

# Molecular Modeling 2017 -- lecture 10

Quaternary and super-quaternary structure

Evaluating Models

same/different versus right/wrong

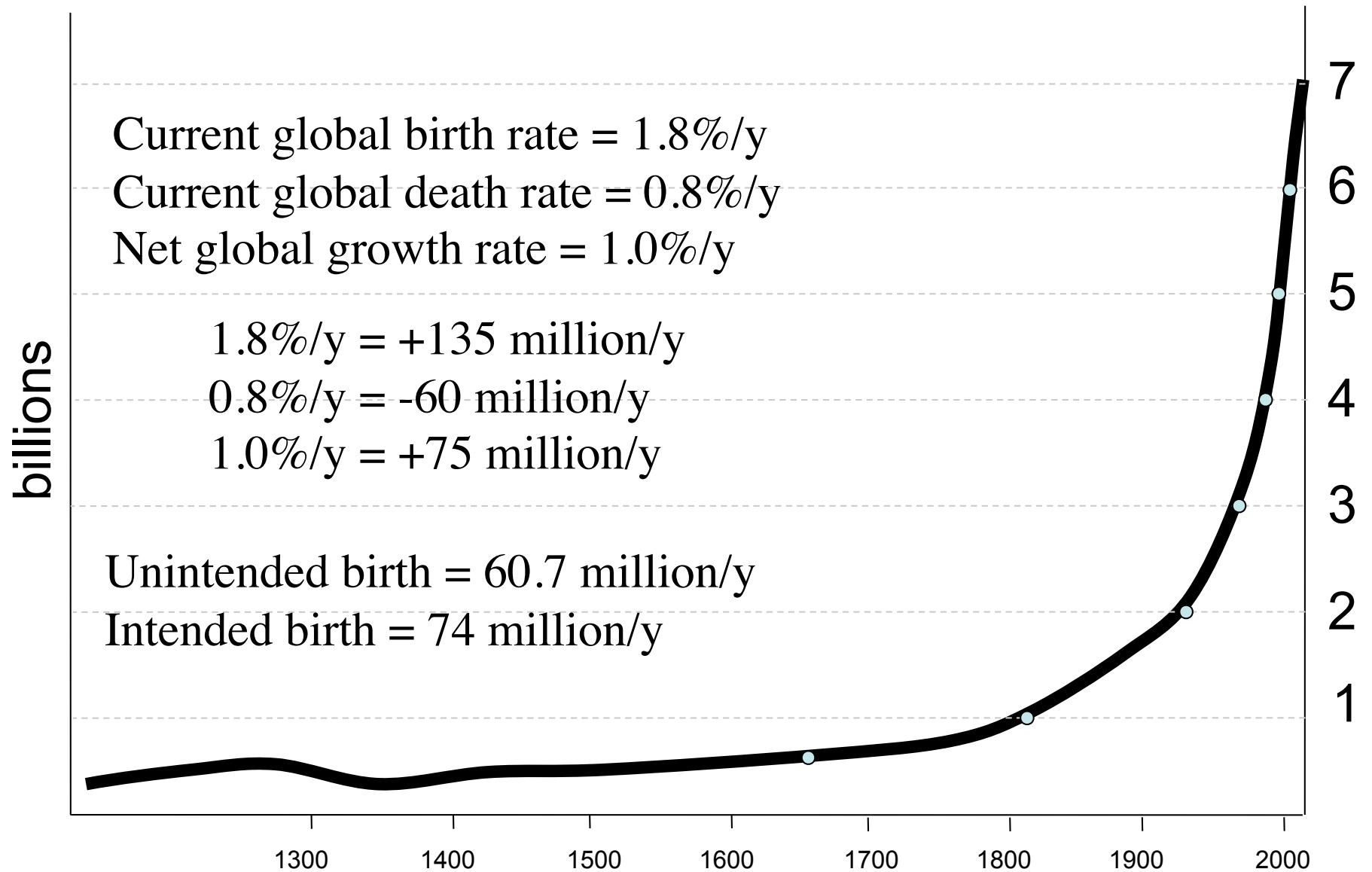
confidence -- 2 types

Fixing errors

