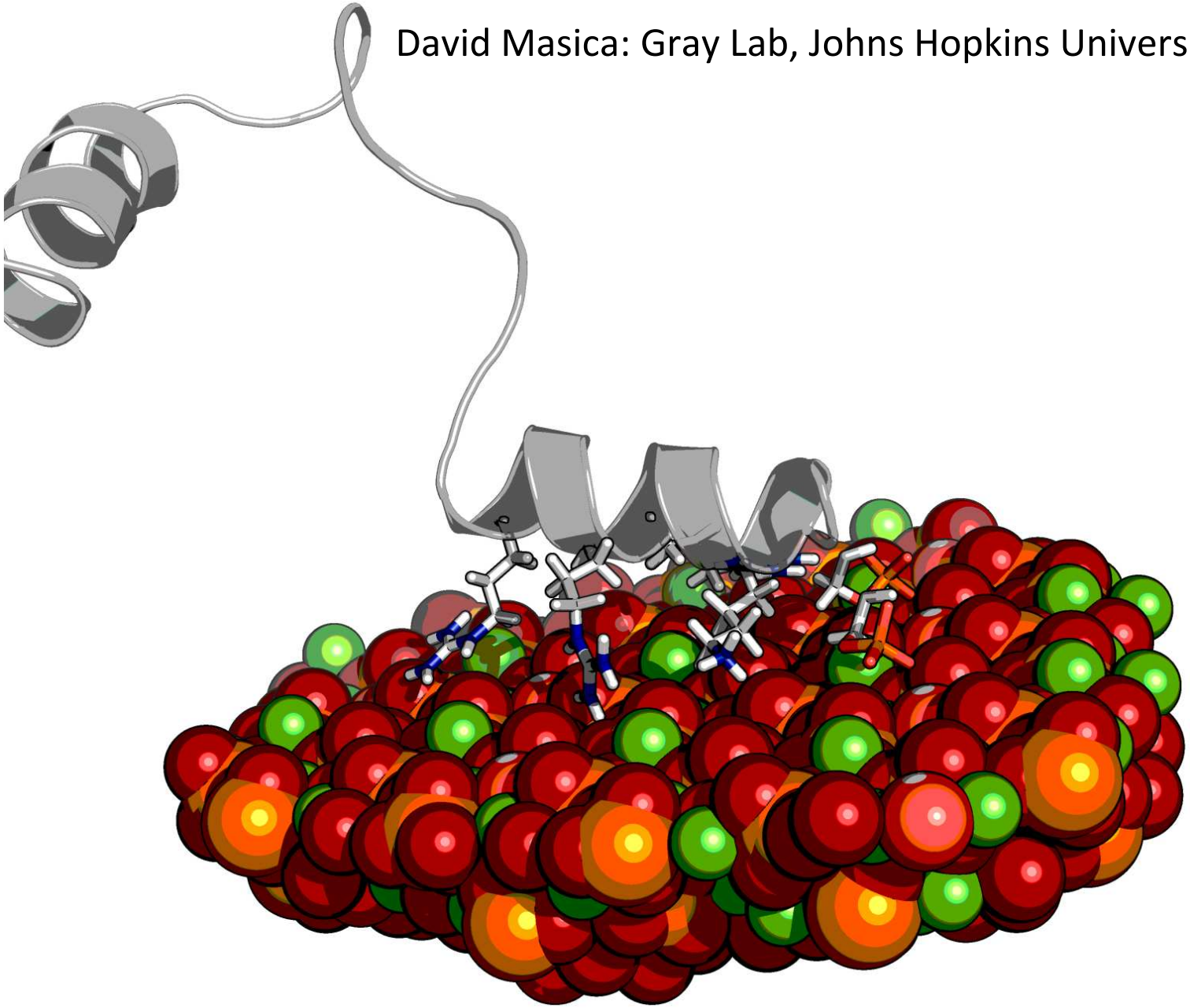
Solution- and Adsorbed-State Ensembles of Biomineralization Proteins with RosettaSurface

David Masica: Gray Lab, Johns Hopkins University



Unpublished
&
Confidential

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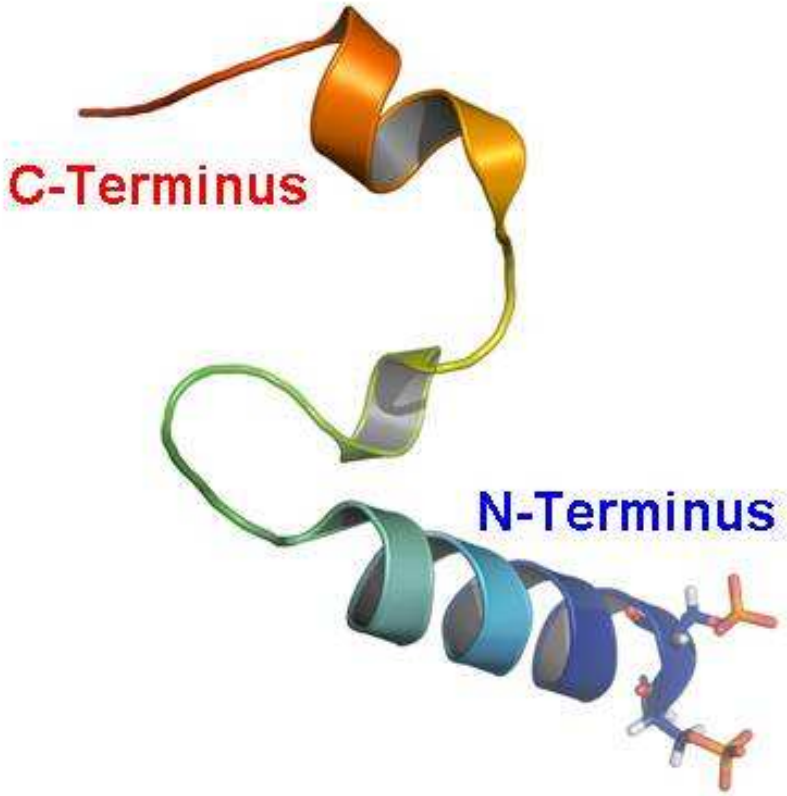
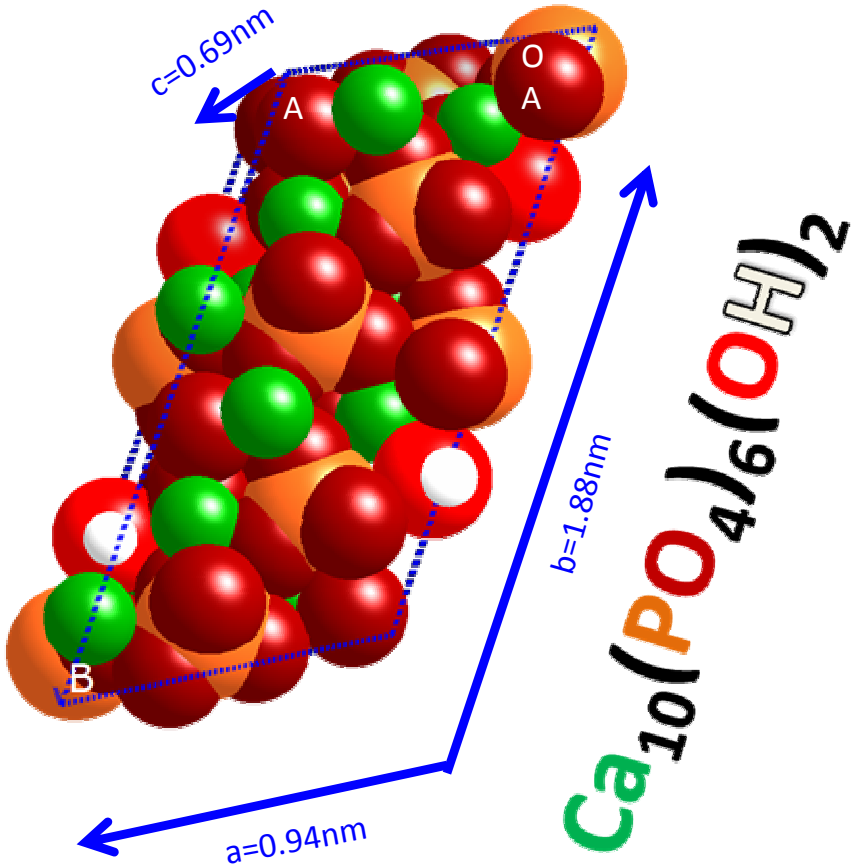
Beckman Foundation

