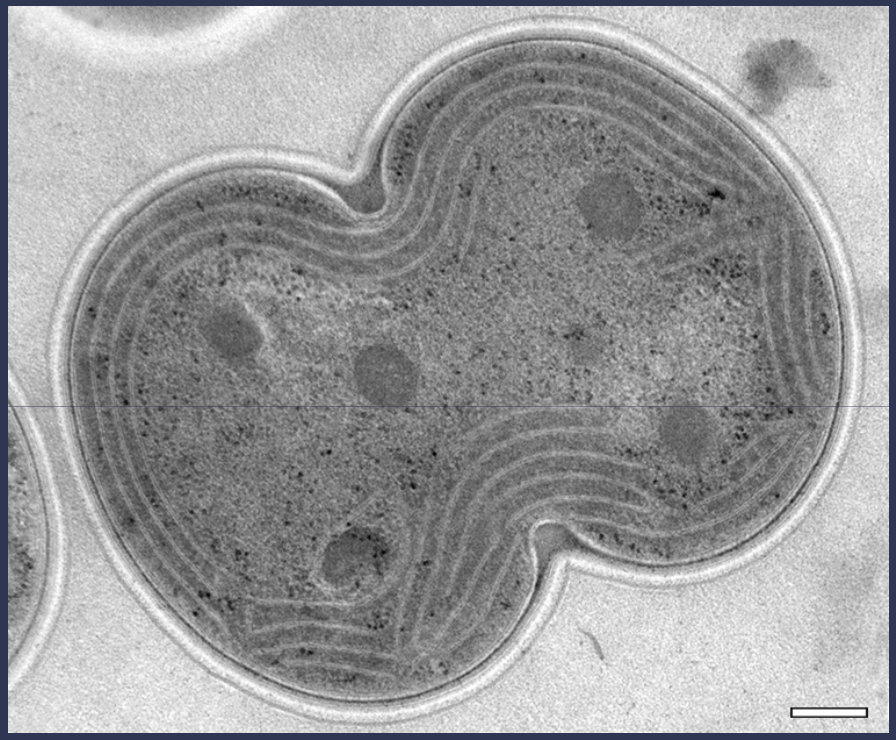
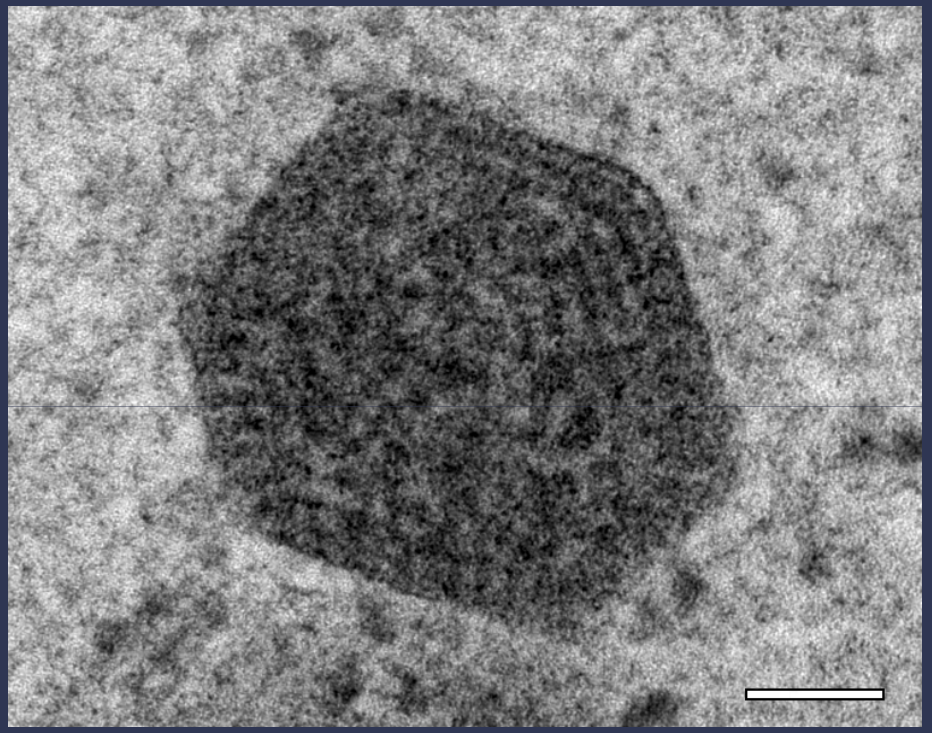
Elucidating the Structural Basis of Carboxysome Function: A Progress Report

The Carboxysome, a Prokaryotic Organelle Composed Entirely of Protein, in Cyanobacteria and Chemoautotrophs



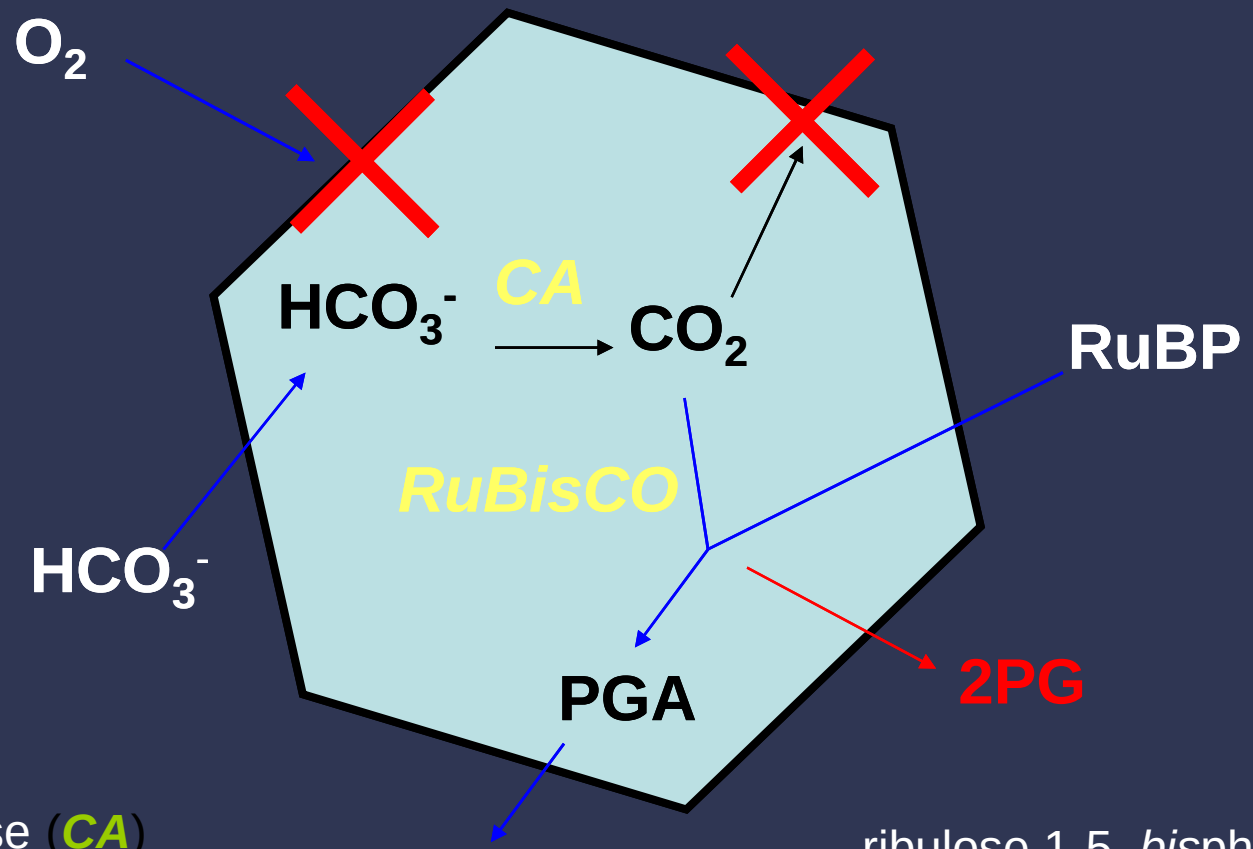
Scale bar = 200 nm



Scale bar = 50 nm

Transmission electron micrographs of *Syn* 6803 cells courtesy of Robby Roberson, Wim Vermass and Allison van de Meene (Arizona State University).

The carboxysome is a bacterial organelle for fixing CO2



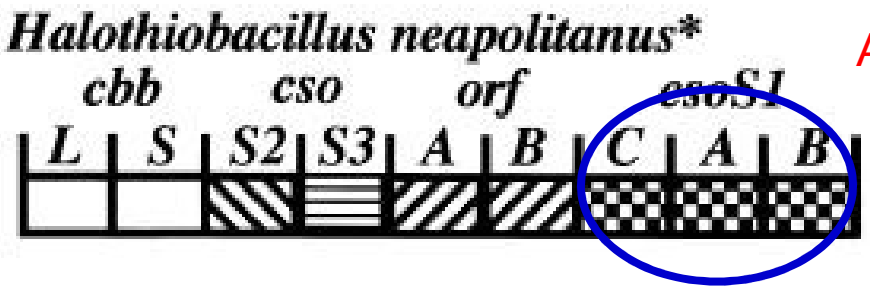
carbonic anhydrase (***CA***)

Ribulose bis-phosphate (RuBP)

ribulose 1-5, *bis*phosphate carboxylase-oxygenase (***RuBisCO***).
3-phosphoglycerate (PGA)

2PG, 2-phosphoglycolate (Eisenhut et al., 2006)