# MOVE UPSTREAM

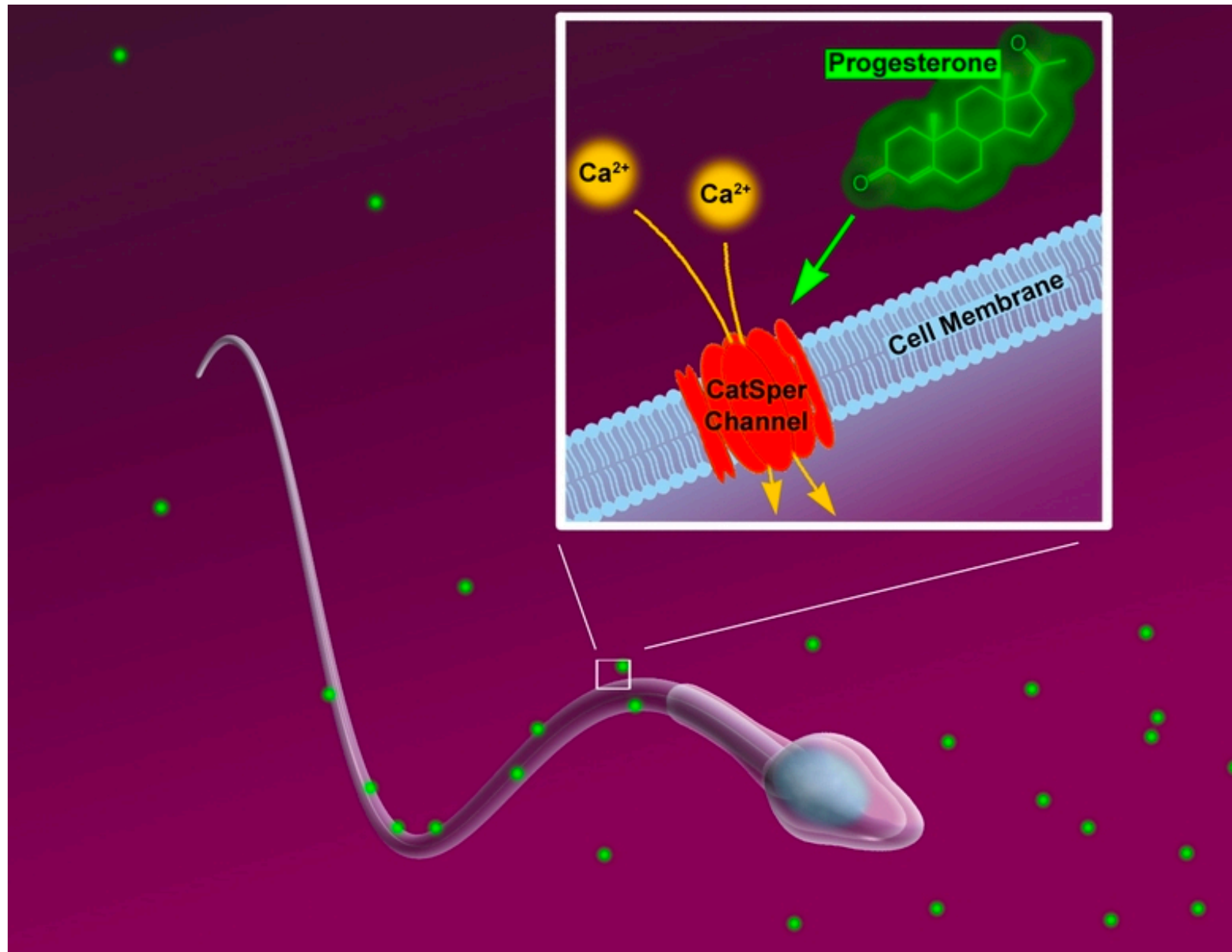
A Call to Solve  
Overpopulation

KAREN I. SHRAGG

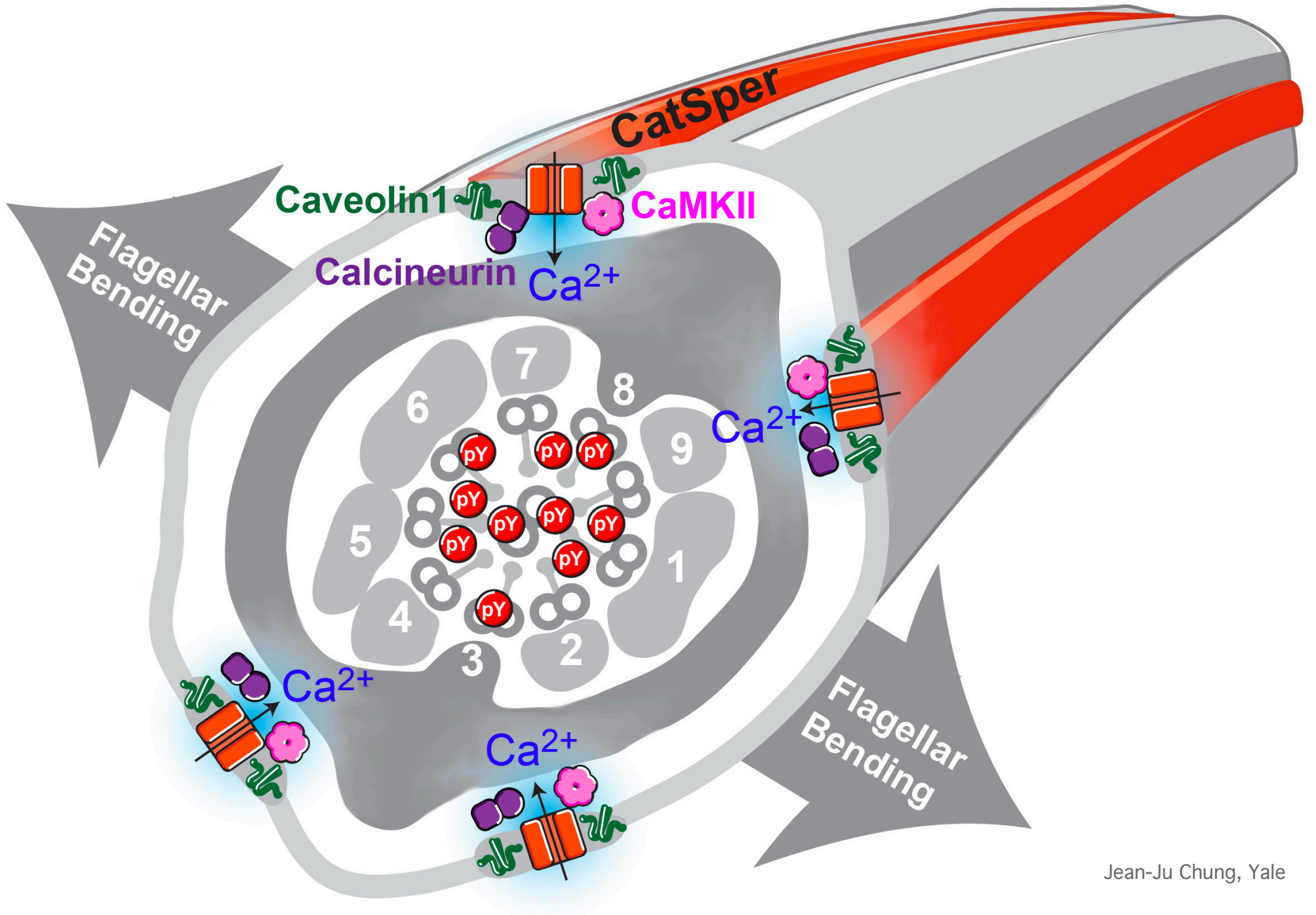


**Global population is growing exponentially!**

The overpopulation problem, up close.