Statherin and Hydroxyapatite (HAp): an Evolved Protein-Surface Interaction

Statherin inhibits the growth of HAp crystals



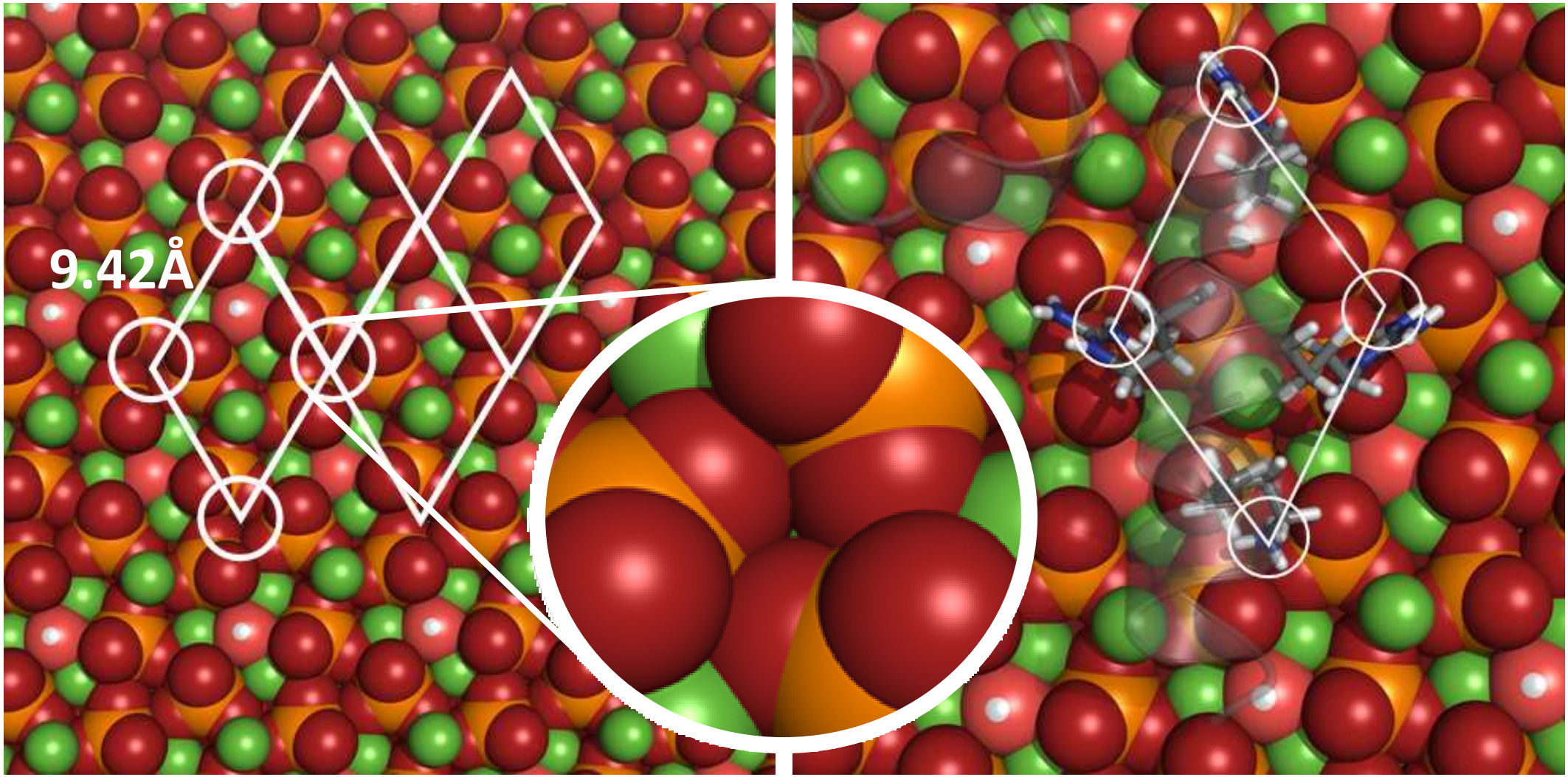
HAp is the primary component In bone and tooth



Goobes, G. et al., *Proceedings of the National Academy of Sciences* **103** (44), 16083 (2006).

The iPOT (interstice of the Phosphate-Oxygen Triad) Motif: a Plausible Molecular Recognition Site

$Ca_{10}(PO_4)_6(OH)_2$



Makrodimitris, K.; Masica, D. L.; Kim, E.; Gray, J. J.; *J. Am. Chem. Soc.* **2007**, 129, 13713-13722.

Key Questions

- Do solution- and adsorbed-state protein folds differ significantly?
- Can RosettaSurface accurately fold a protein on a surface starting from an extended chain?
- Can a combined RosettaSurface-NMR protocol solve a protein structure on a surface?
- Specific or promiscuous binding?

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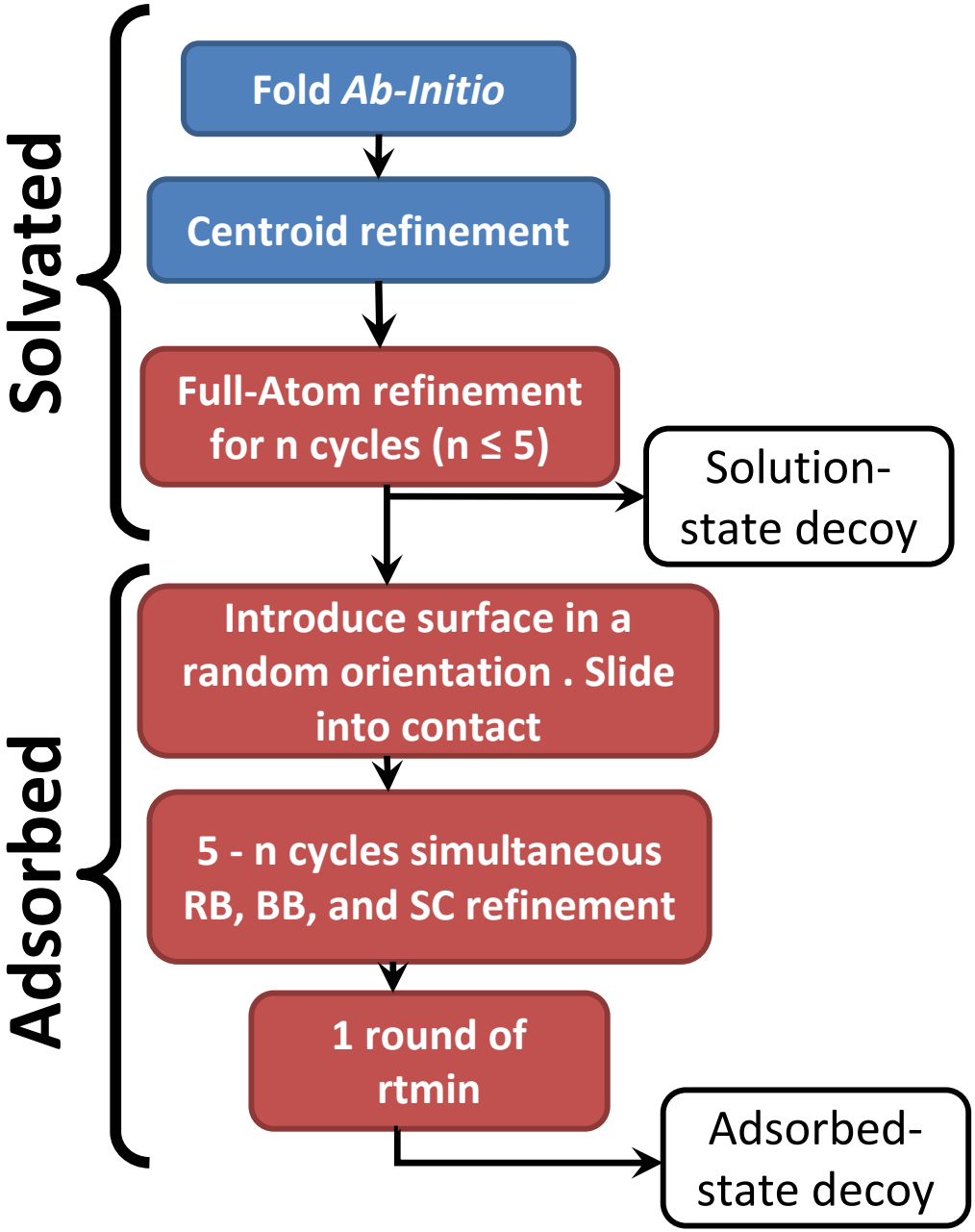
Flow Chart



Centroid | Full Atom

Key features

- Start from linear backbone
- Solution- and adsorbed-state decoys
- Decision to create encounter complex is random

Energy Fcn

- $W(R_g) = 0$
- Self electrostatics
- Final discrimination by non-weighted physical potential

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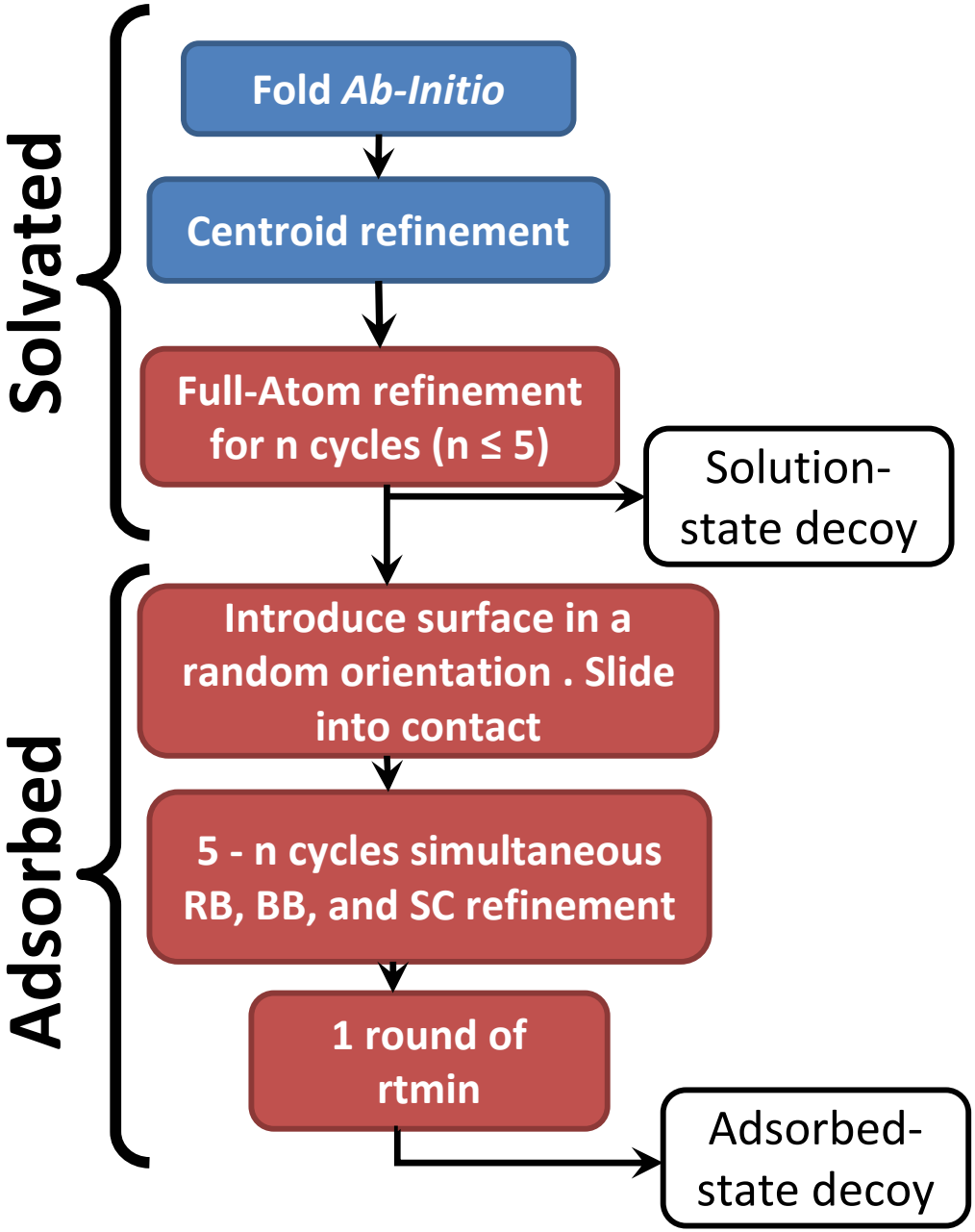