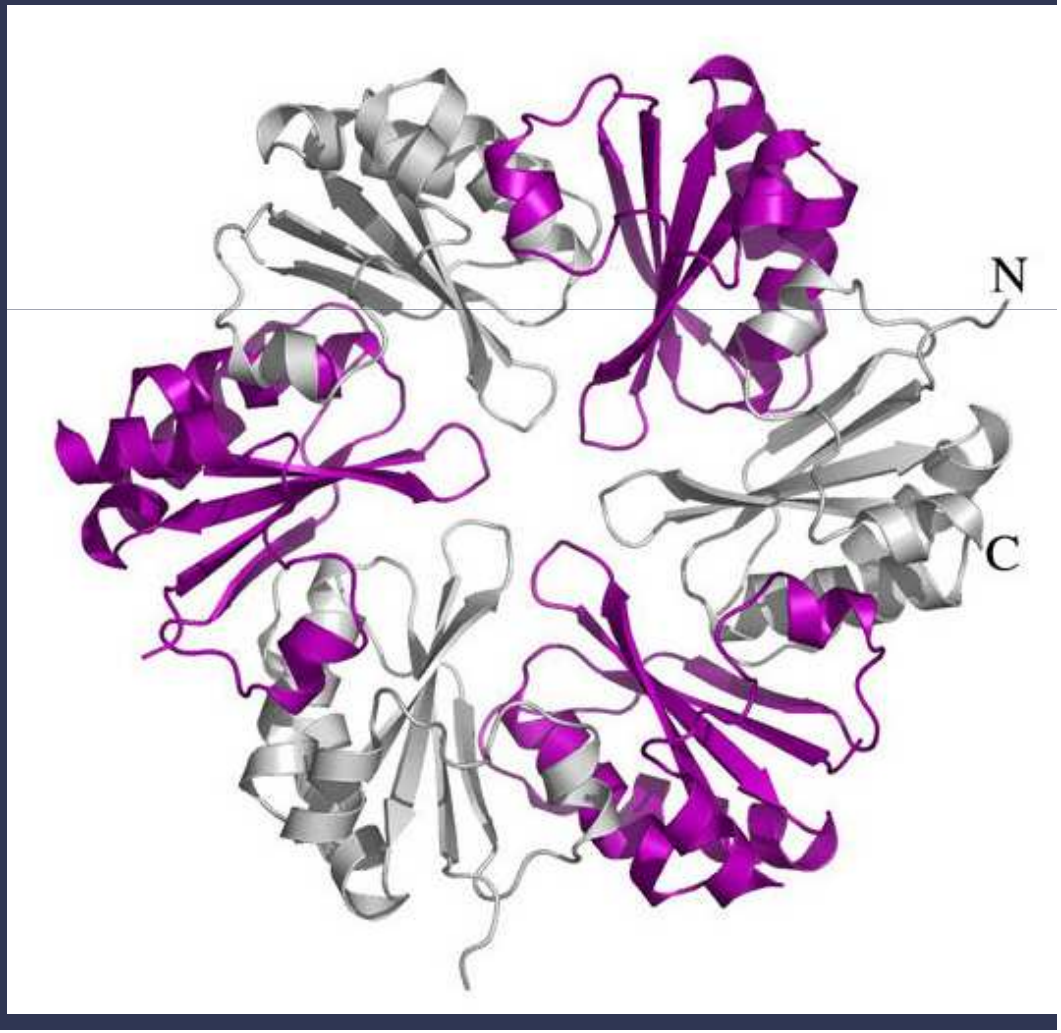
Carboxysome Genes

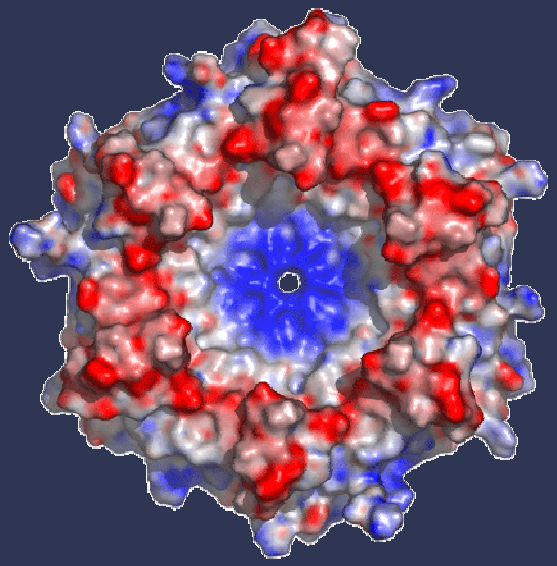


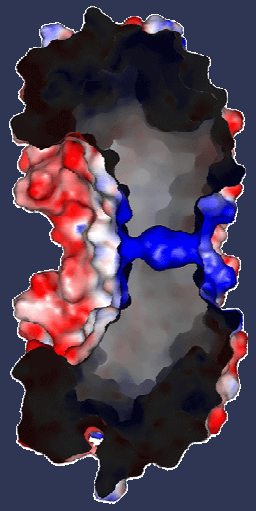
Alpha

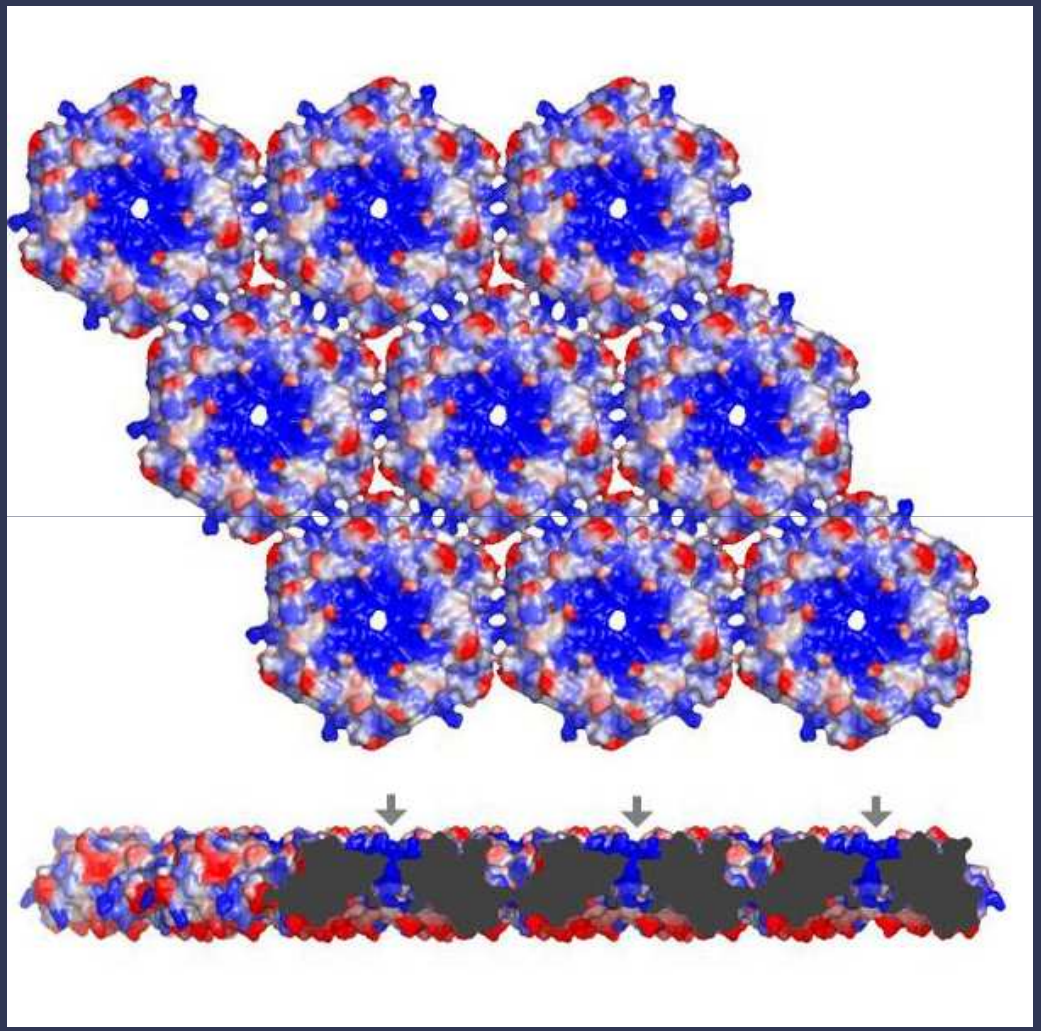
Beta

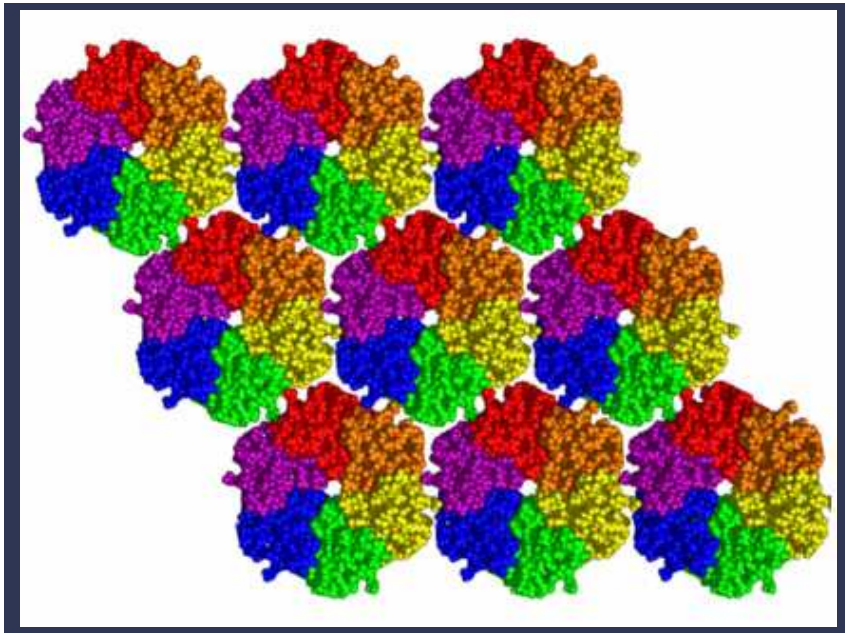
Alpha


N
C

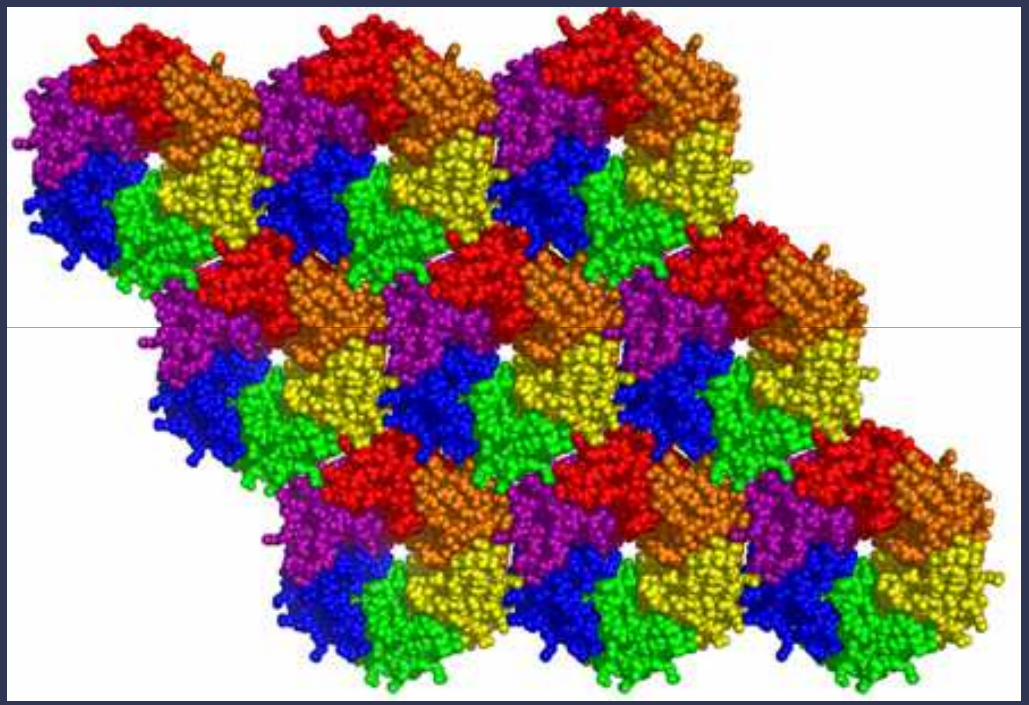








CcmK2 *Syn* PCC6803 (Beta)

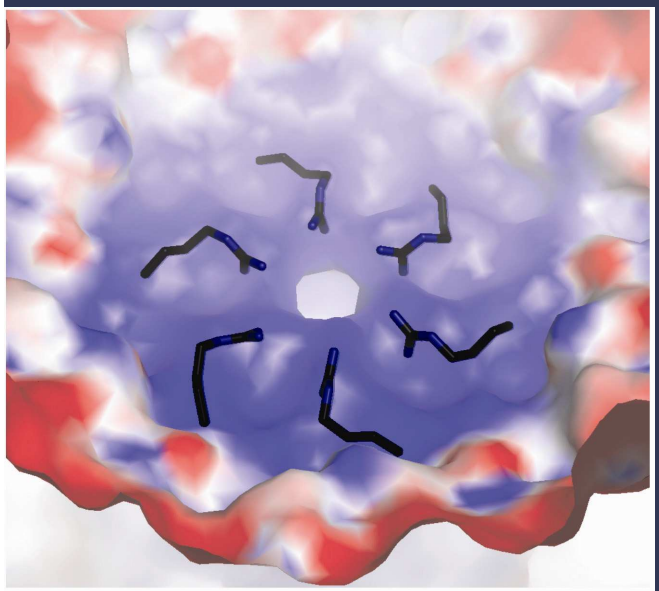


CsoS1A *H. neopolitanus* (Alpha)

Tsai et al., PLoS Biology, June 07

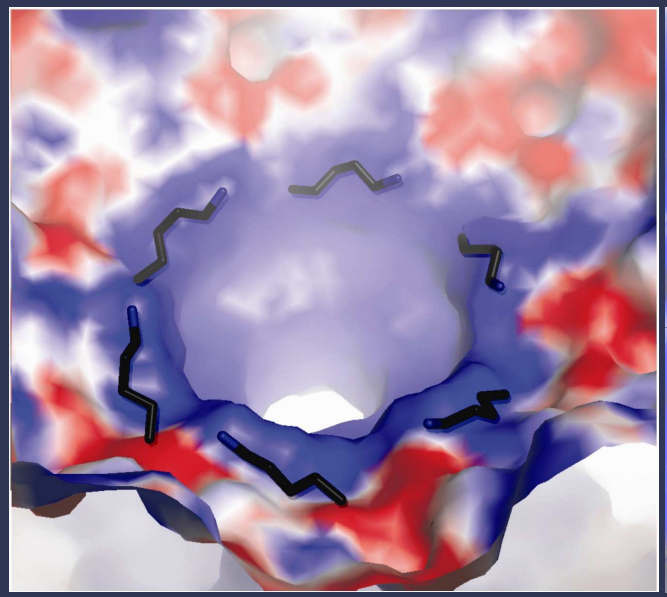
A comparison of pores

CcmK4



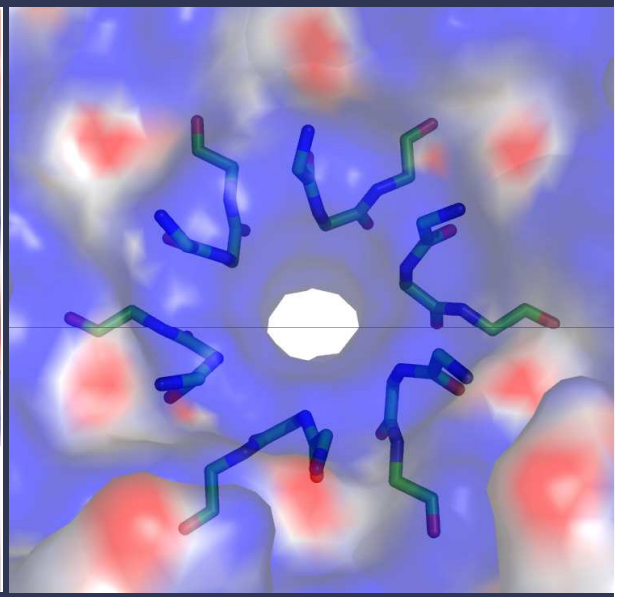
Arg 38

CcmK2/CcmK1



Lys 36

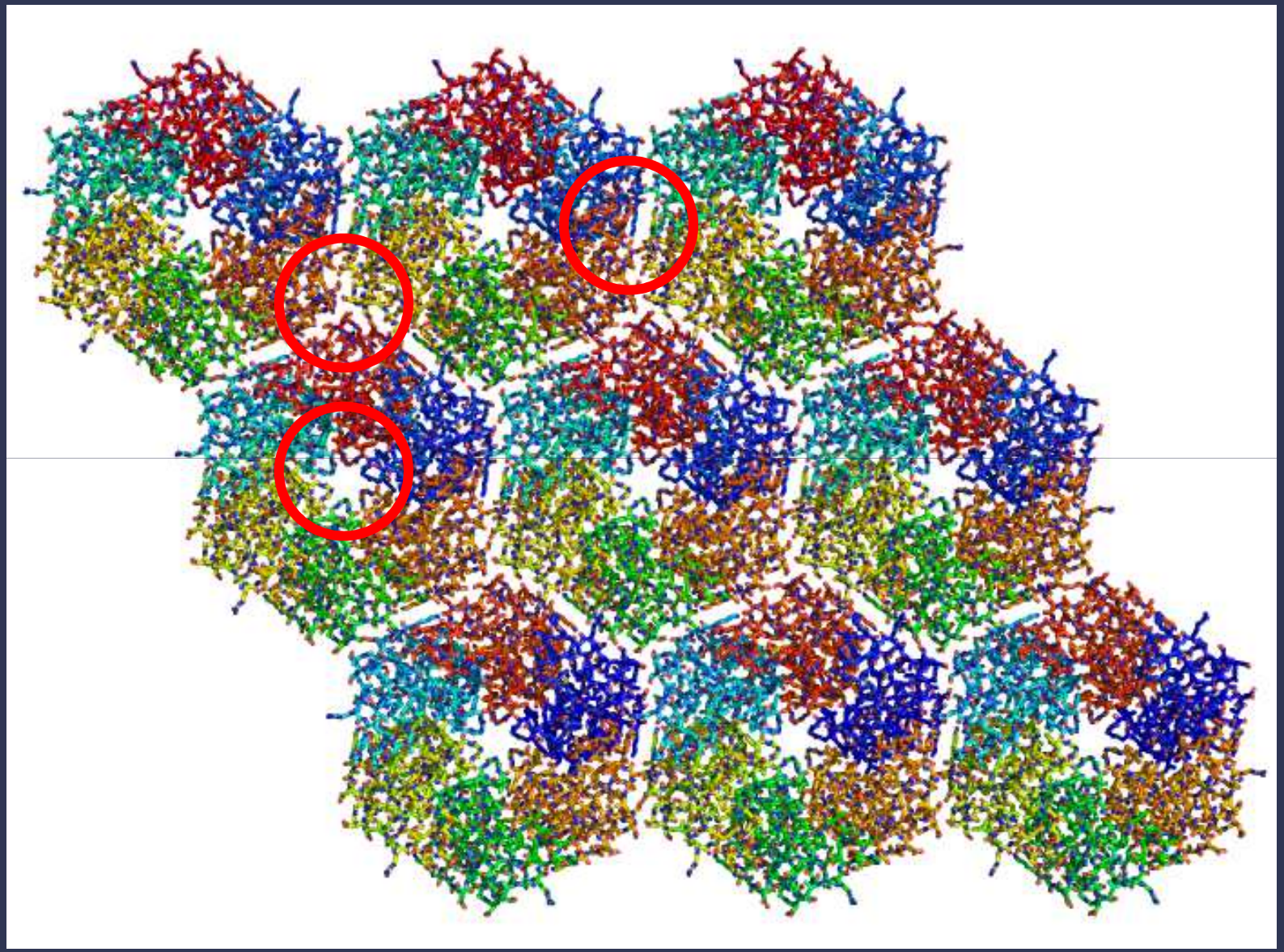
CsoS1A



Loss of charge in pore: K→ F
G triplet, inward facing amides

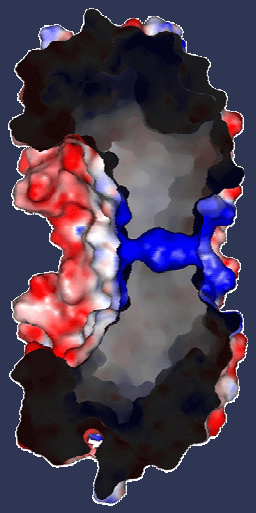
```
CcmK2   ----LVGYEKIGSGRVTVI---
CsoS1A  ----LVGRQFVGGGYVTVL---
```