# CatSper

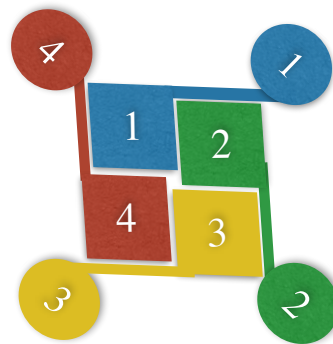
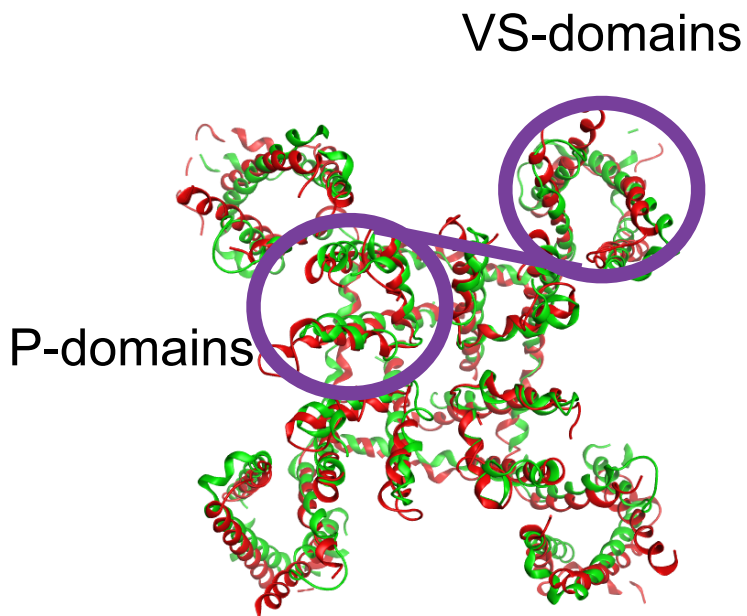
- 4 subunits + 5 associated proteins
- All 9 required for male fertility
- Responsible for sperm hyperactive motility
- Expressed exclusively in sperm
- 6-transmembrane Voltage-gated calcium channel (Cav)
- Heterotetramer
- Not glycosylated
  - > **candidate for contraceptive vaccine**
- Structure not known.

# Homologs of CatSper with known structures

Arcobacter butzleri Cav<sub>1</sub> ← homotetramer

Vertebrate TRPV (capsaicin, cold)

4-fold tandem fusion



heterotetramer has 6 "arrangomers"

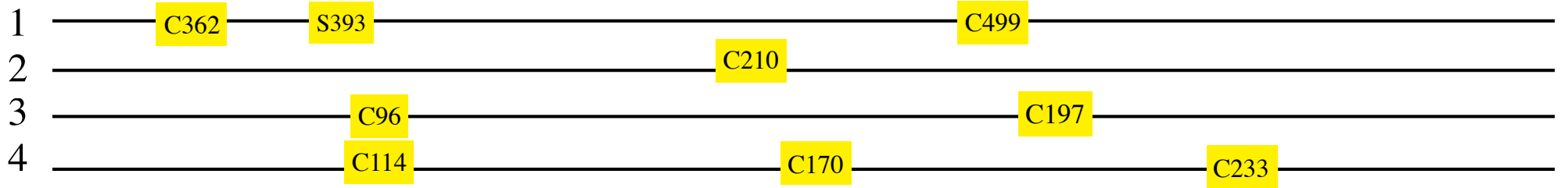
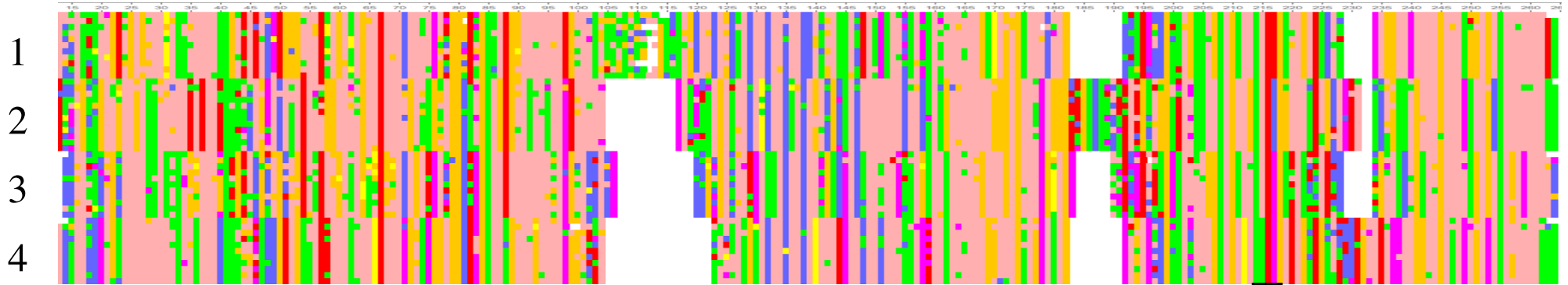


?

