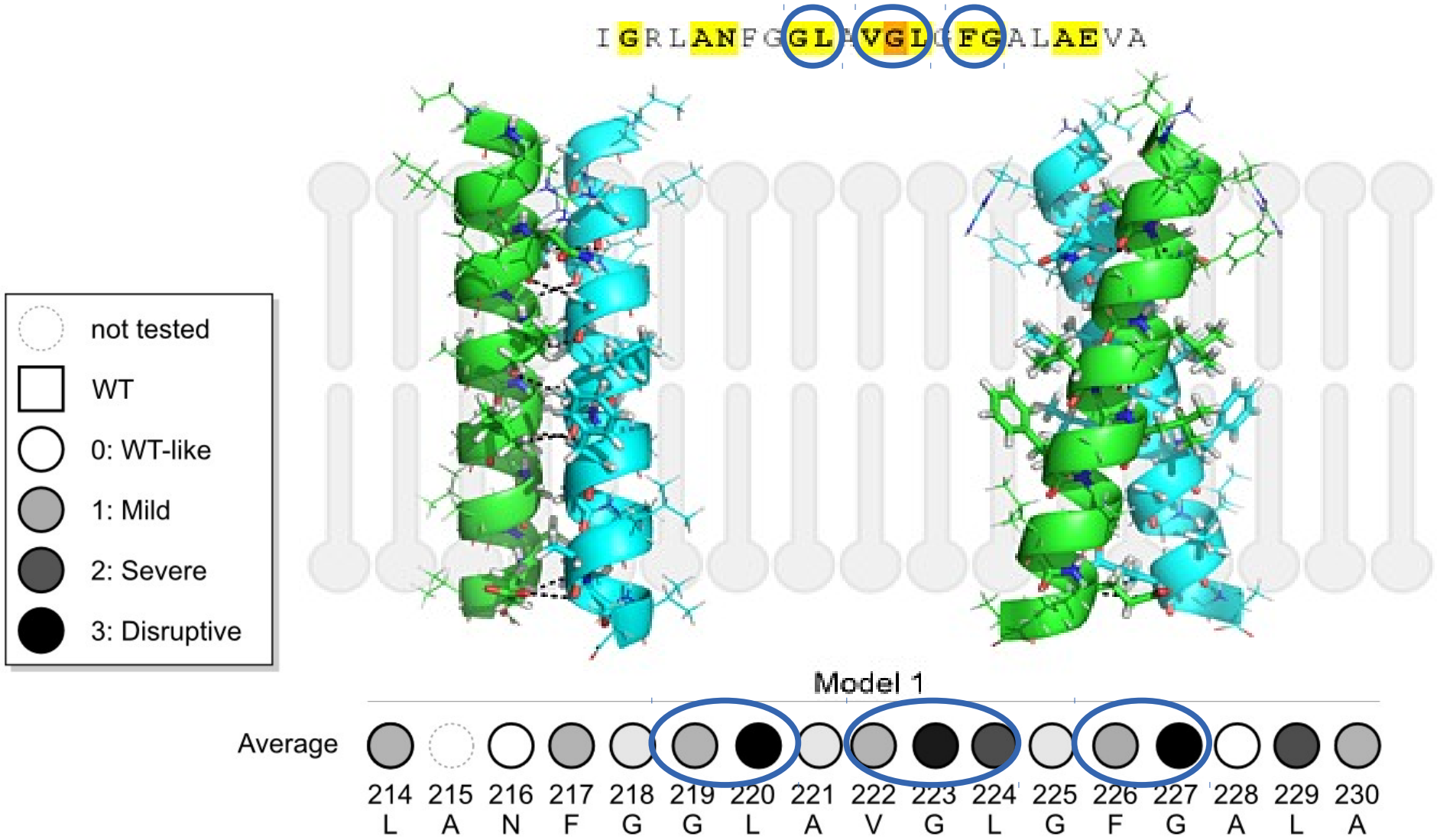
Protein	PDB	Residues	Cα RMSD (Å)
ErbB1	2m20	645-657	0.77 ± 0.05

Mitochondrial kinase



Pagliarini Lab, UW Madison

Mitochondrial kinase preliminary mutagenesis



Experiments by Pagliarini Lab and Ambalika Khadria

Conclusions

- Predict known dimers accurately
  - Backbone geometry/hydrogen bonds
  - Sidechain hydrogen bonds
- Predict alternative conformations
- Preliminary experiments indicate accurate prediction of interface for unknown proteins

Thank You

Collaborator
Ben Mueller

Advisor
Alessandro Senes

Experiments
Ambalika Khadria and Loren Lapointe

CATM