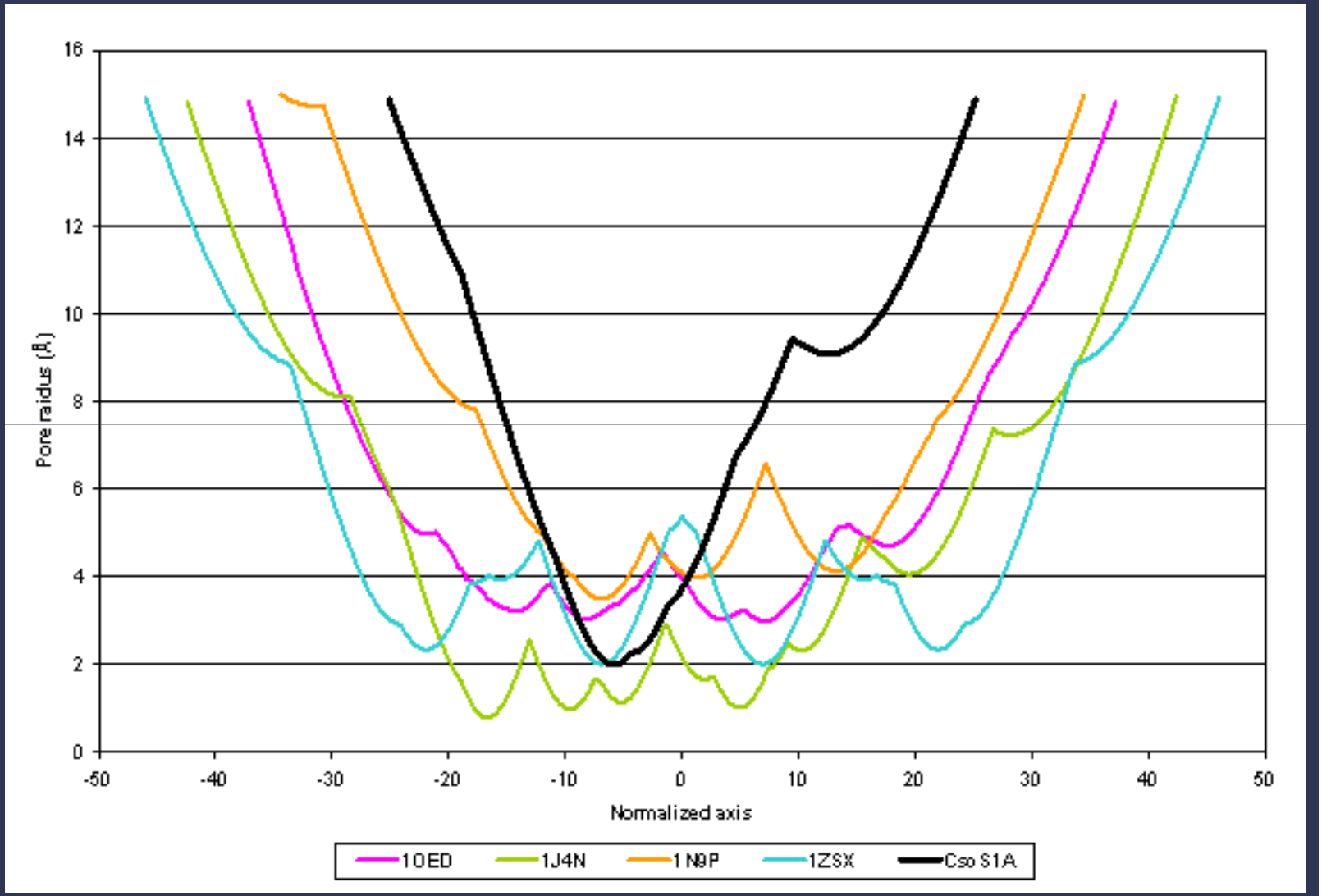
Sulfate Soaks



Tsai et al., PLoS Biology, June 07

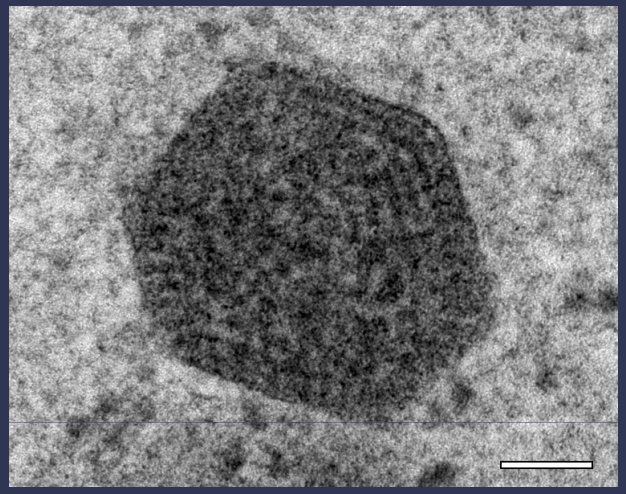
Analogy to membrane channel proteins

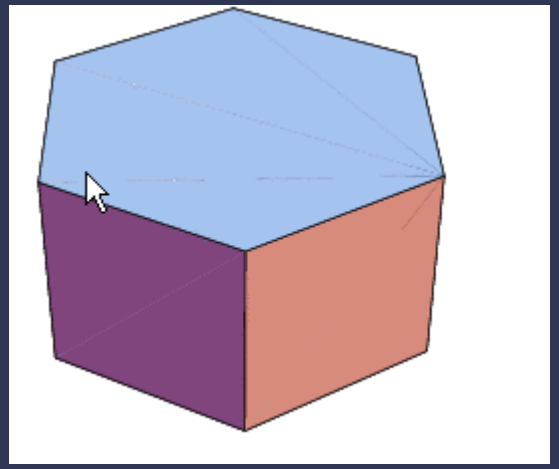


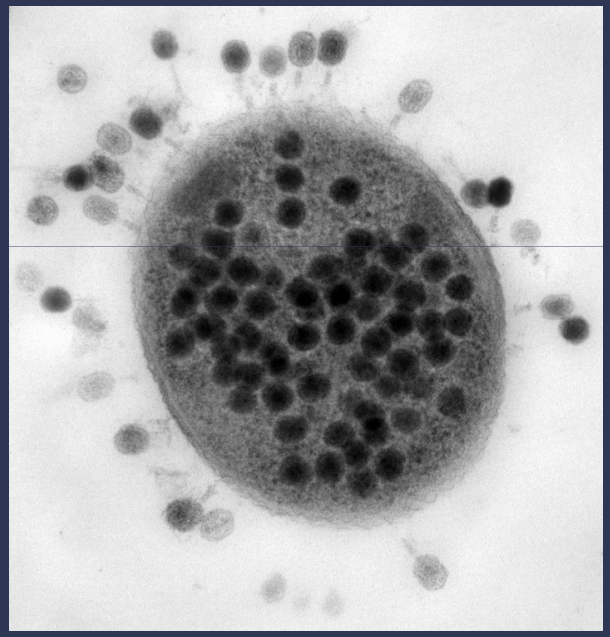


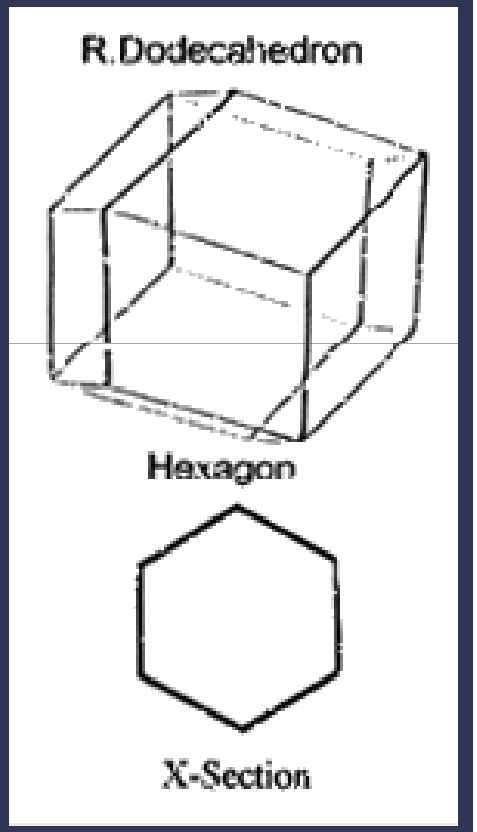
Tsai et al., PLoS Biology, June 07

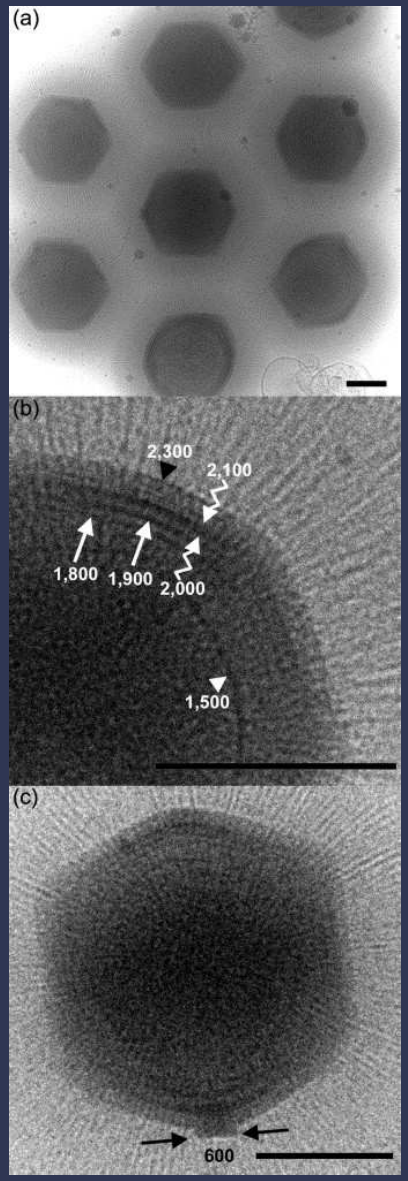
Possible Geometries?