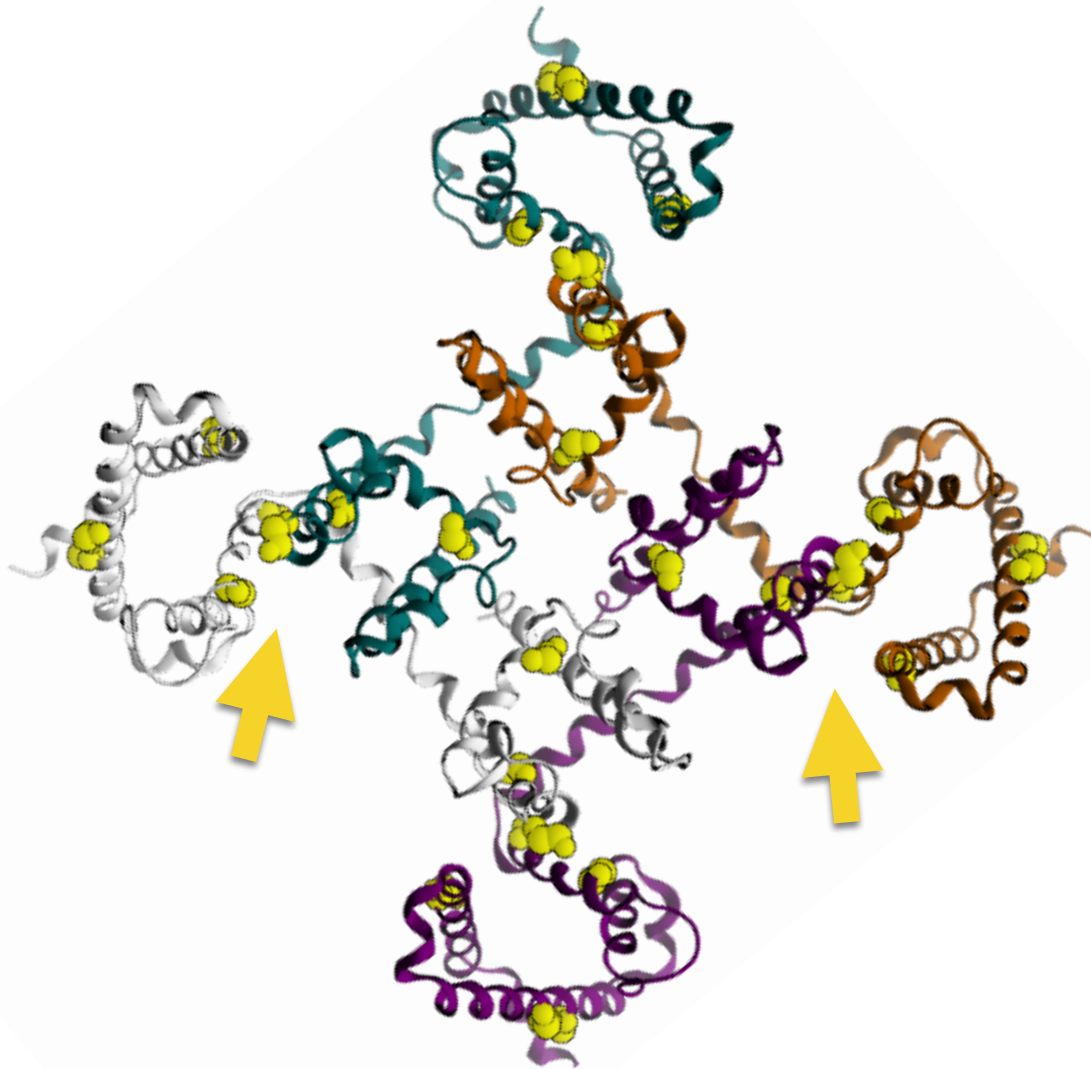
# Conserved cysteines in mammalian CatSper



———— Voltage Sensitive domain ————— Pore domain —————



Cys in all 4 mouse CatSper mapped onto  
onto **acrobacter** monomer -->  
homotetramer