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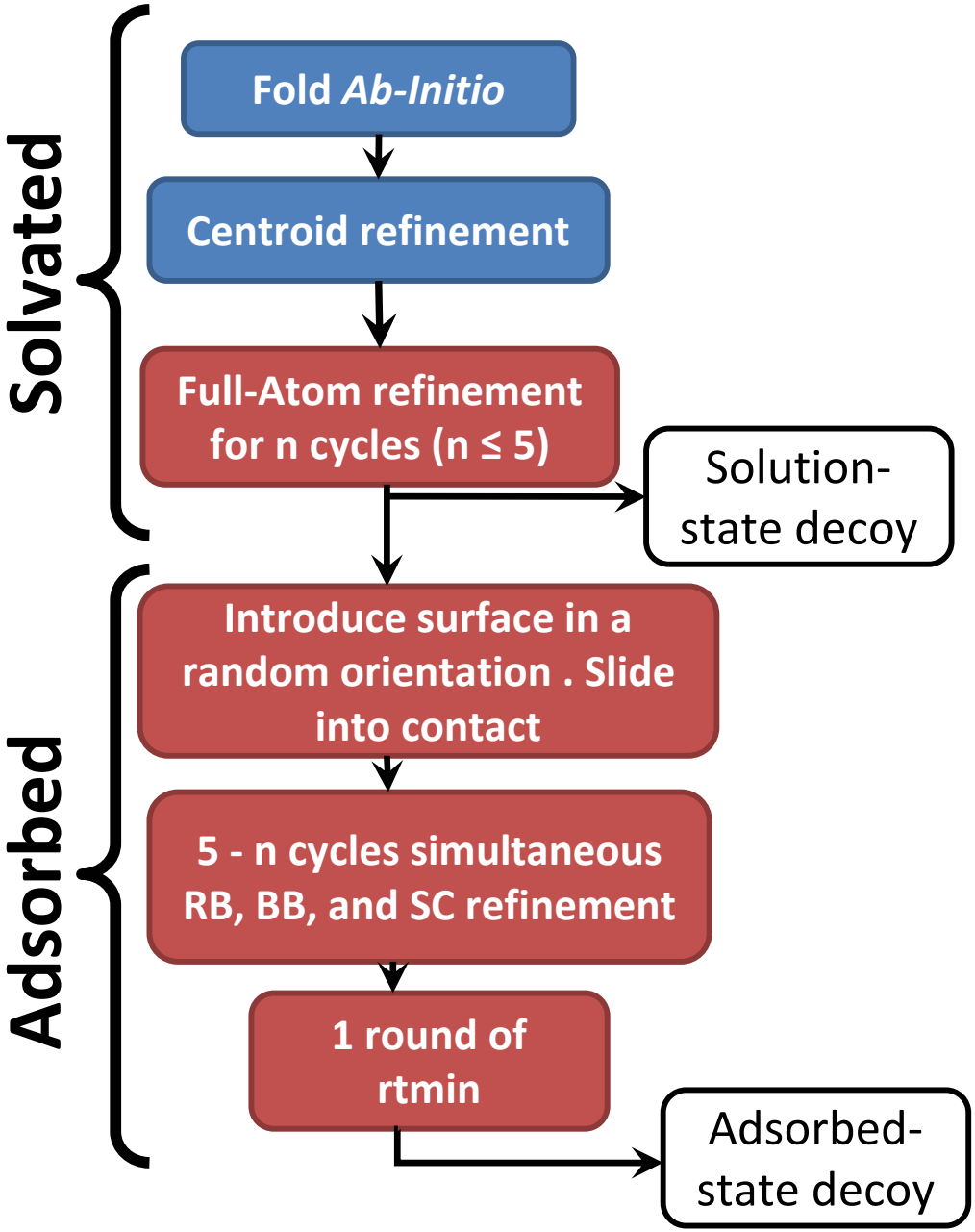