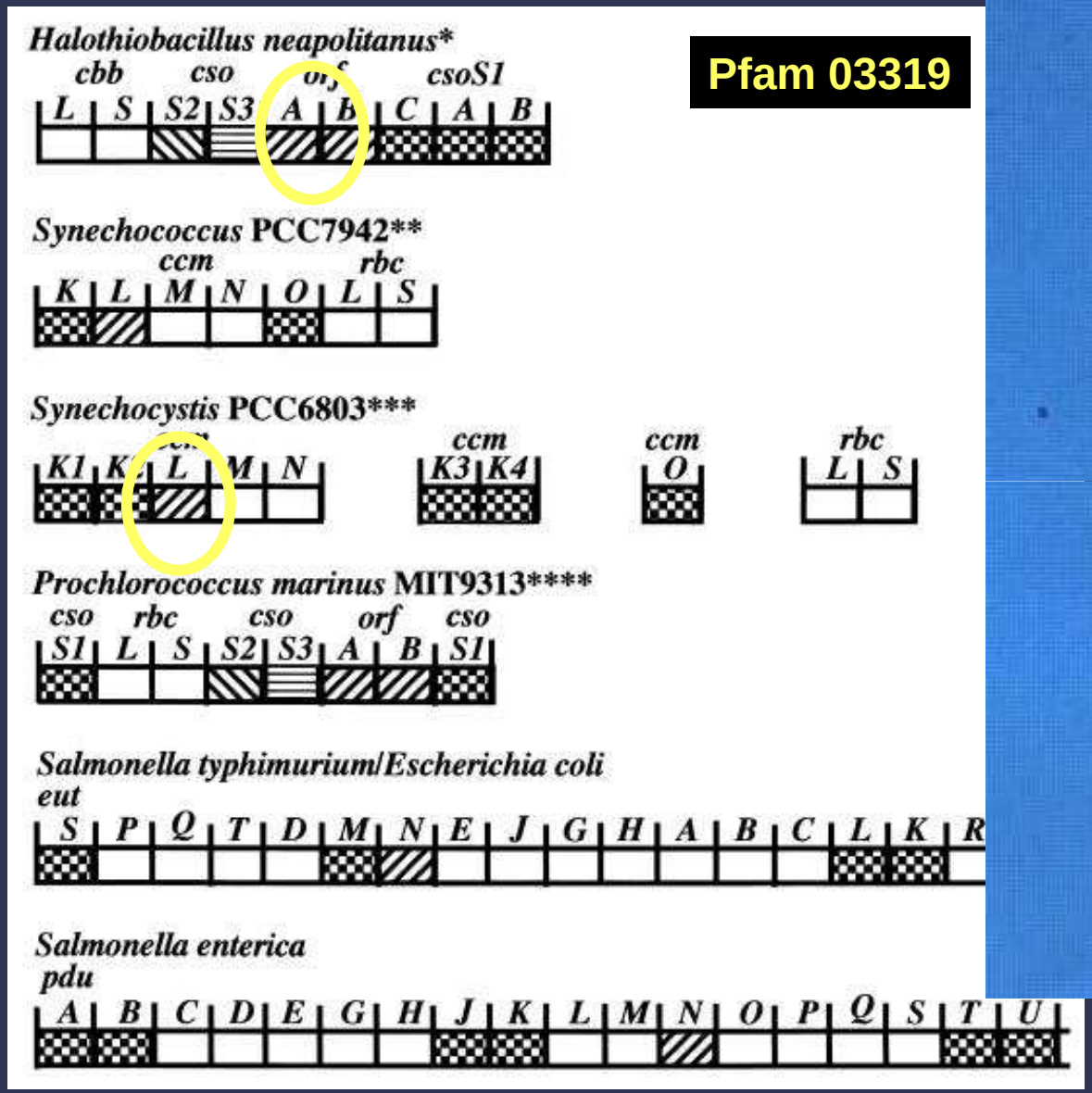




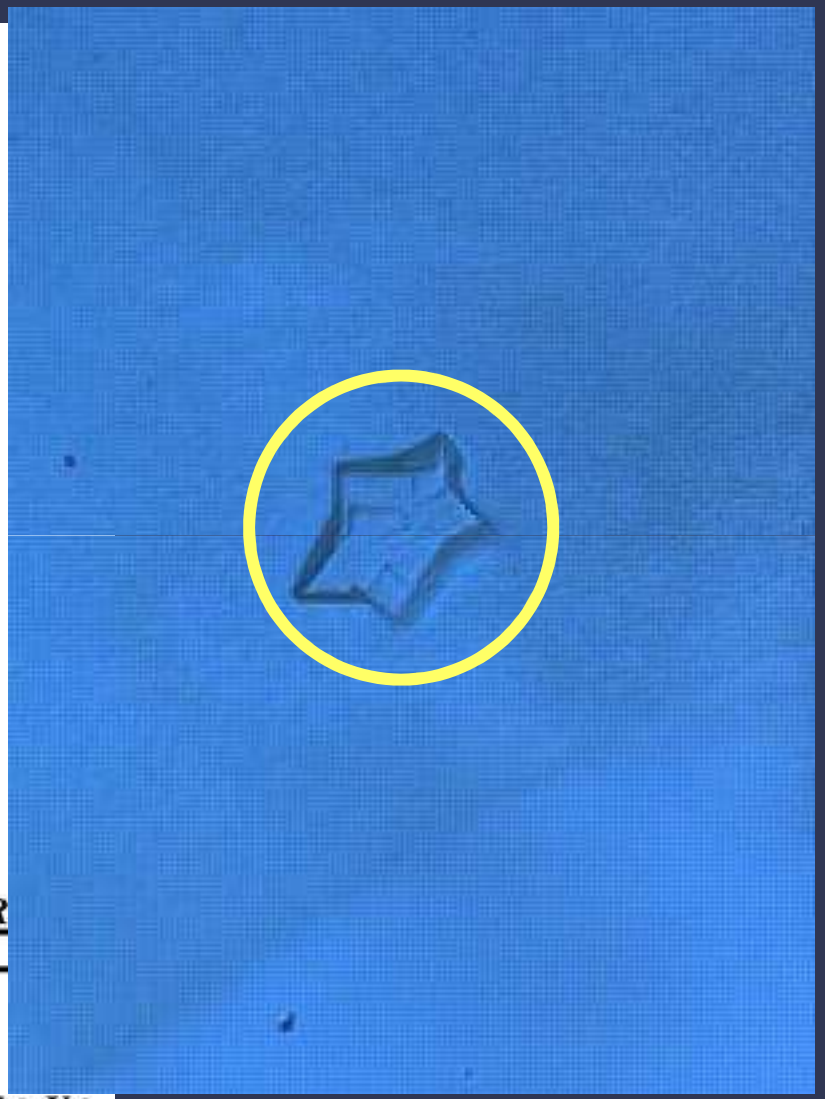


And two tomography studies……

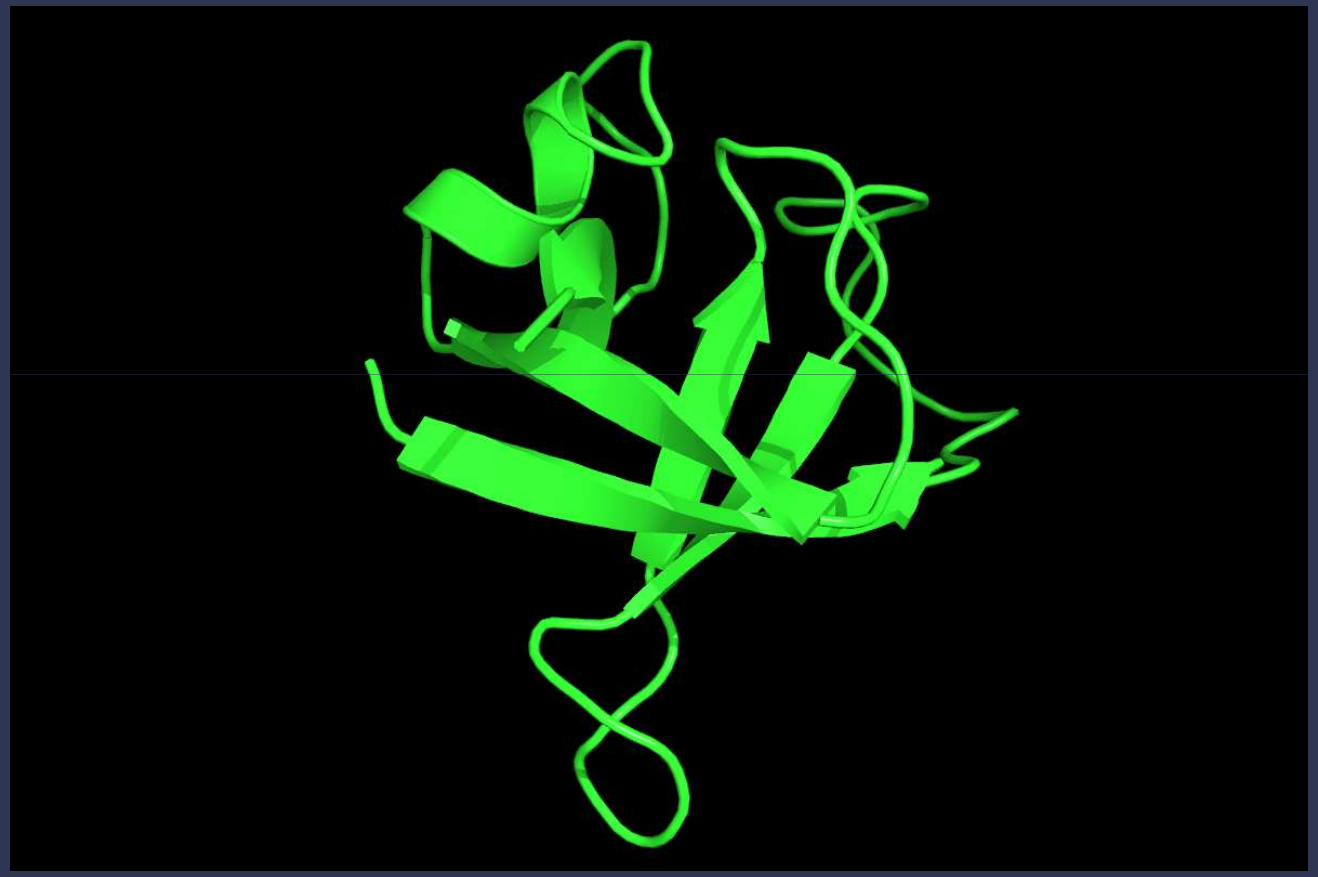
Pentameric Defects?