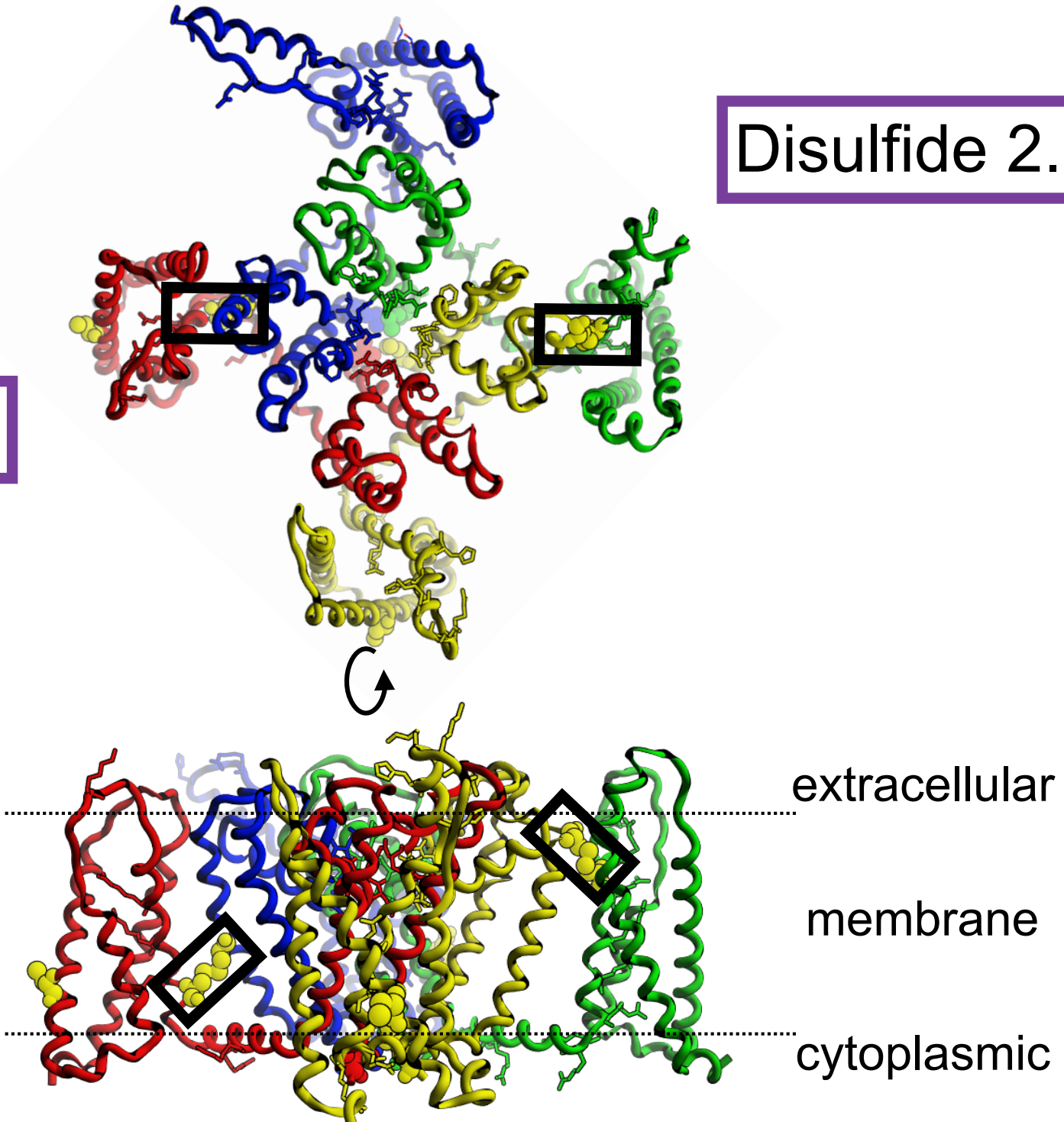


Which conserved Cys  
could be interacting?

Disulfide 1.

Disulfide 2.

Can't cross-pair  
because of  
different depths



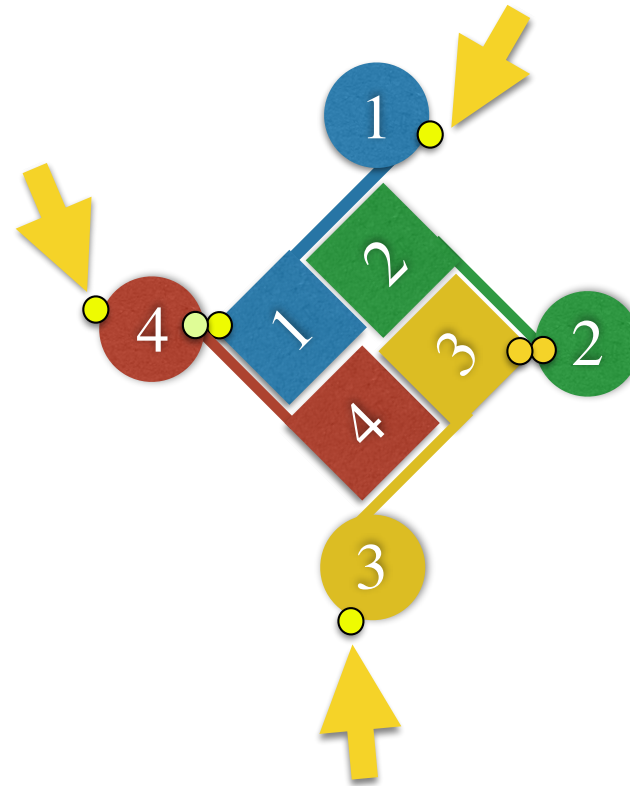
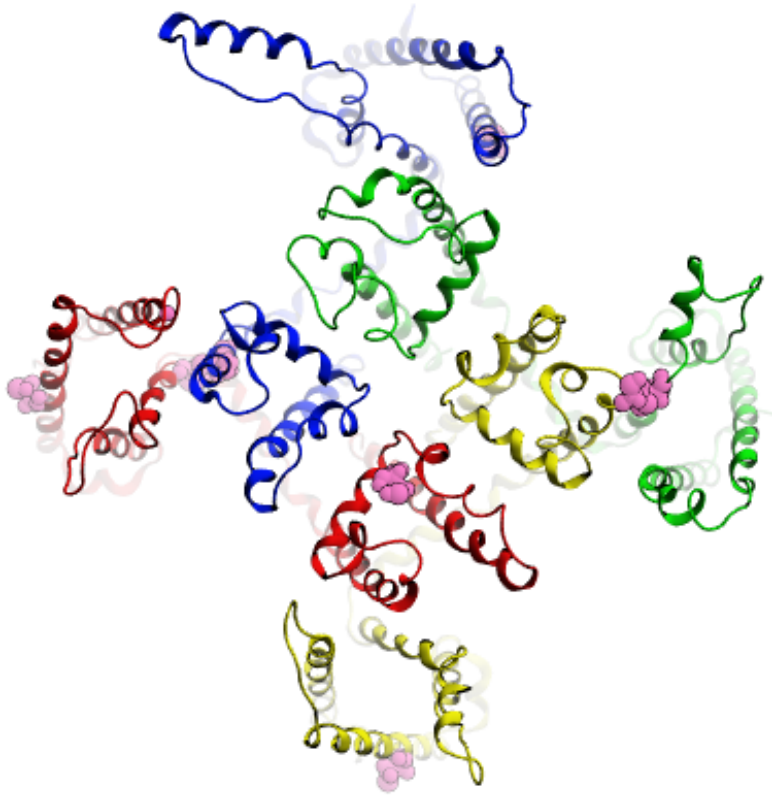
extracellular