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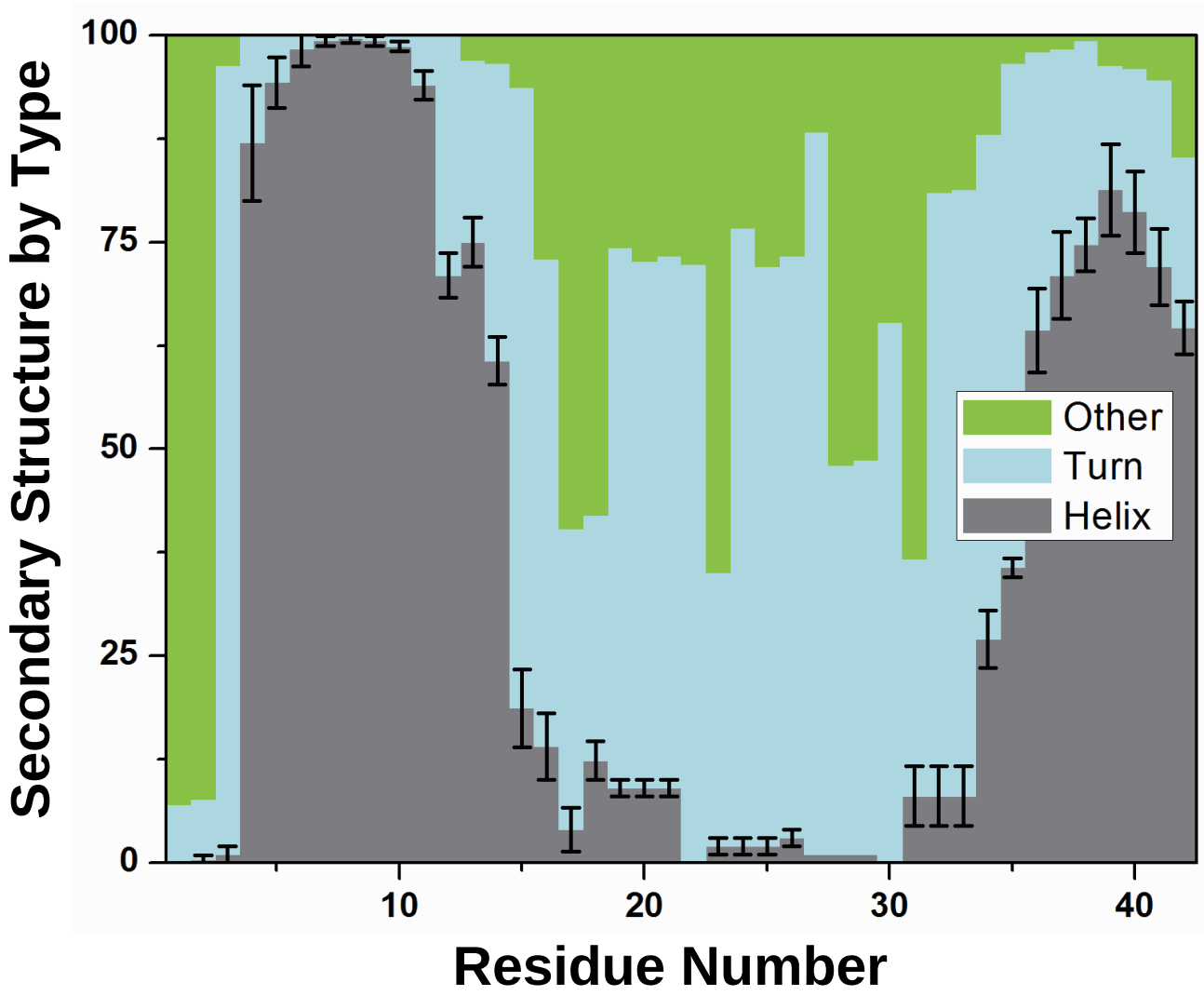
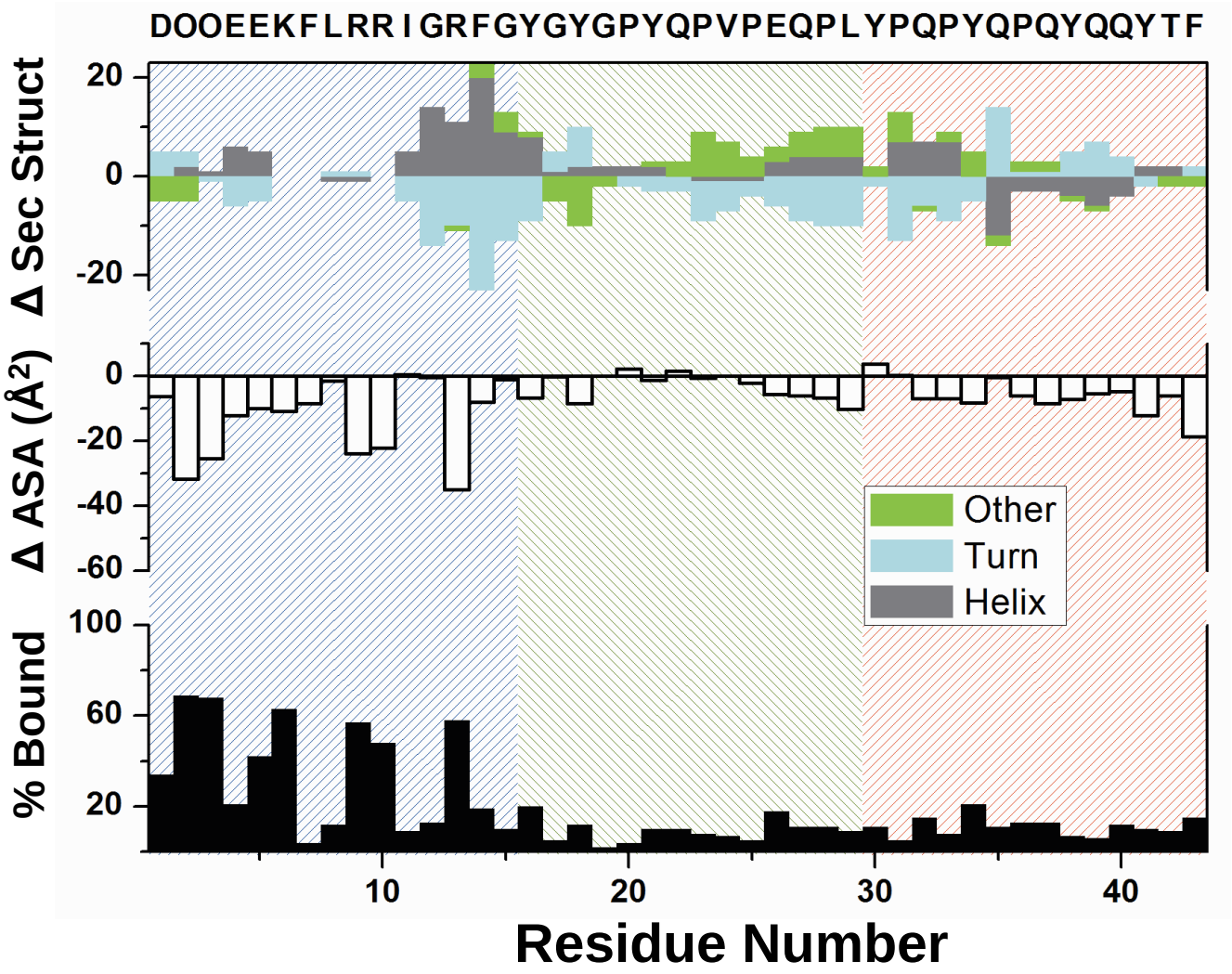
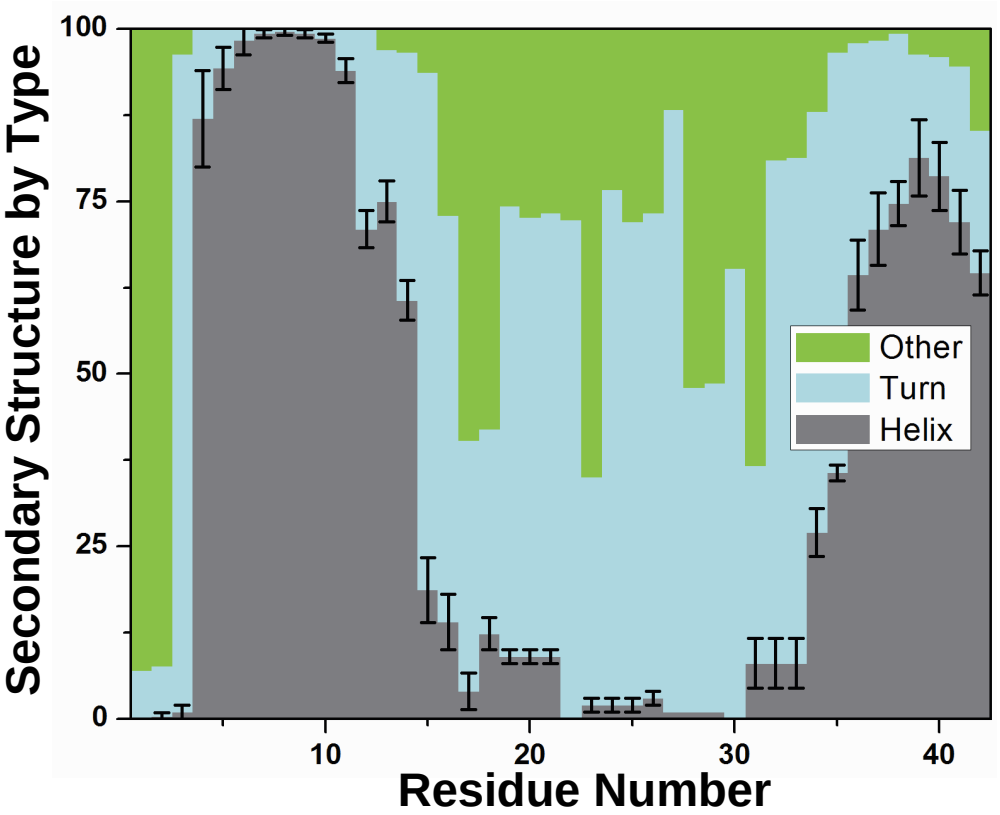
Solution-State