Pfam 03319

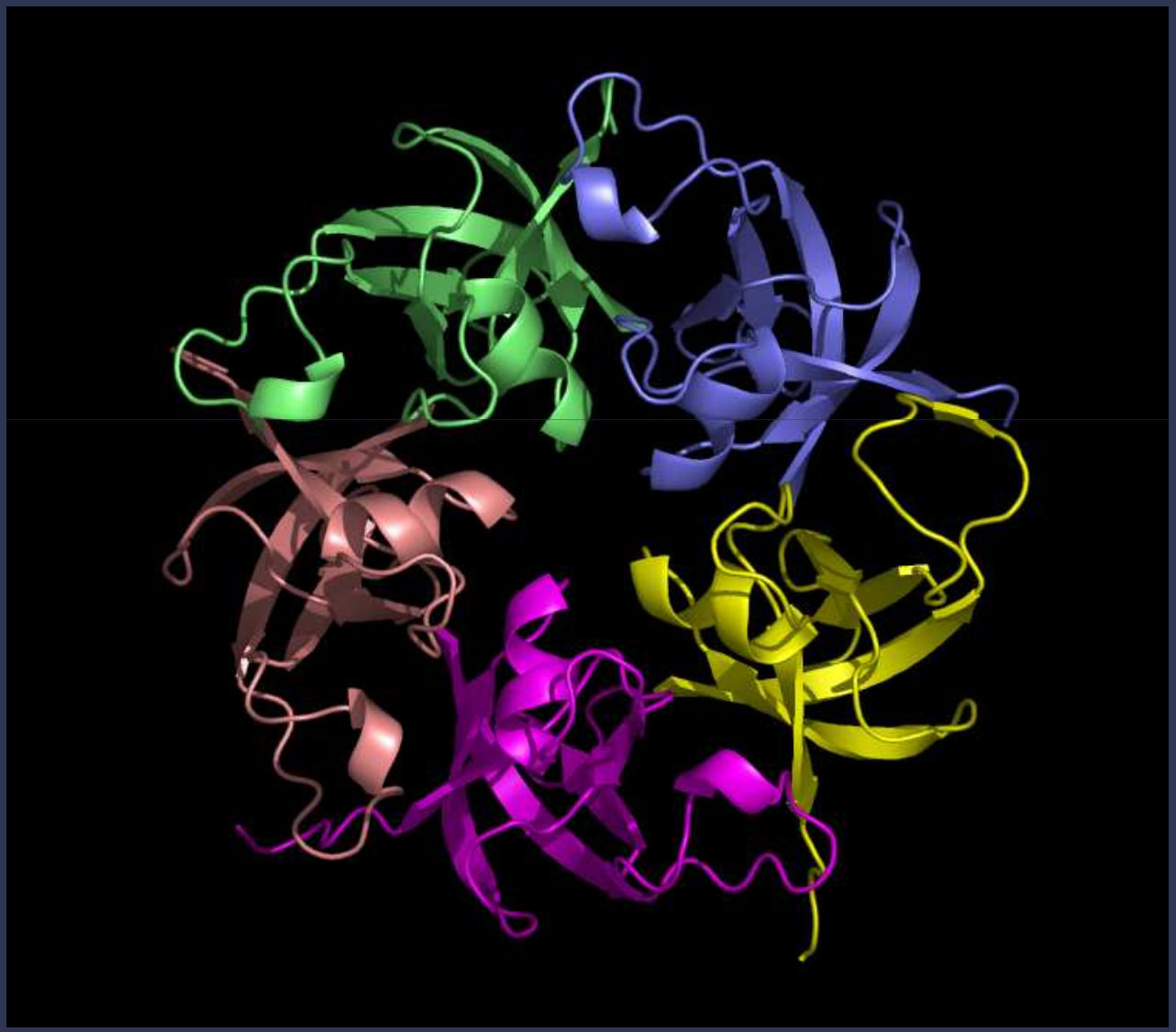


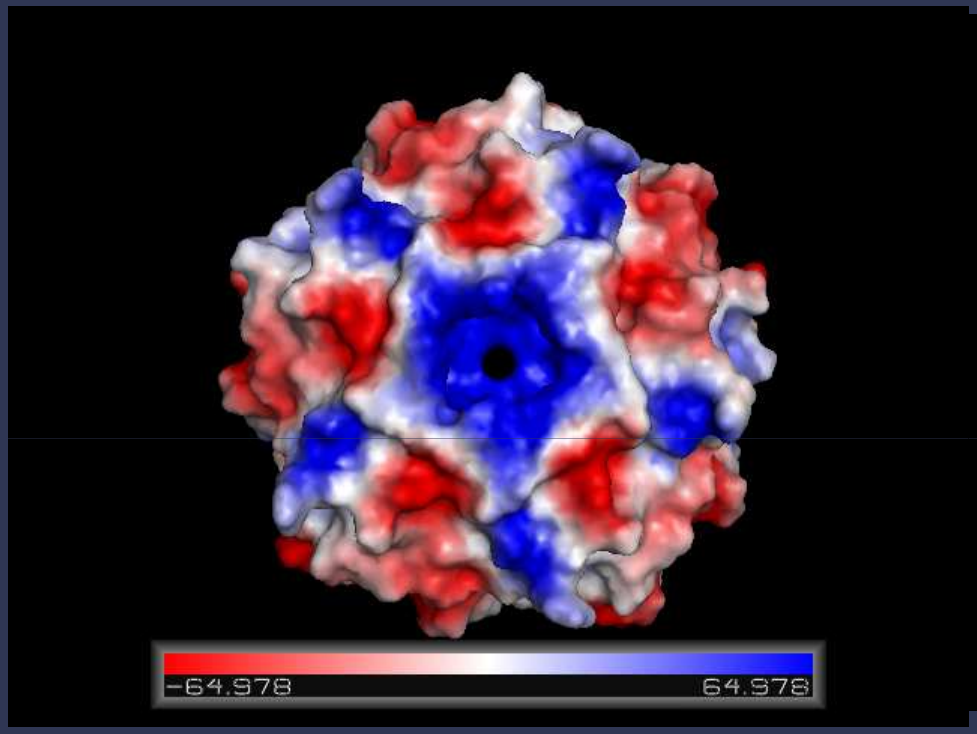
OrfA structure

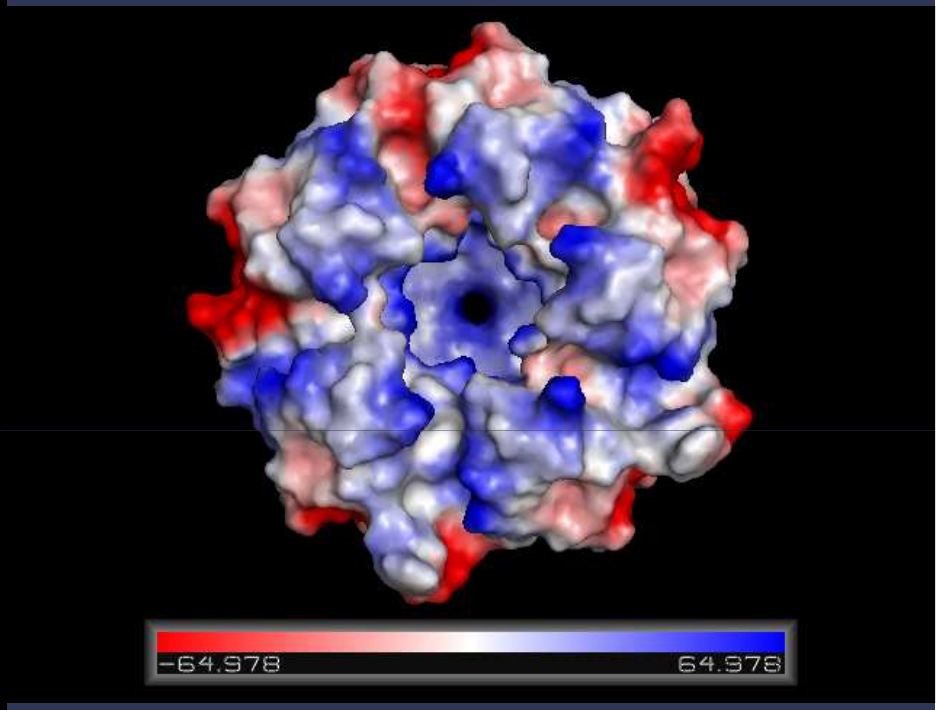


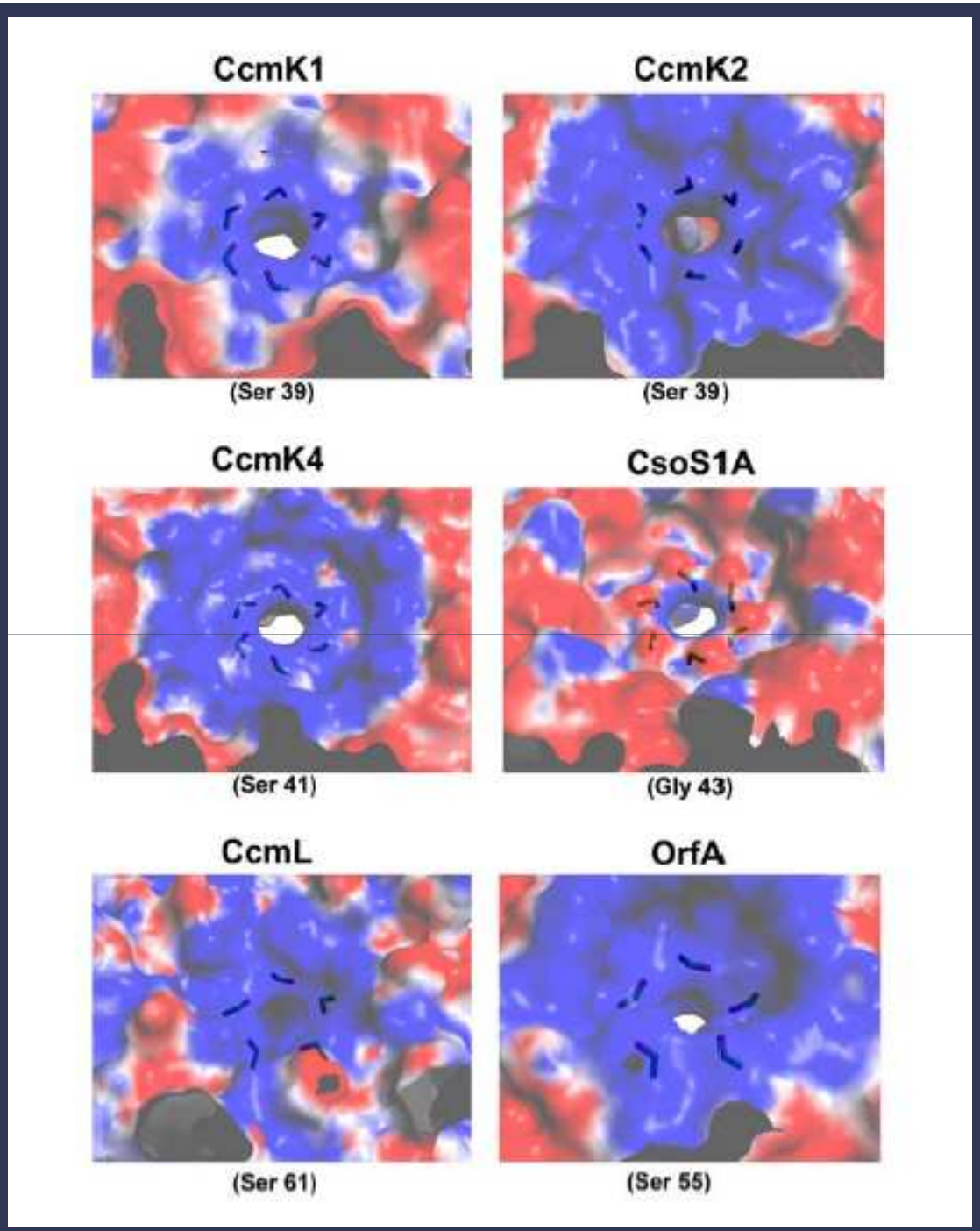
Tanaka et al. Science 2008

OrfA structure



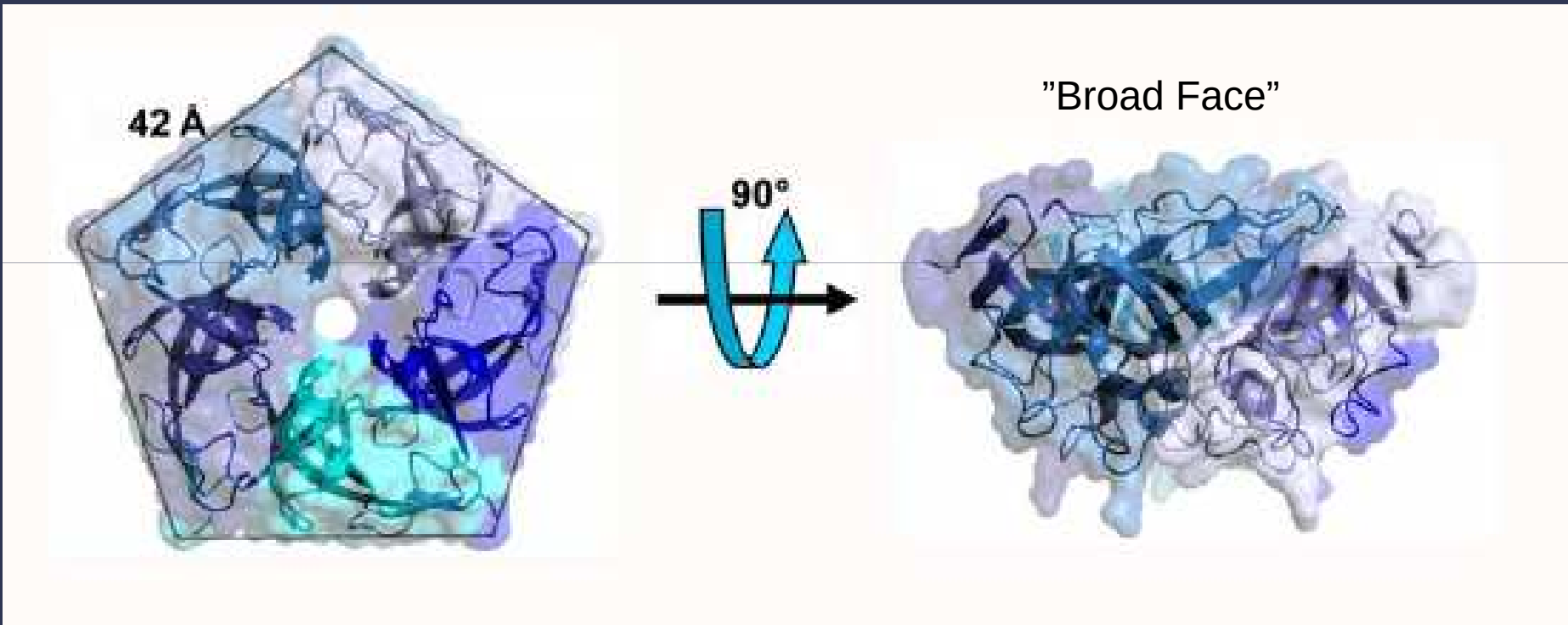

-64.978
64.978


-64.978
64.978



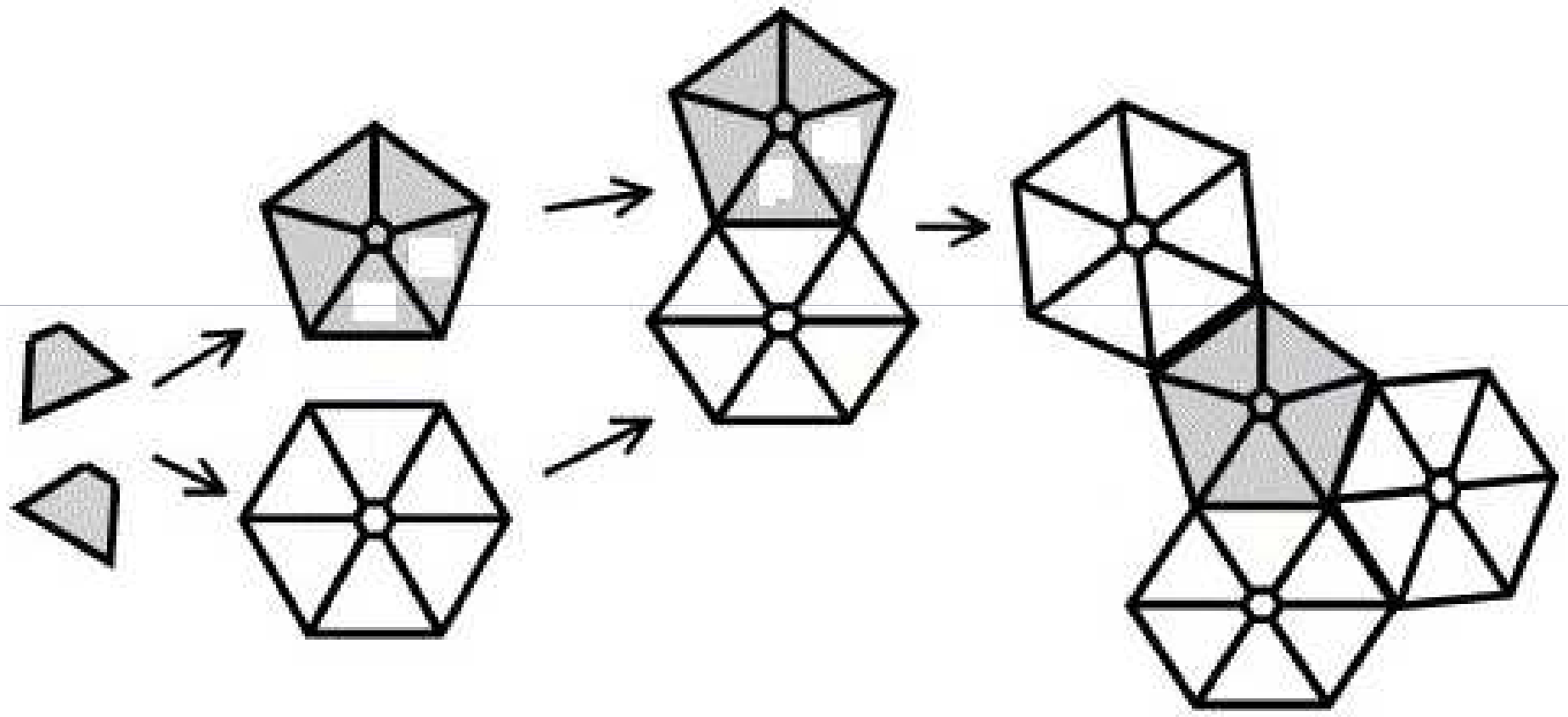
Tanaka et al. Science 2008

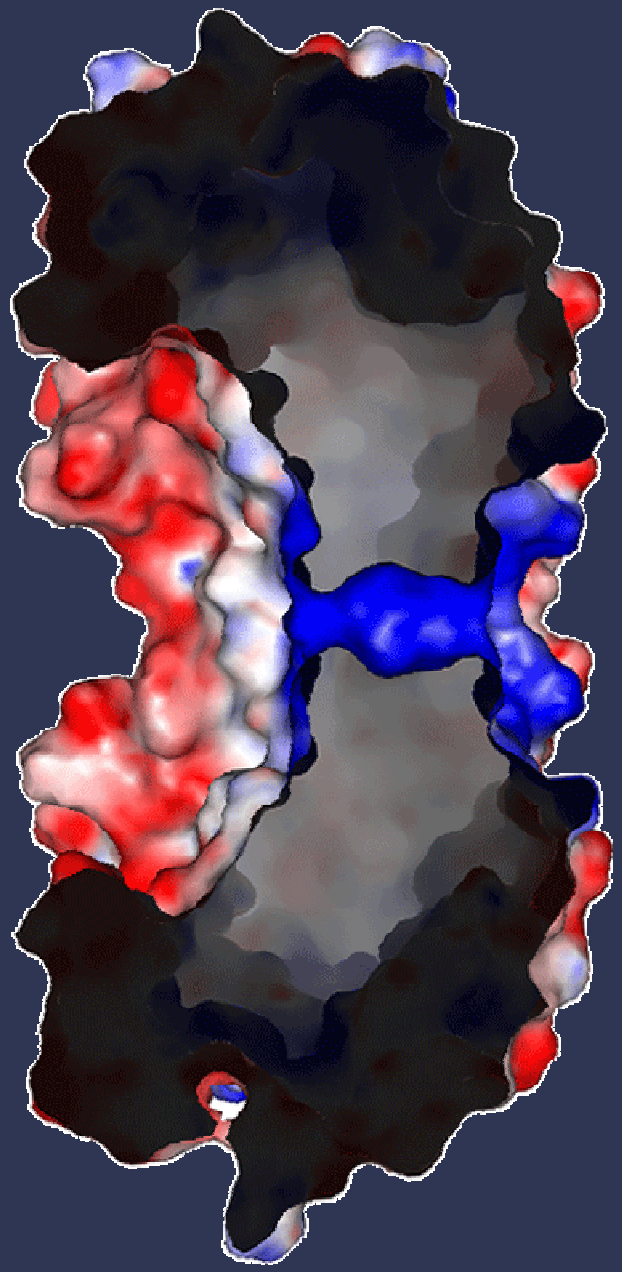
The Pentamer is a Truncated Pyramid

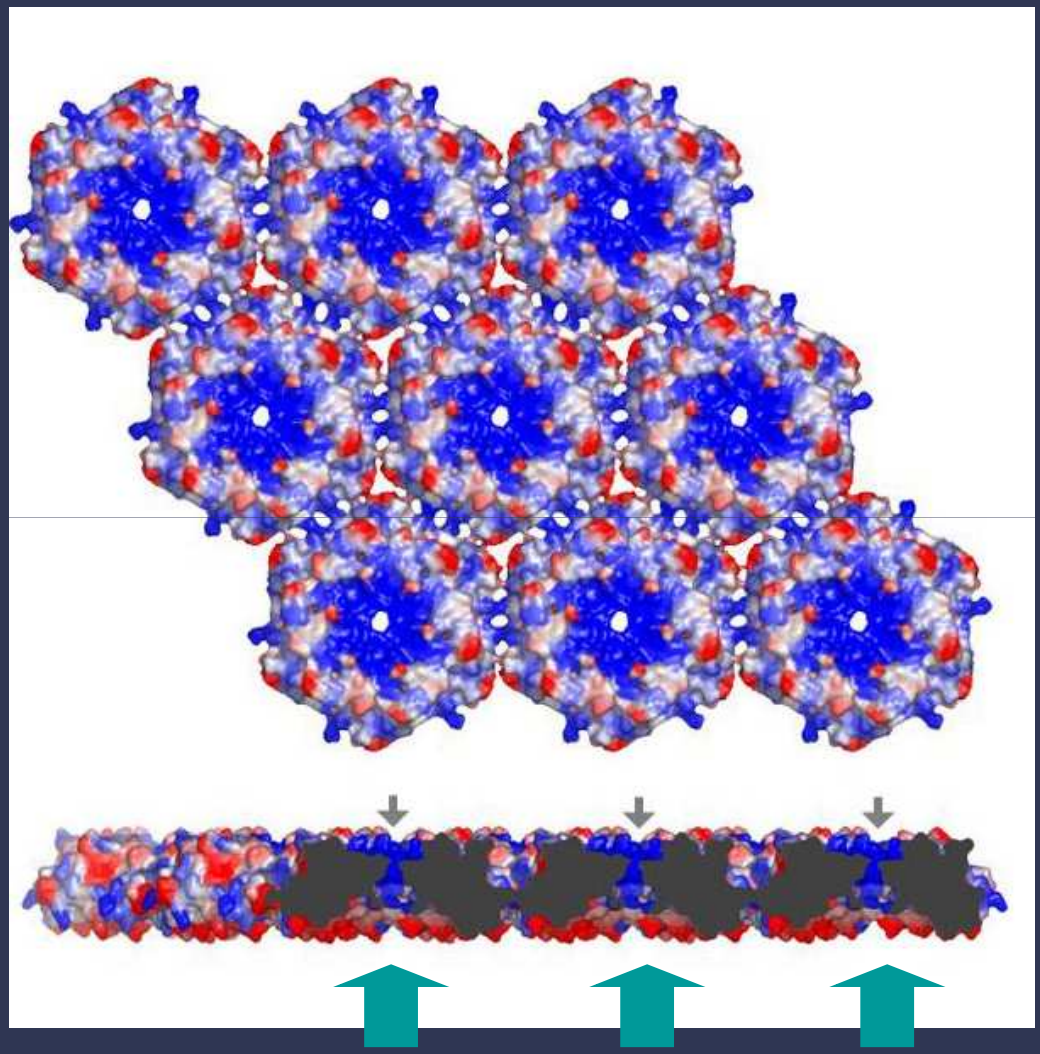


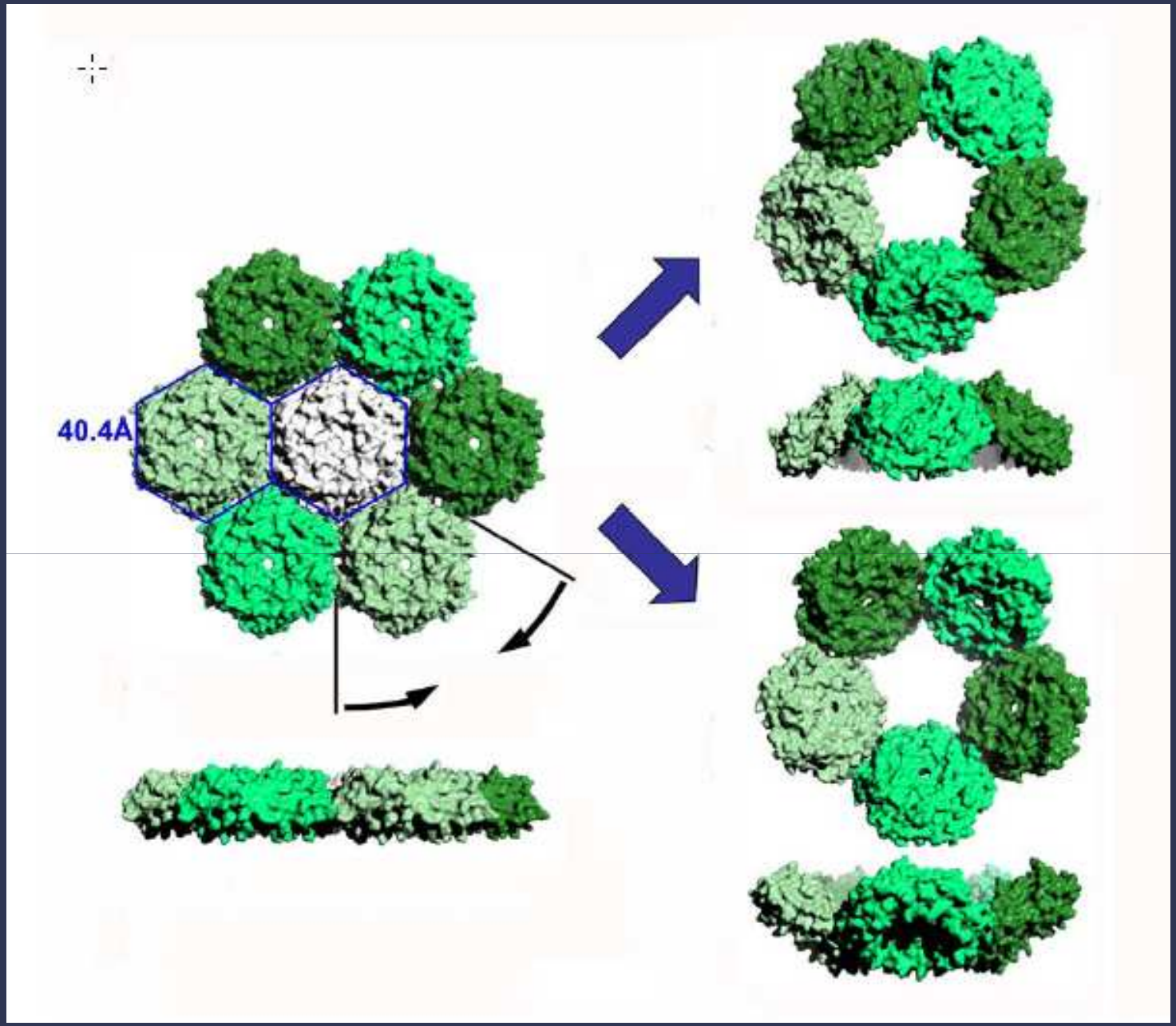
Tanaka et al. Science 2008

Assembly of an Icosahdron