membrane

cytoplasmic



# hanging conserved Cys?

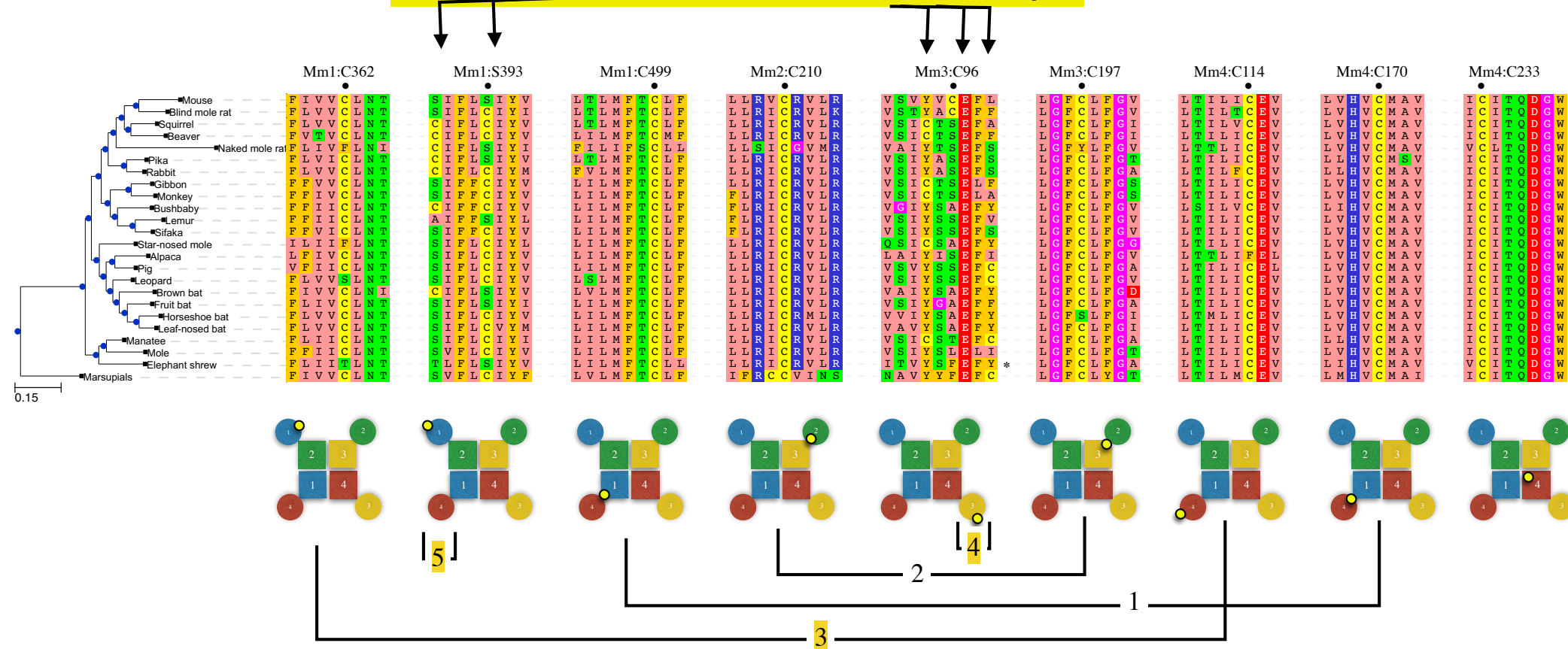


What could they be doing?