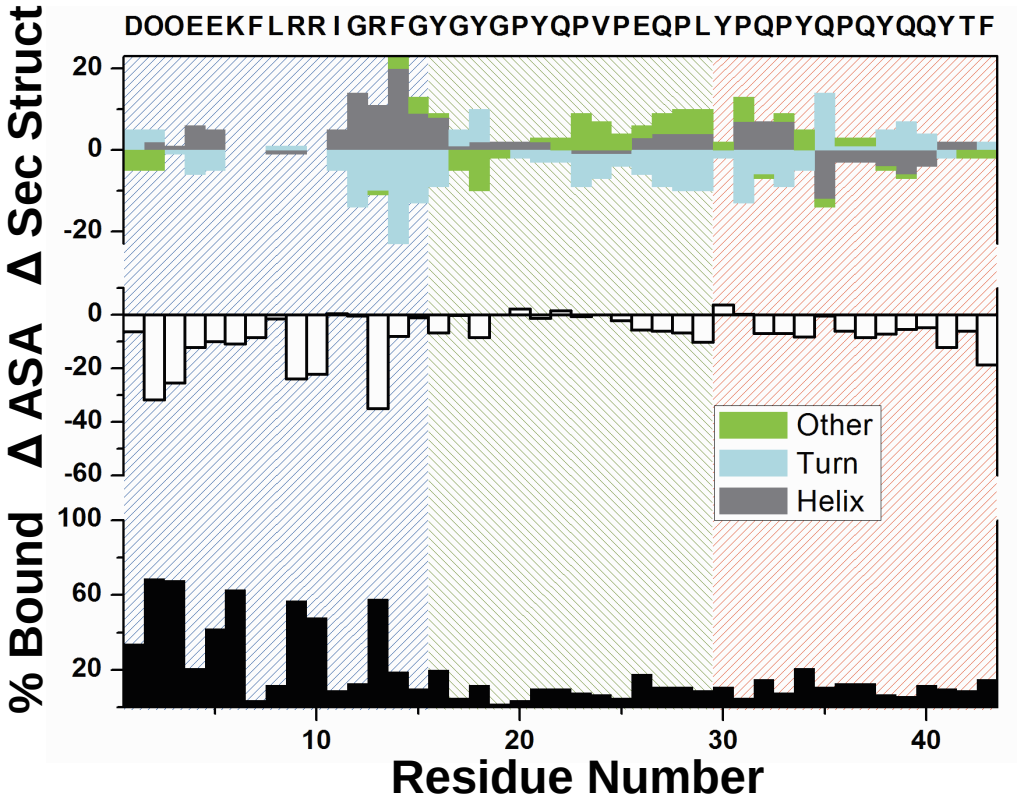


Adsorbed-State

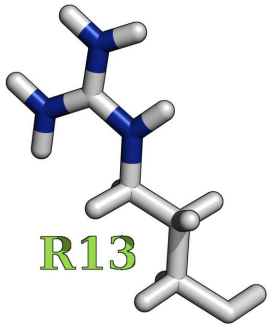


Solution-State -> Adsorbed-State

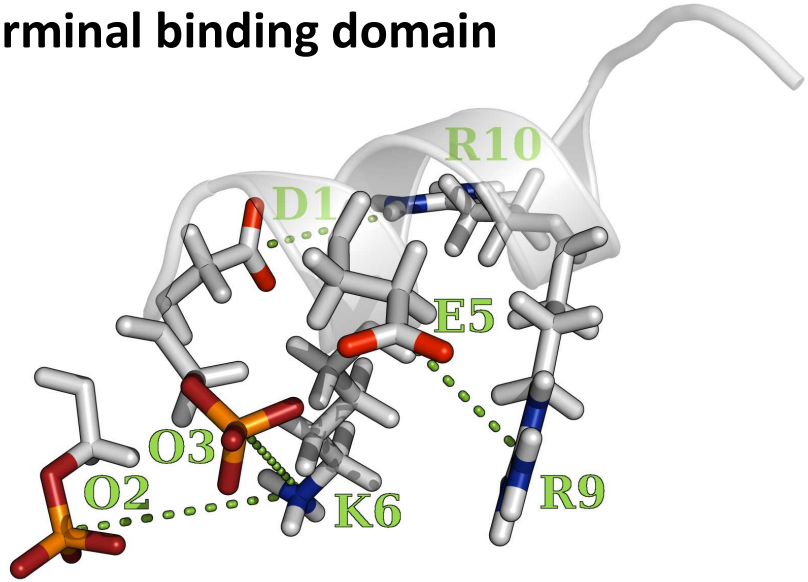




Folding Event Around Glycine 12

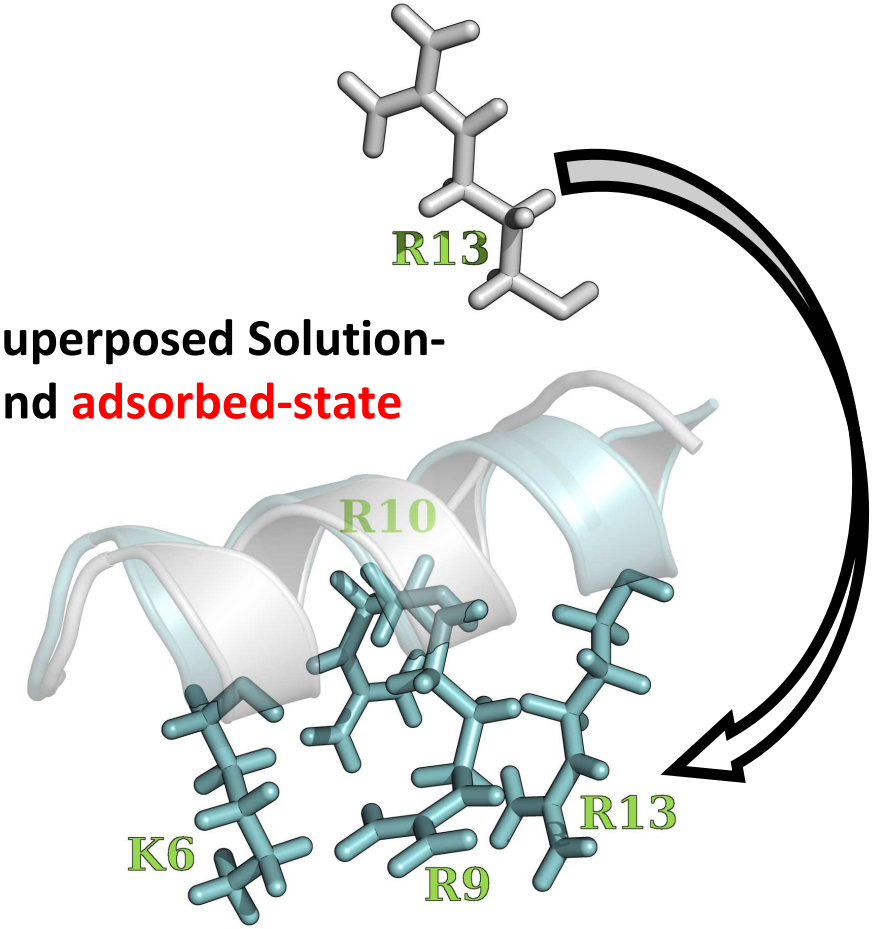


Solution-state 15mer
N-terminal binding domain



R13 anti-parallel to binding motif

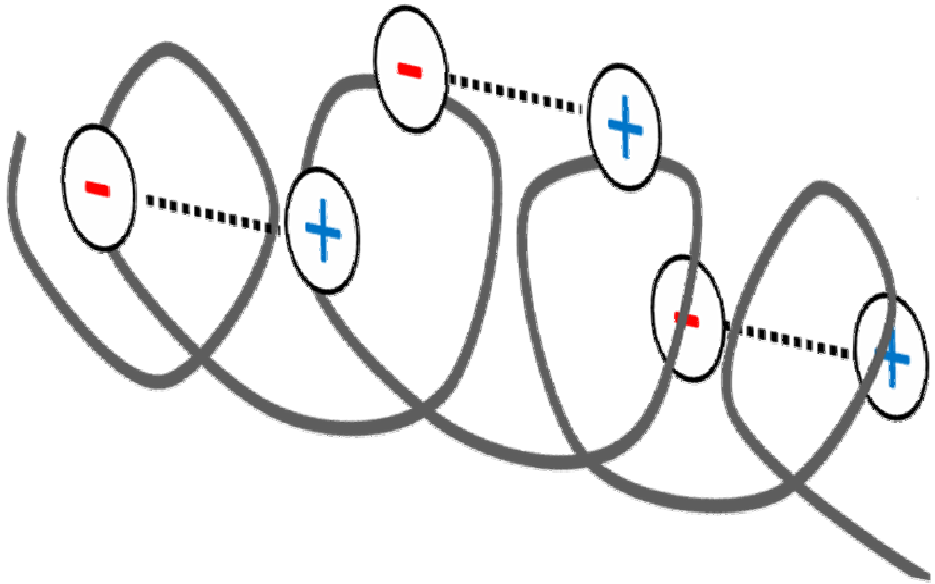
Superposed Solution-
and **adsorbed-state**