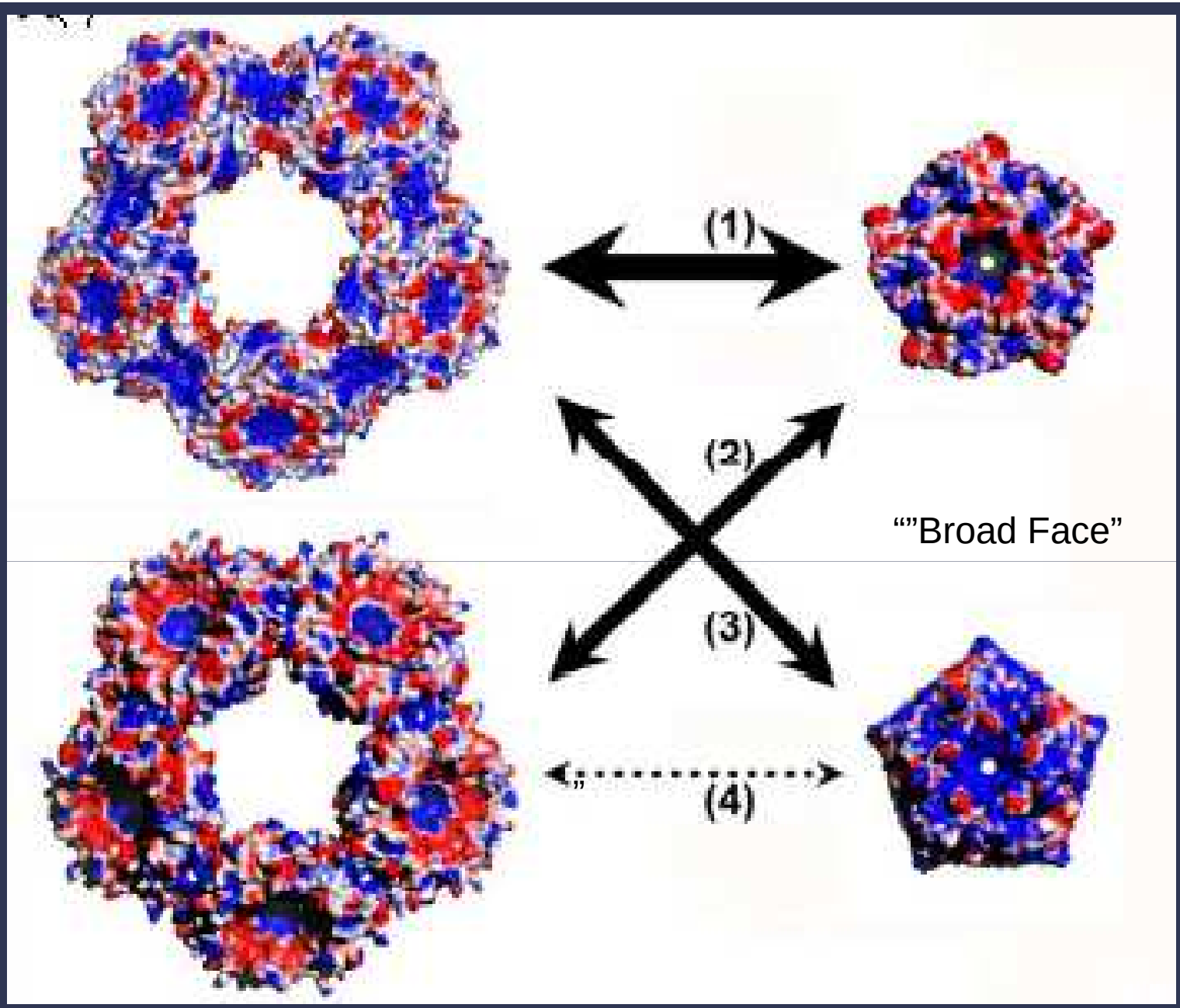




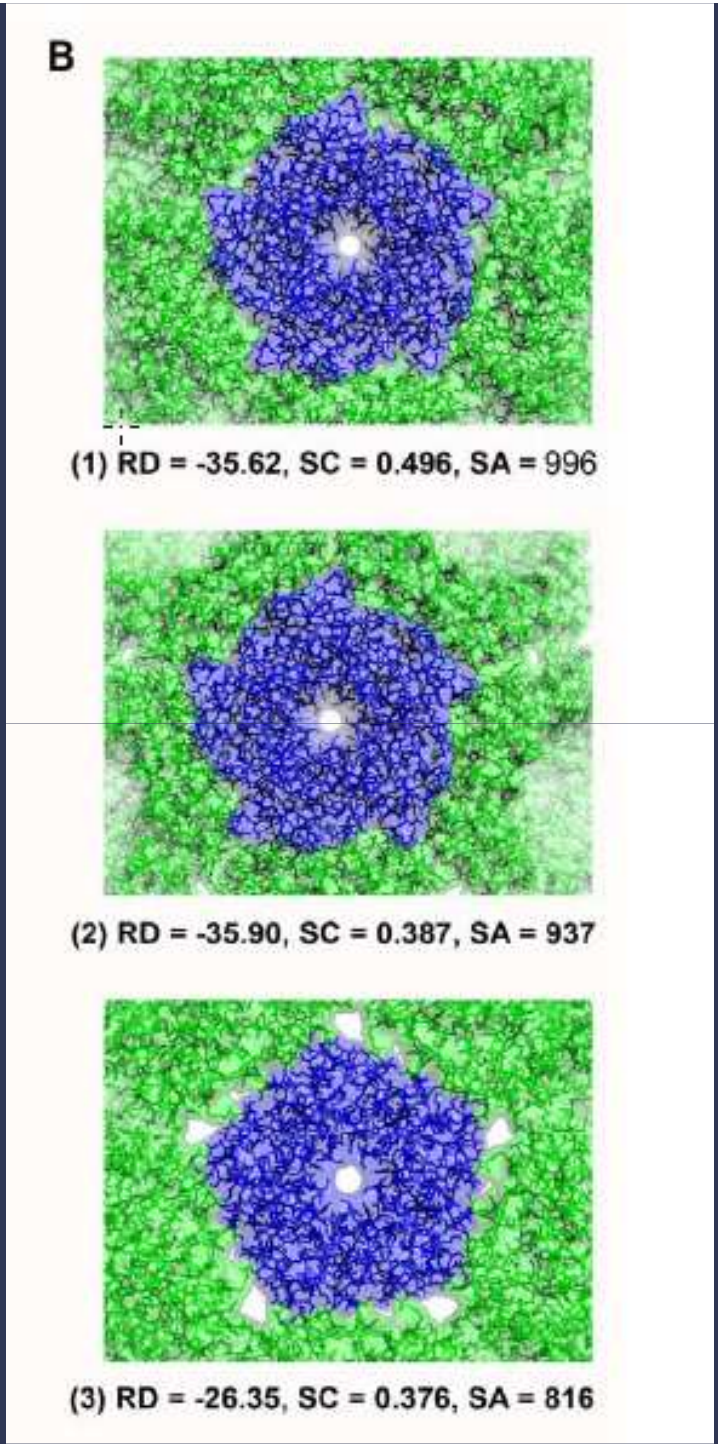




Tanaka et al. Science 2008

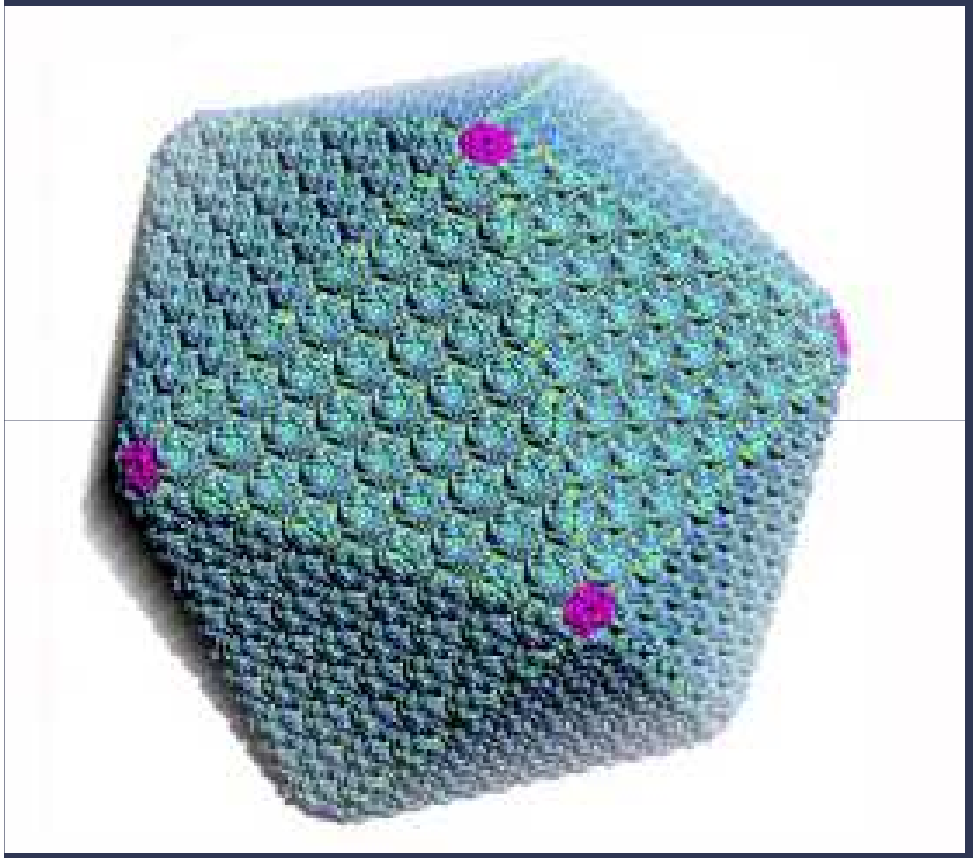


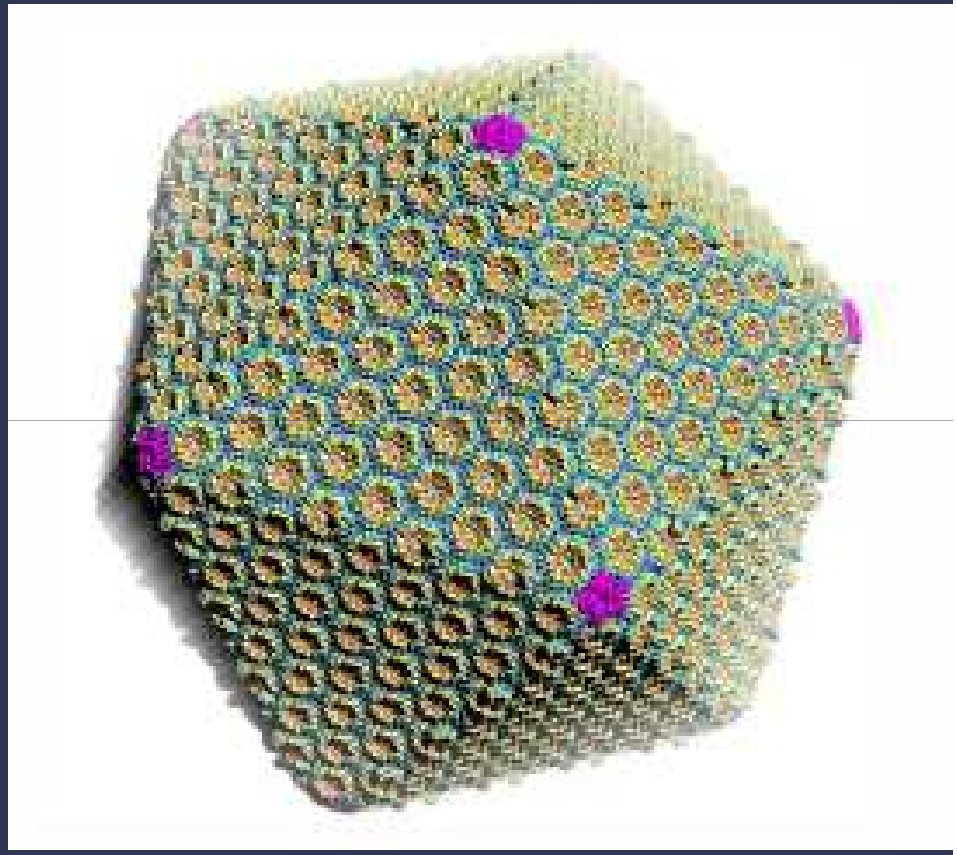
Tanaka et al. Science 2008



Tanaka et al. Science 2008

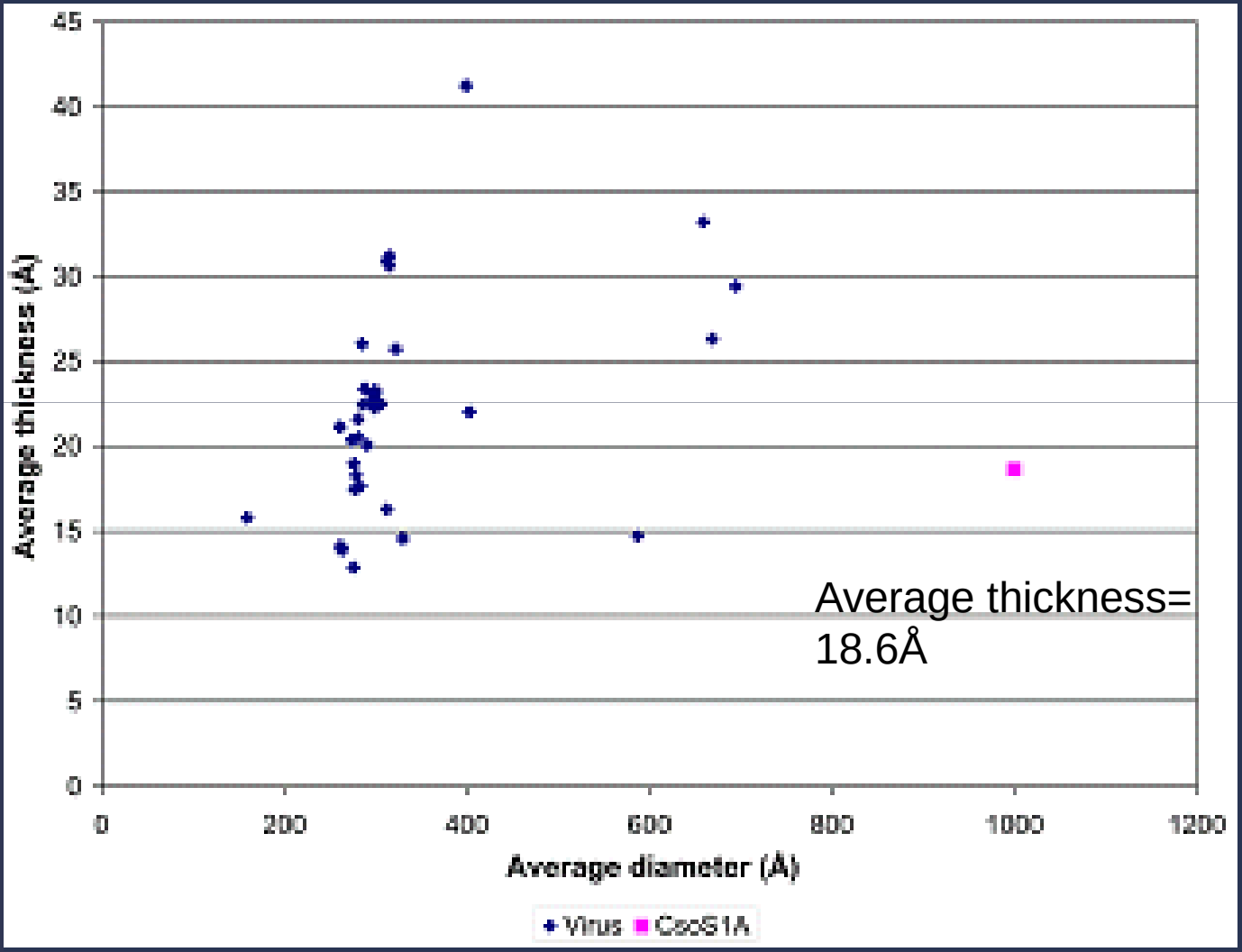
T=75 Models of the Carboxysome





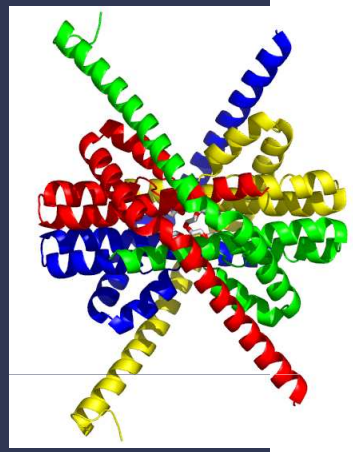
Tanaka et al. Science 2008

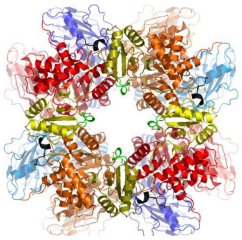
The carboxysome shell is relatively thin compared to the shells of viruses



Tsai et al., PLoS Biology, June 07

Next Steps…..





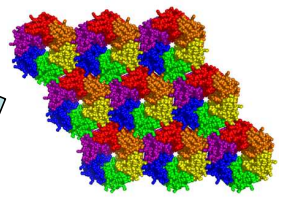