# Phylogenetic analysis of mammalian CatSper

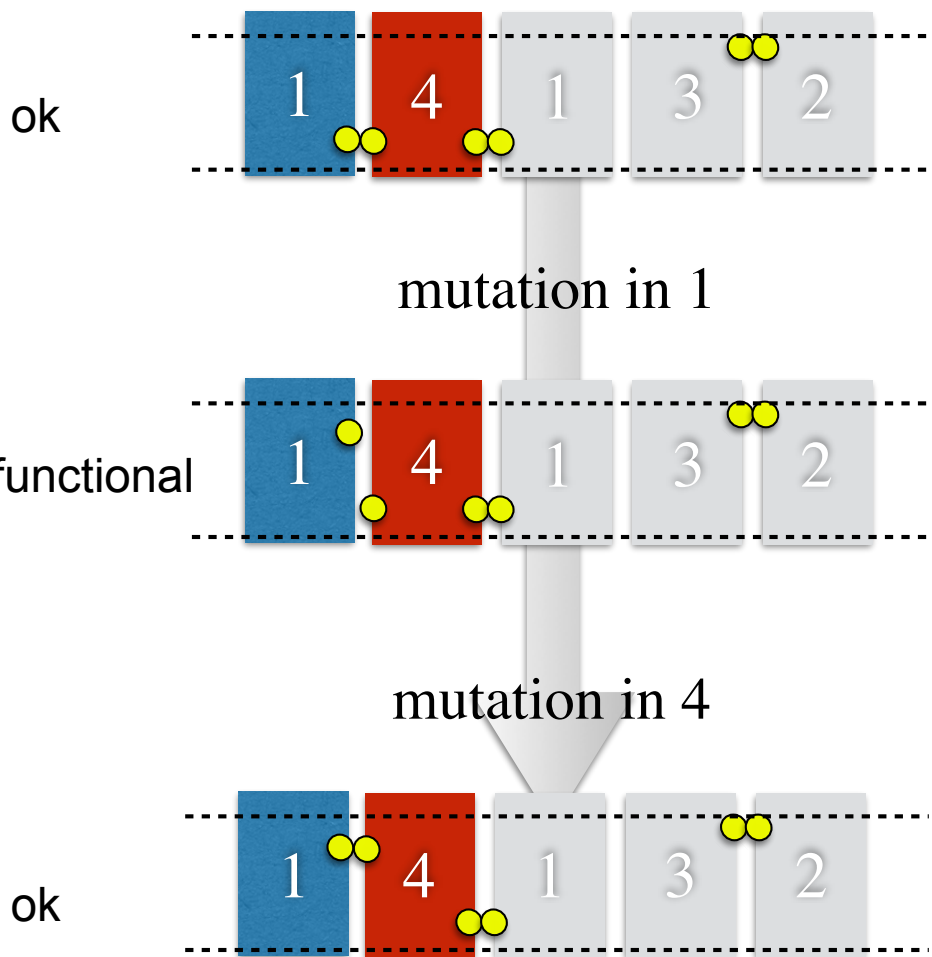
variable position conserved Cys



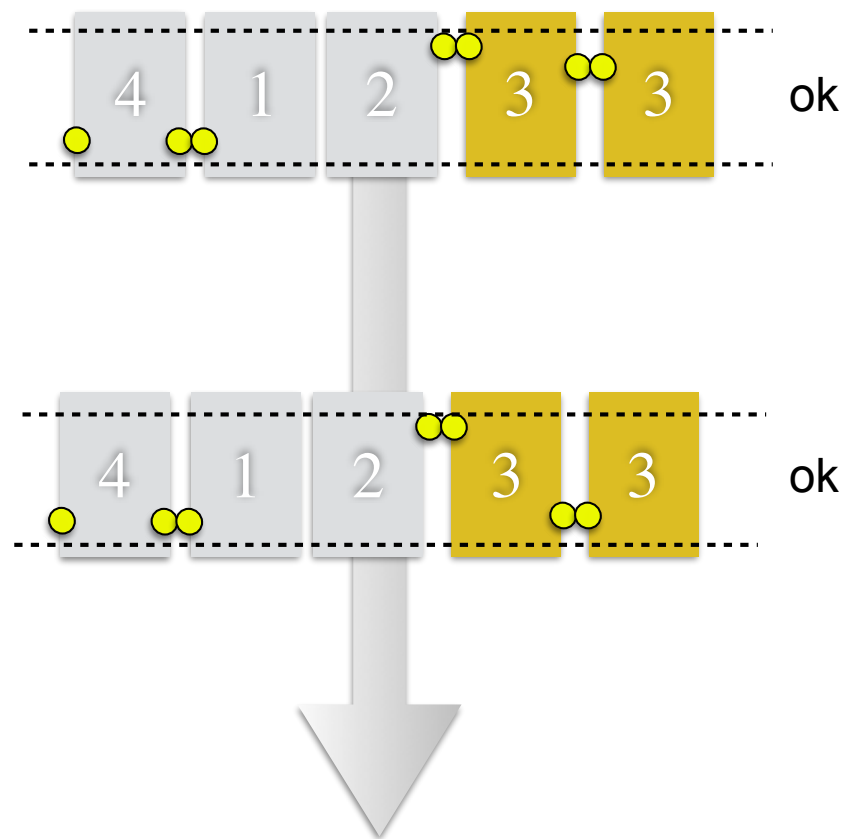
single position conserved Cys

Why are some cysteines moving around and other staying put?