R13 parallel to binding motif

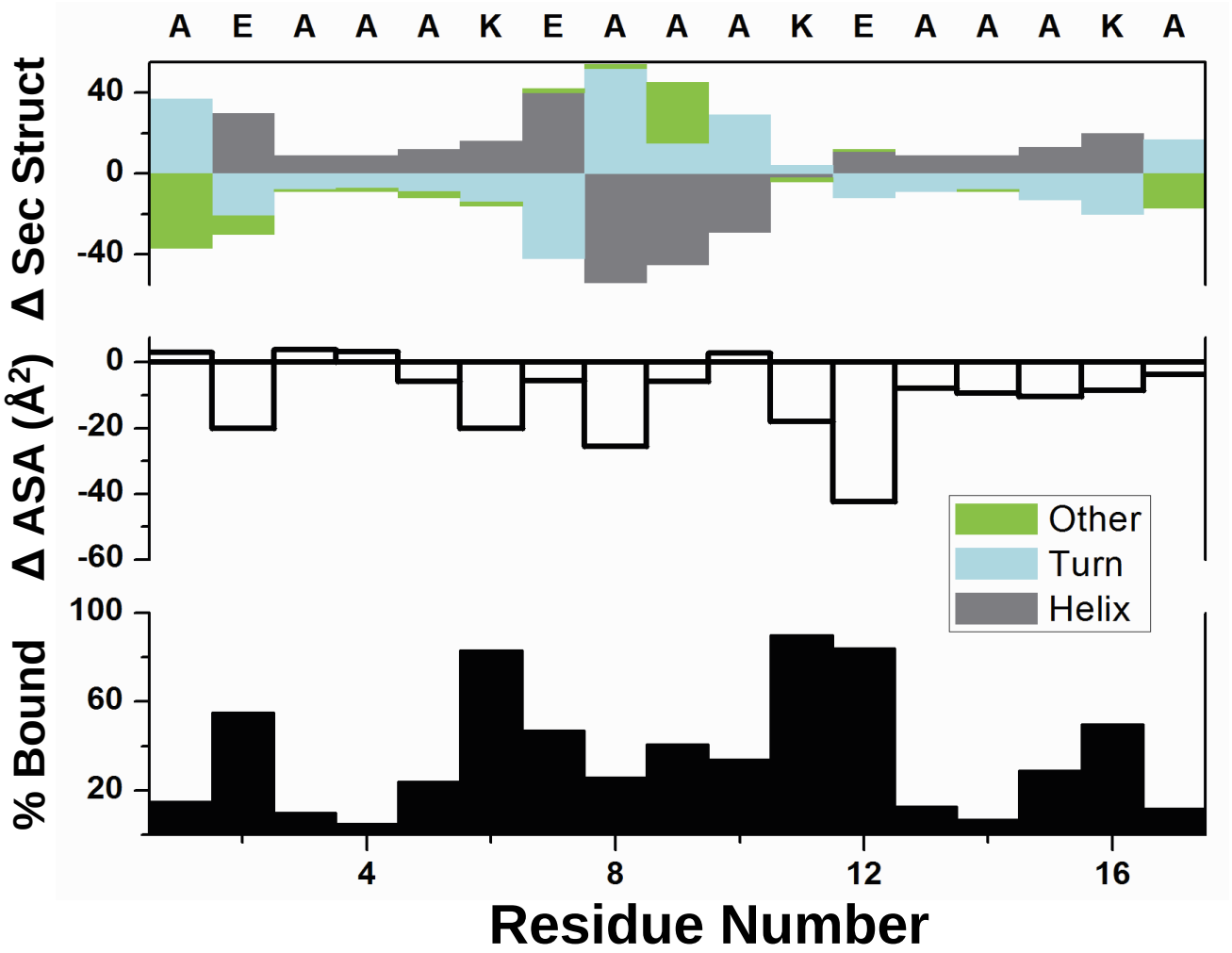
A Control: the Schematic

Predicted helical fold stabilized by electrostatic interactions

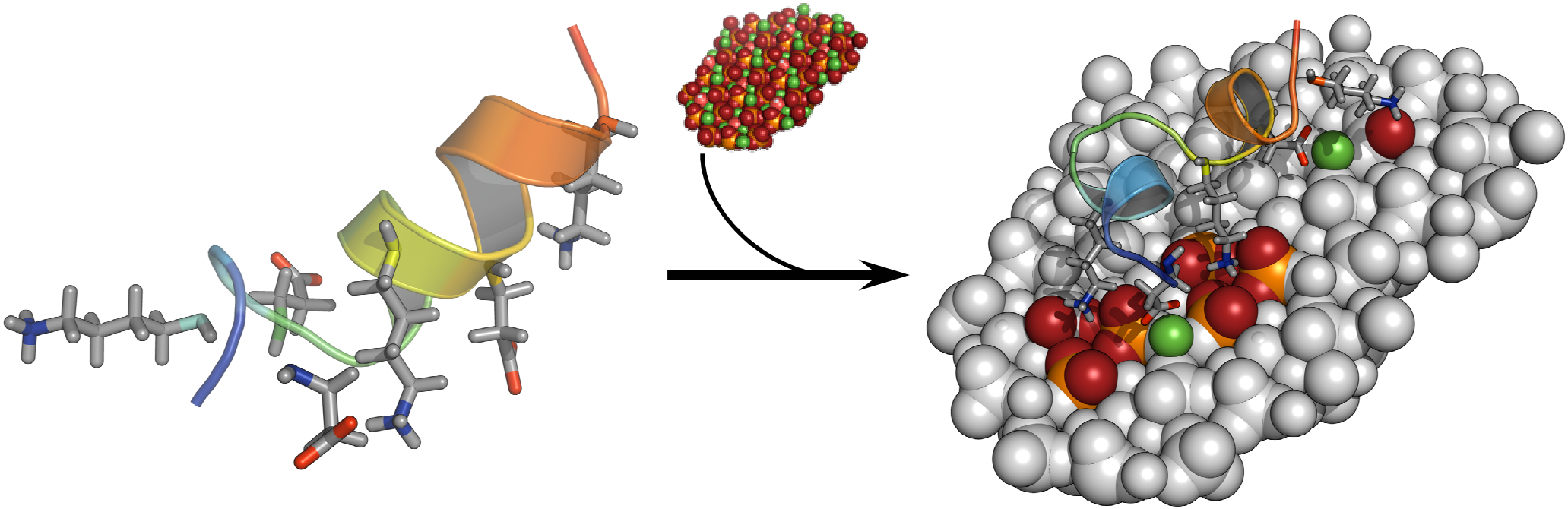


Marqusee, S. et al., *Proceedings of the National Academy of Sciences* (1989).

A Control: Statistics from the Top 100

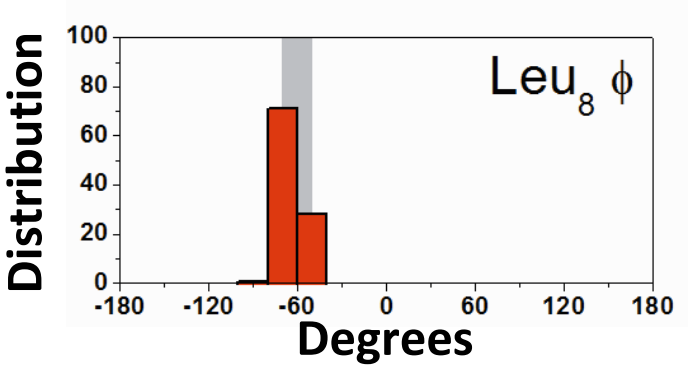


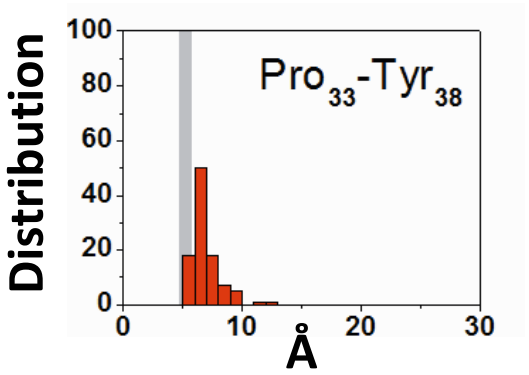
A Control: Representative Structures

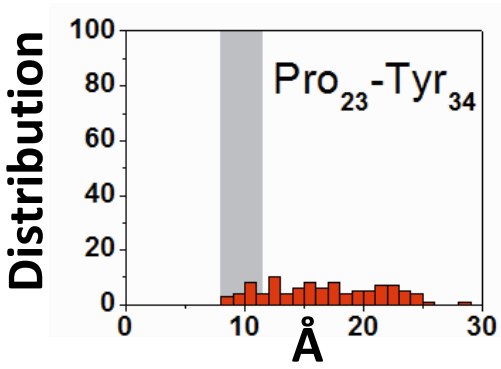


Comparison with High-Resolution Solid-State NMR Measurements, for the Statherin-HAp System

Unbiased Simulation:







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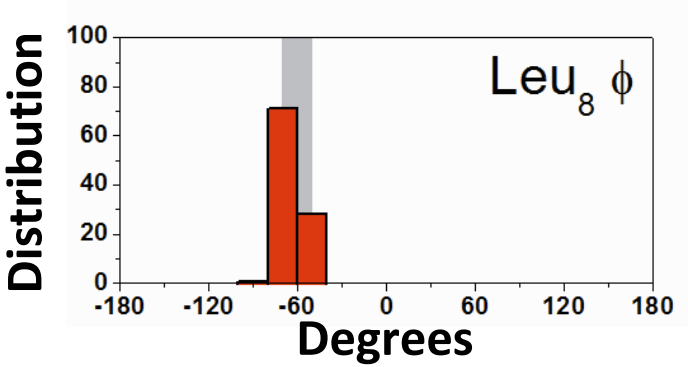
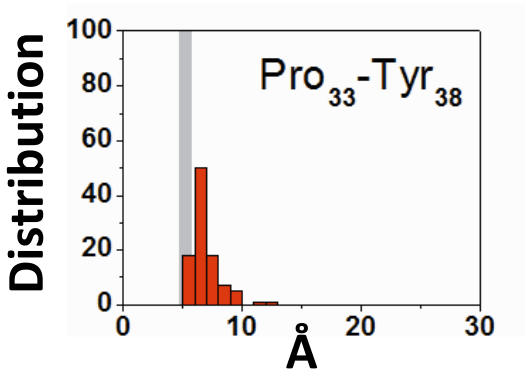
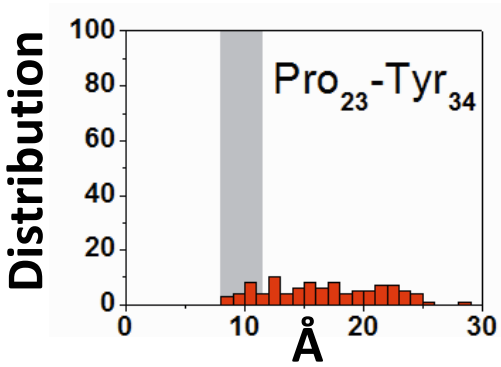
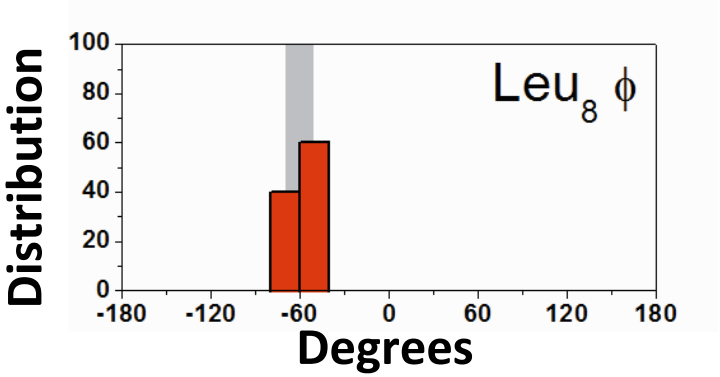
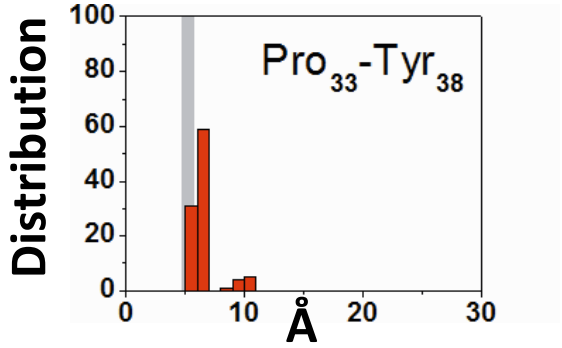


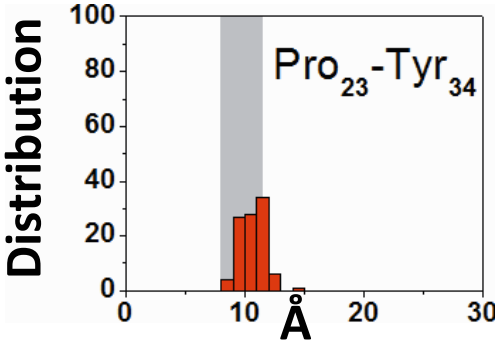




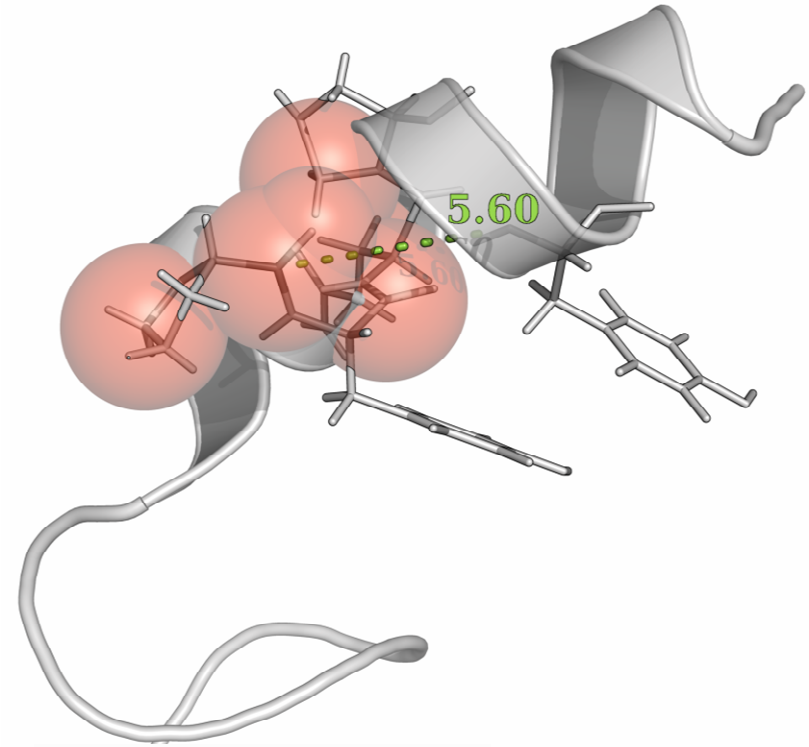
Biased Simulation:

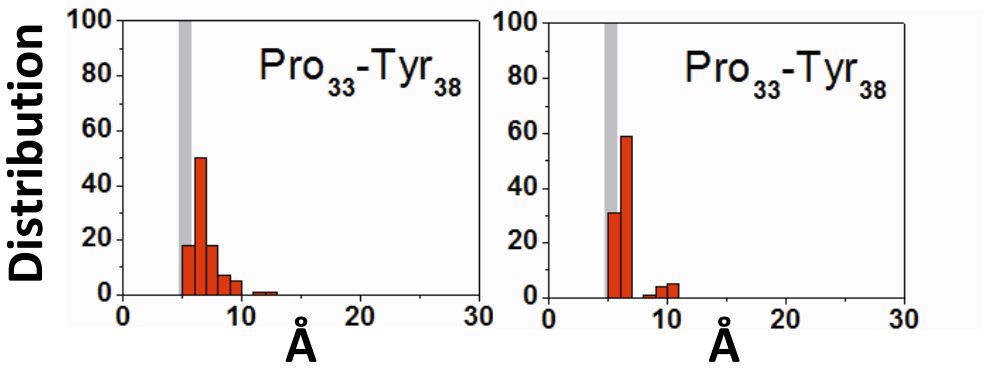




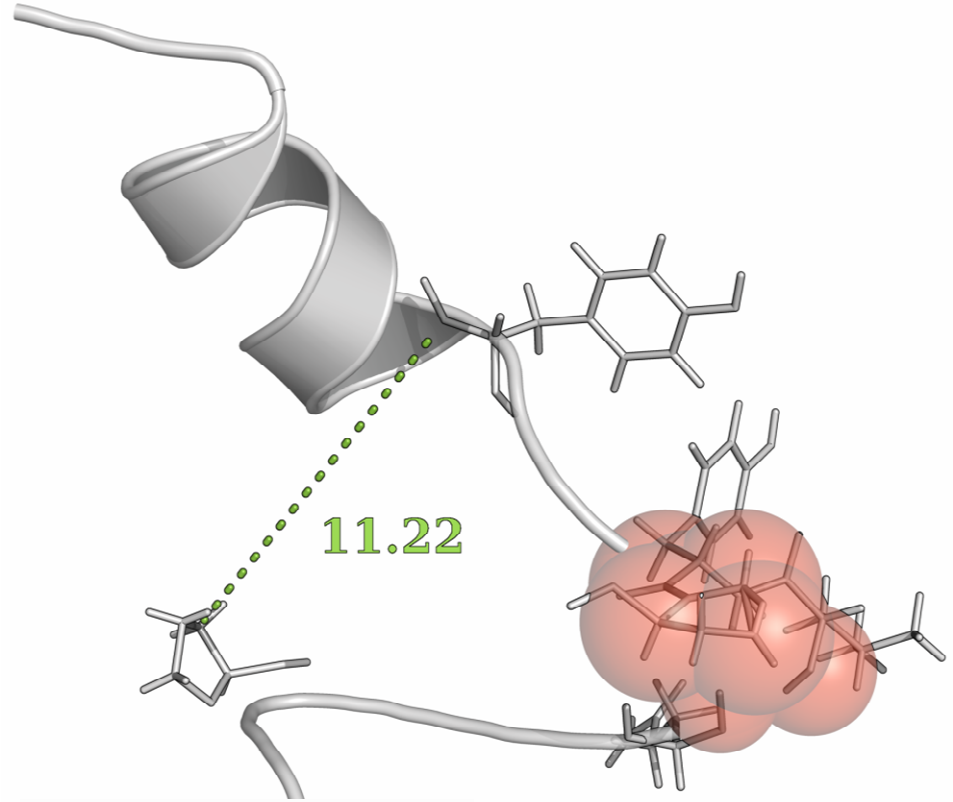


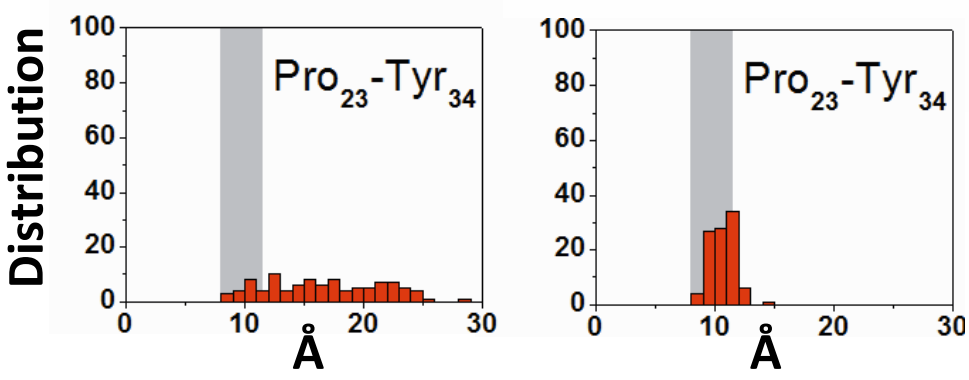
Enforcing Two NMR Constraints Created Clashes





Unbiased → Biased

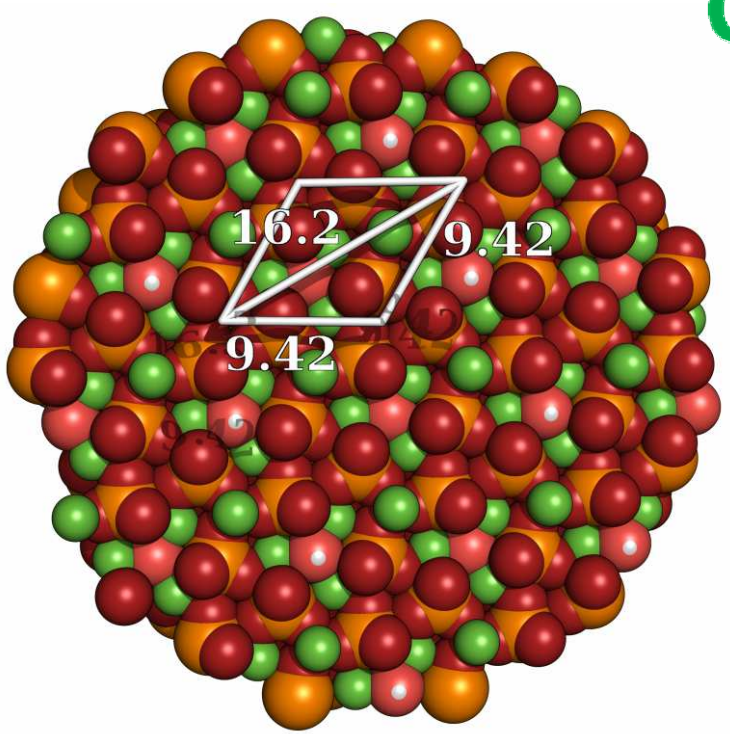




Unbiased → Biased

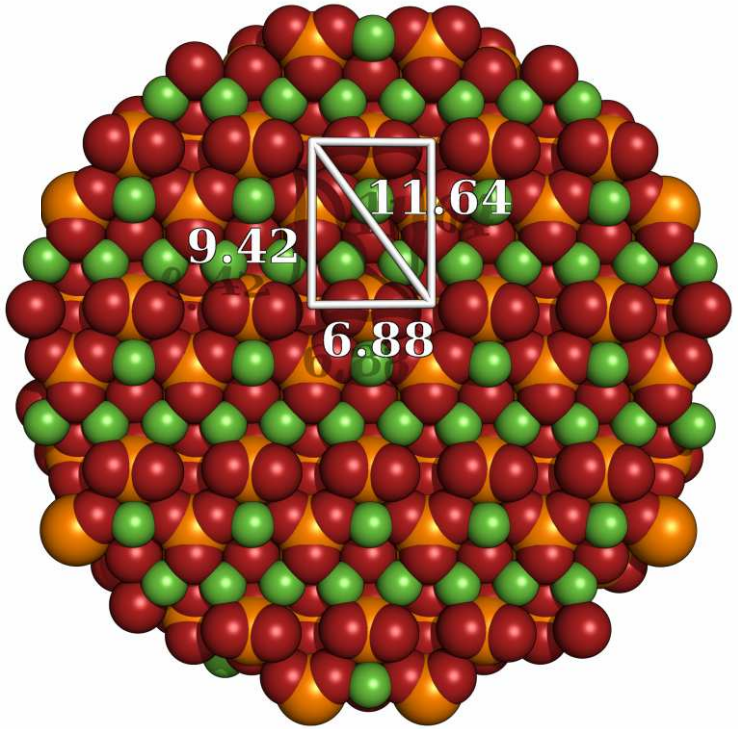
Specificity?