RuBisCO

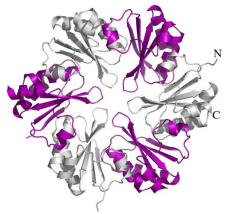
? CcmM ? CcmN

RbcX

~1200 Å

CcmL /(OrfA)



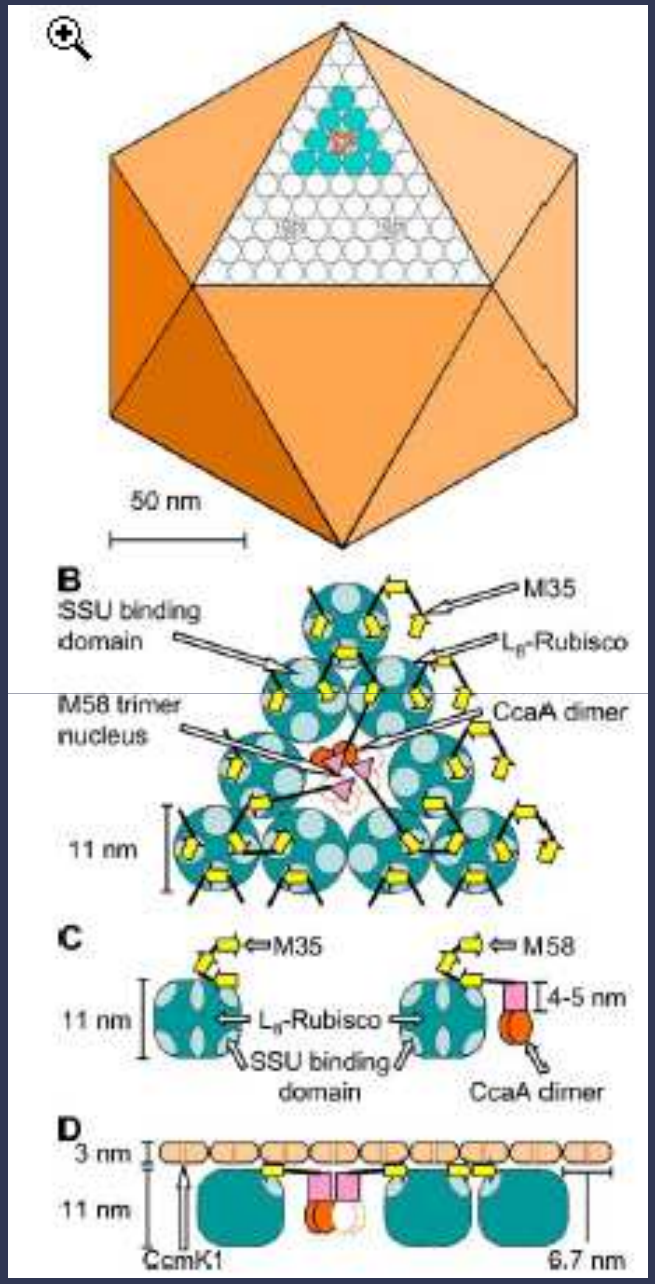


CcmK2,4

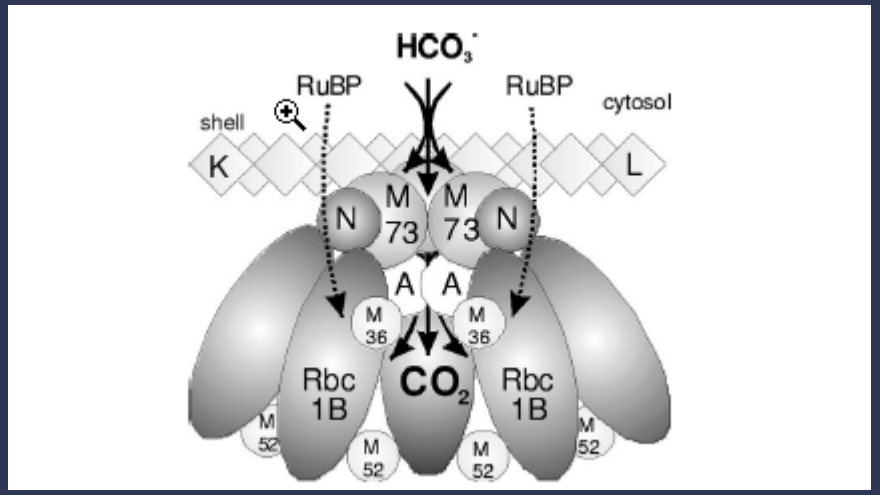
The Carboxysome:

Nature's CO_2-fixing machine

Yeates et al., Biochem. Soc. Trans. 2007



Long et al., JBC 2007



Cot et al., J. Bact 2007

JBC Papers in Press. Published on March 10, 2008 as Manuscript M709214200
The latest version is at http://www.jbc.org/cgi/doi/10.1074/jbc.M709214200

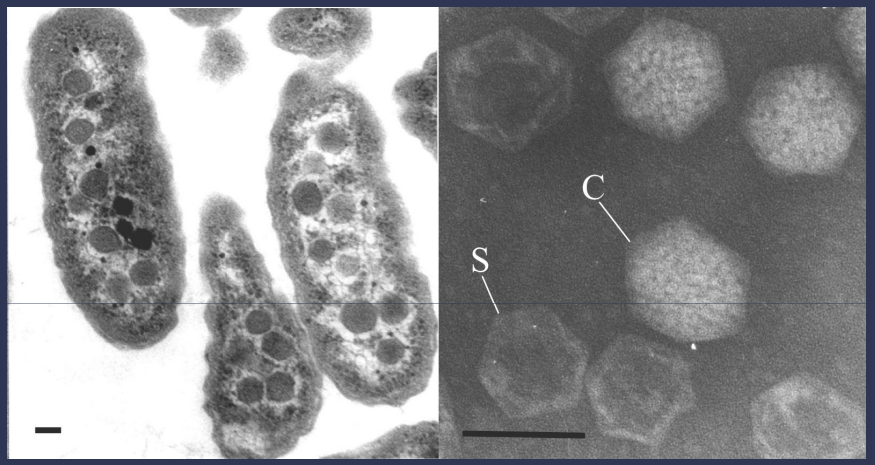
BIOCHEMICAL AND STRUCTURAL INSIGHTS INTO BACTERIAL ORGANELLE FORM AND BIOGENESIS.

Joshua P Parsons [1]*, Sriramulu D Dinesh[2]*, Evelyne Deery,[1] Helen K Leech[1], Amanda A Brindley[1], Dana Heldt[1], Steffi Frank[1], C. Mark Smales[1], Heinrich Lünsdorf[3], Alain Rambach[4], Mhairi H Gass[5], Andrew Bleloch,[5] Kirsty J McClean[6], Andrew W Munro[6], Stephen E J Rigby[7], Martin J Warren[1] and Michael B Prentice[2]

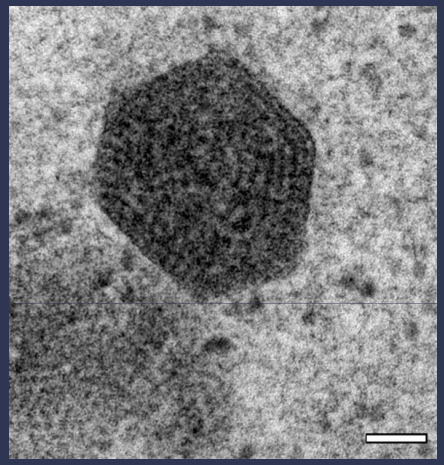
1. Protein Science Group, Department of Biochemistry, University of Kent, Canterbury, Kent CT2 7NJ, UK
2. Departments of Pathology and Microbiology, University College Cork, Ireland
3. Department of Vaccinology, Helmholtz Center of Infection Research, Braunschweig, Germany
4. CHROMagar, 4 place du 18 Juin 1940, F-75006 Paris, France
5. SuperSTEM facility, Daresbury Laboratories, Daresbury, UK
6. Faculty of Life Sciences, Manchester Interdisciplinary Biocentre, University of Manchester, 131 Princess Street, Manchester M1 7DN
7. School of Biological and Chemical Sciences, Queen Mary, University of London, Mile End Road, London E1 4NS, UK

Other Bacterial Microcompartments

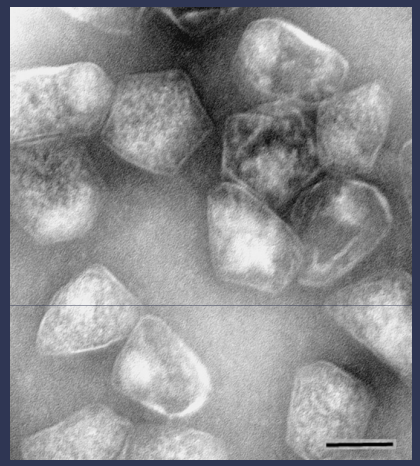
A



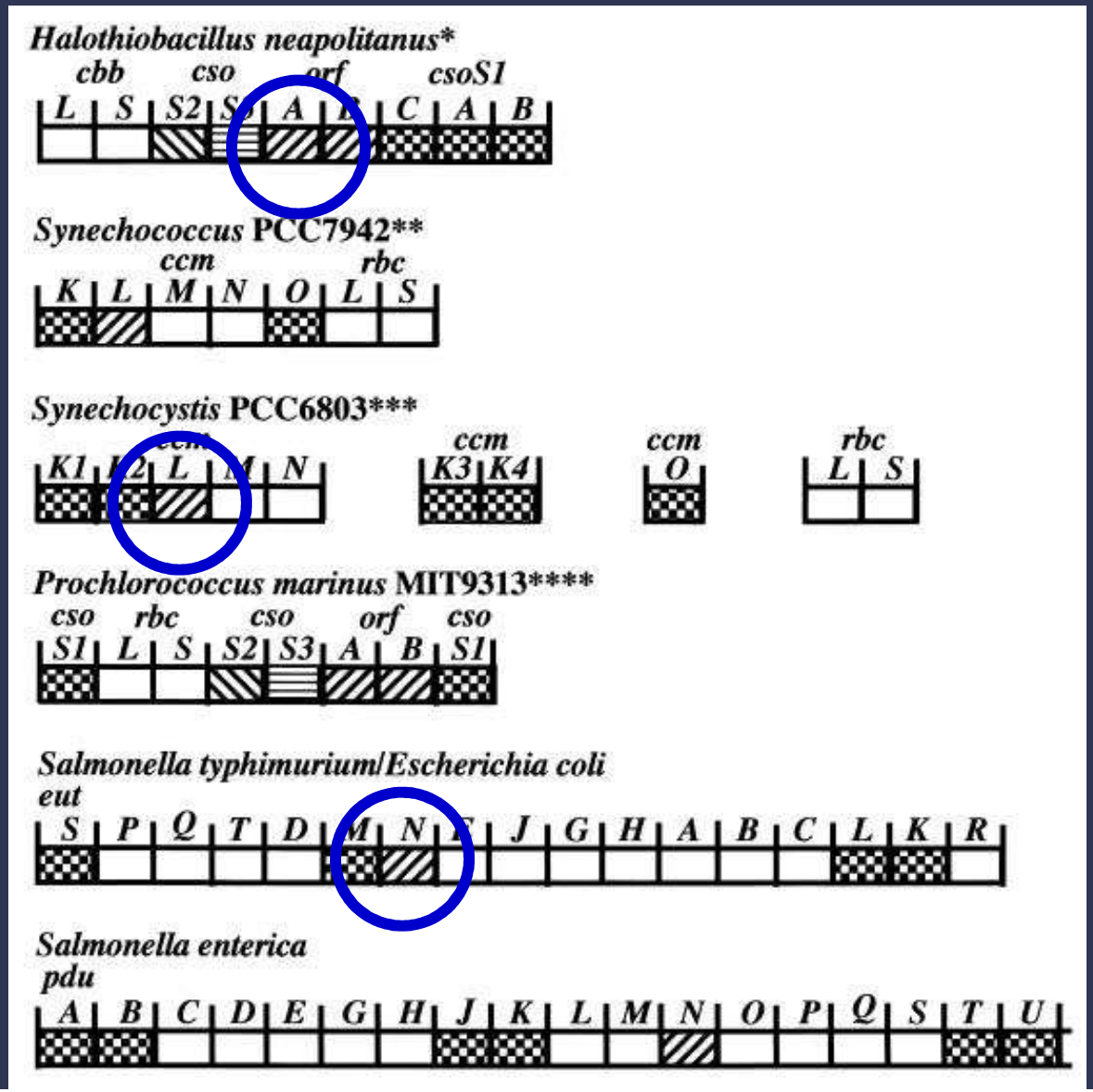
B

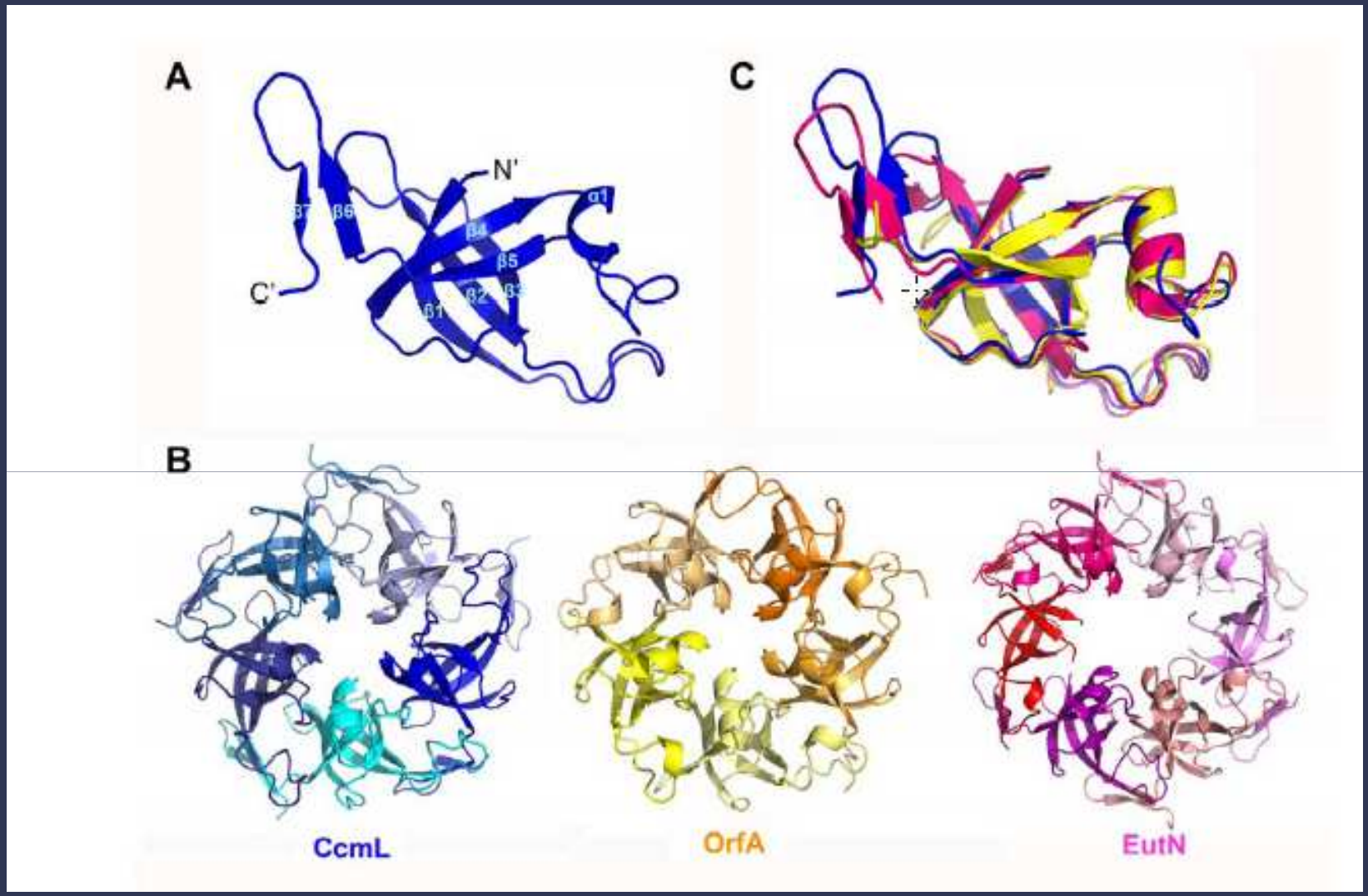


C



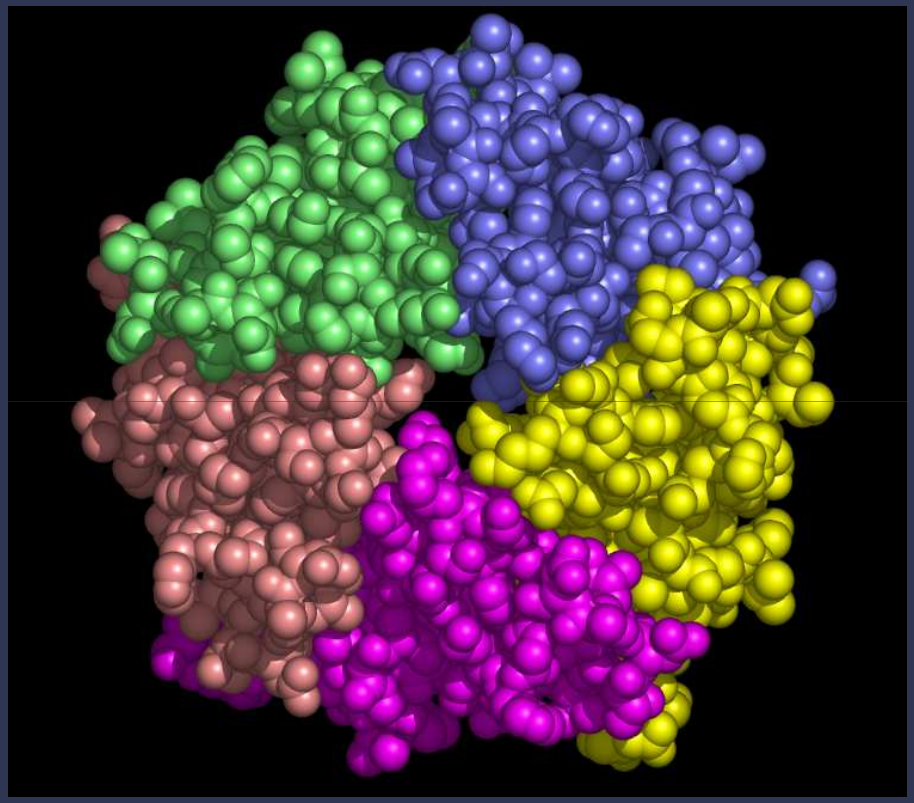
Other Bacterial Microcompartments



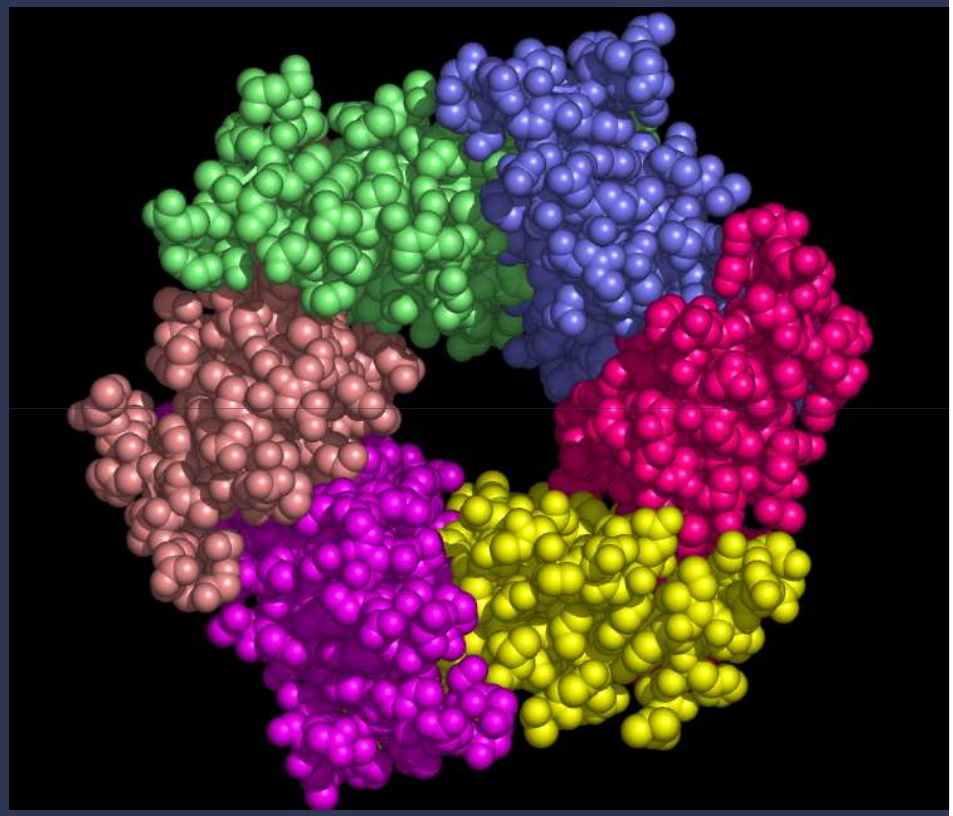


Tanaka et al. Science 2008

OrfA/CcmL



EutN



And other unusual shell protein building blocks forthcoming….

Summary

- We have sufficient structural data to model the carboxysome shell
- Bacterial microcompartments are surprisingly widespread and diverse in function
- And likely to be architecturally diverse in their building blocks

Opportunities…..

- Cyanobacterial Annotation Tools for Undergraduates (DOE)/Database needs
- Cyanobacterial Molecular Biology Workshop, June 2010 Lake Delevan, Wisconsin

Acknowledgements

- **UCLA**

Todd Yeates' group

- **USM**

Sabine Heinhorst &
Gordon Cannon's group

- **JGI and UC Berkeley**

Natalia Ivanova
Kostas Mavramommatis
Edwin Kim
Michael Klein
Jay Kinney

FUNDING
USDA and DOE