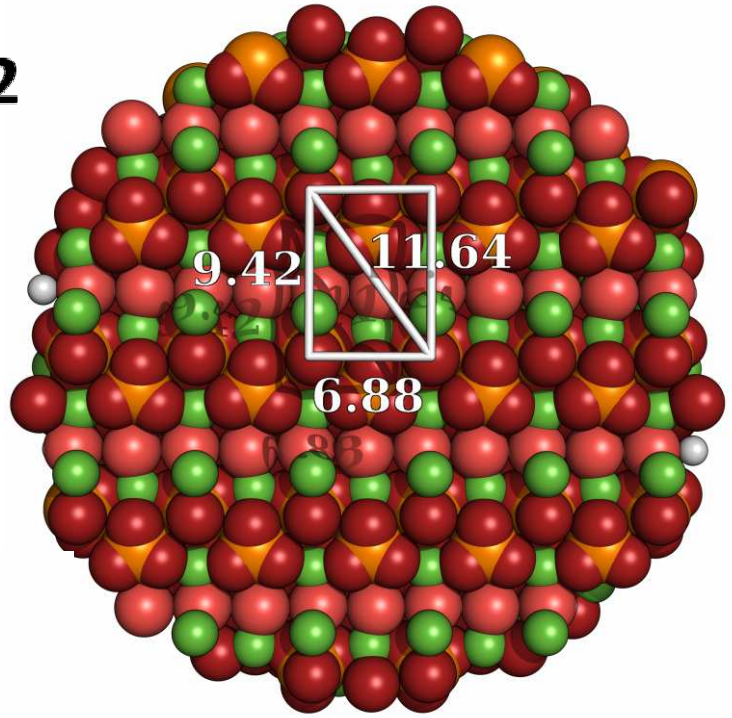
$Ca_{10}(PO_4)_6(OH)_2$



001

010



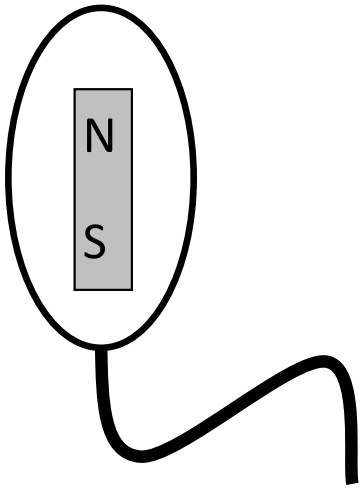


100

Conclusion

- RosettaSurface suggests that statherin undergoes moderate structural change upon binding
- RosettaSurface captures many molecular and atomic features of the statherin-HAp system, and can do so beginning from a fully-extended chain in solution
- A Combined NMR-RosettaSurface protocol may prove useful for determining protein-structures at interfaces
- Similar structures result when adsorbing statherin to the 001, 010, and 100 faces of HAp

Do Proteins Interact with Inorganic Materials in Nature?



Magnetite

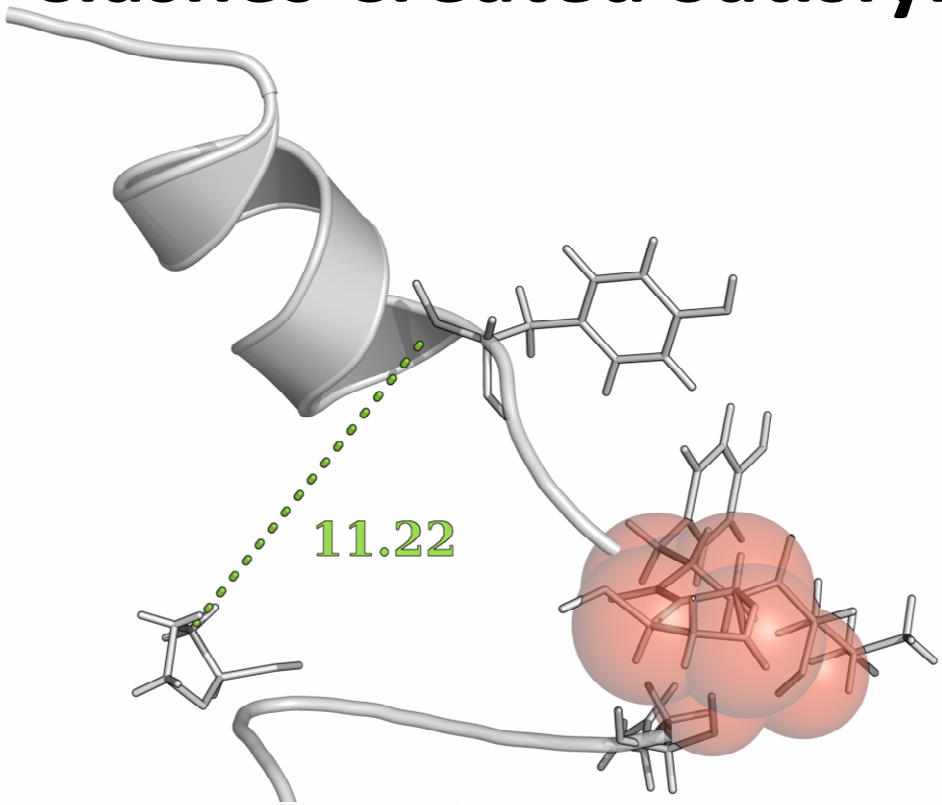
Ice

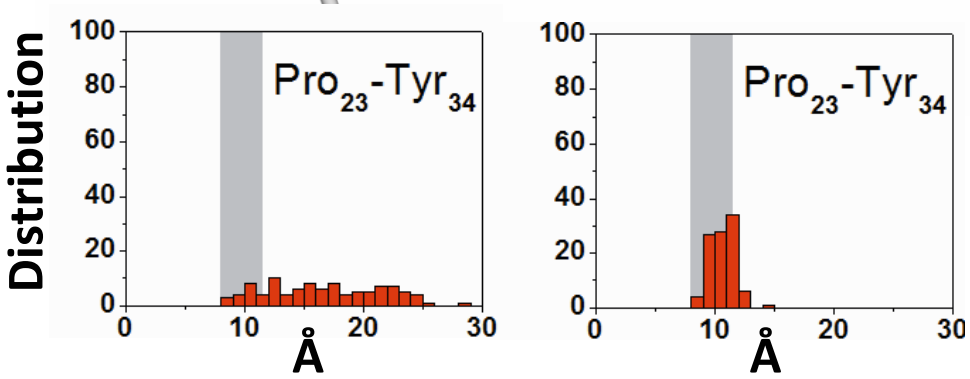
Silica

Hydroxyapatite

Calcite/Aragonite

Clashes Created Satisfying Some NMR Constraints





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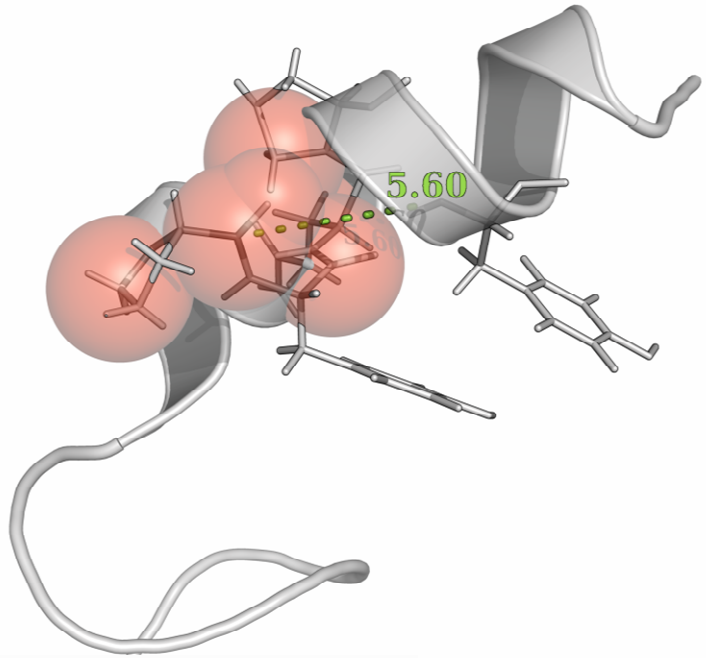

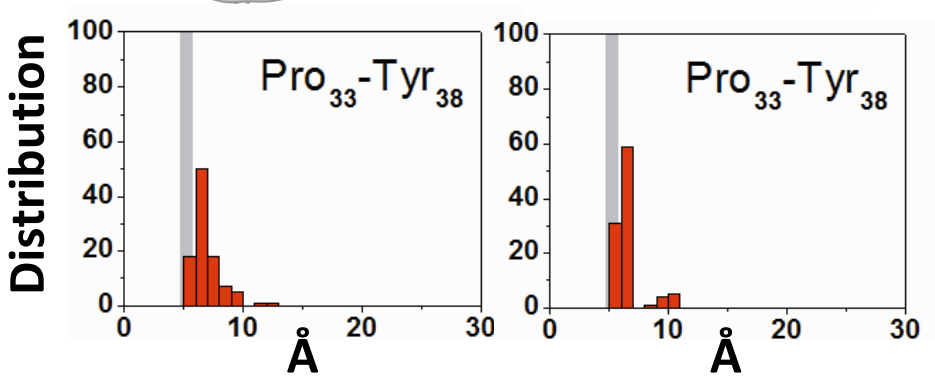

Unbiased → Biased

Simulate Protein-Surface Interactions?

- Why not
  - No structural models solved by experiment, i.e. NMR or crystal structures. Therefore, no training sets and no benchmarks!
- Why
  - Simulation provides the ONLY means of solving the structure of a protein adsorbed to a solid-surface

Why a Second Statherin-HAp Study?

- No other system has been the subject of as many high-resolution solid-state NMR studies; 15 measurements to date.
  - 3 protein-surface intermolecular
  - 7protein intramolecular
  - 5 protein backbone torsion angles