fixed evolutionary position  
is consistent with unlikely  
mutational pathway

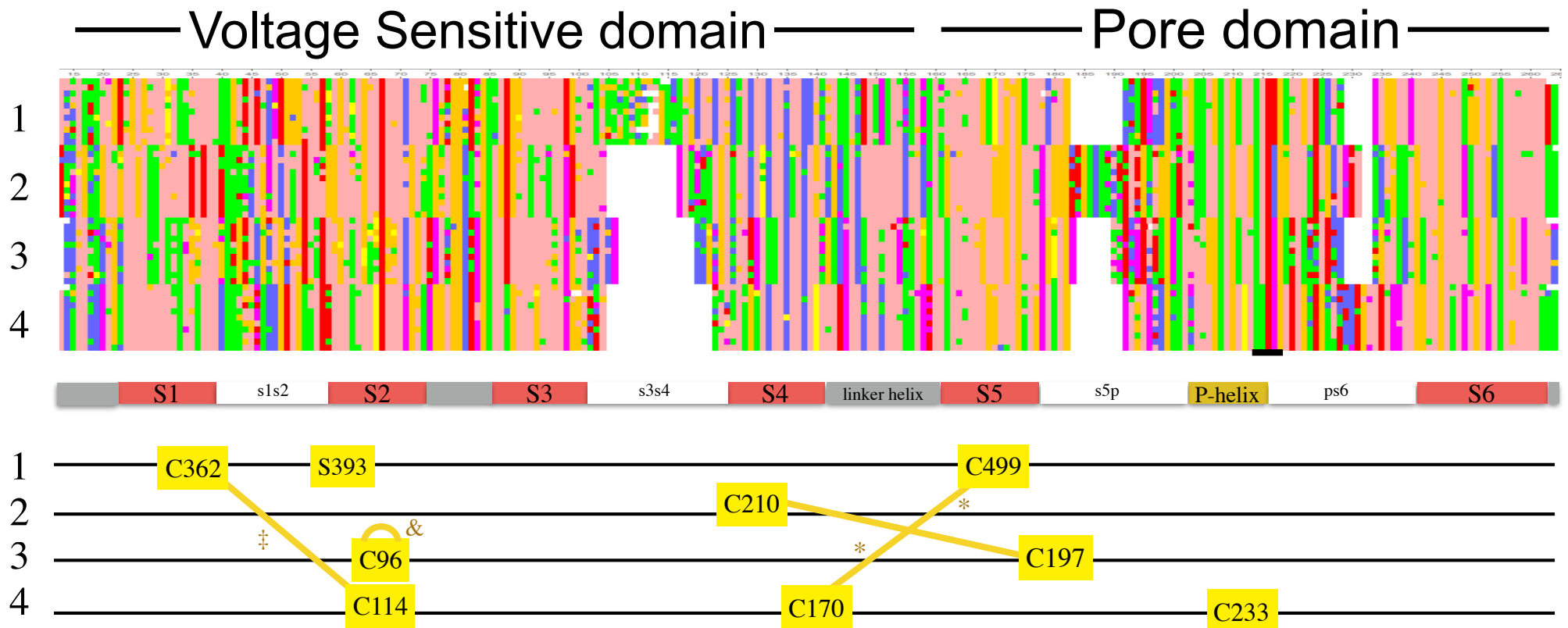


variable evolutionary position  
is consistent with easy  
mutational pathway



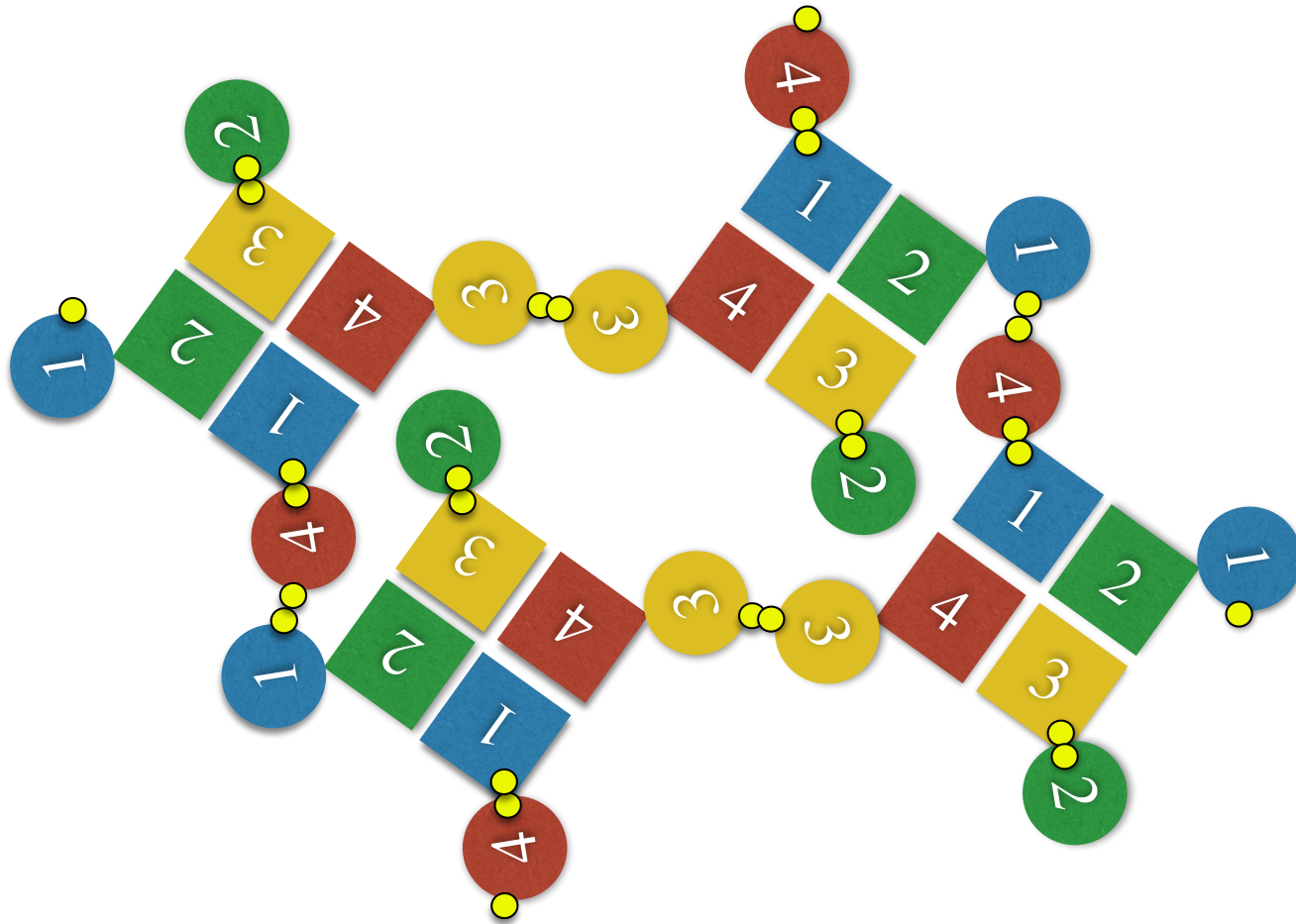
Variable position means self-reacting.

# Connecting the last remaining conserved cysteines

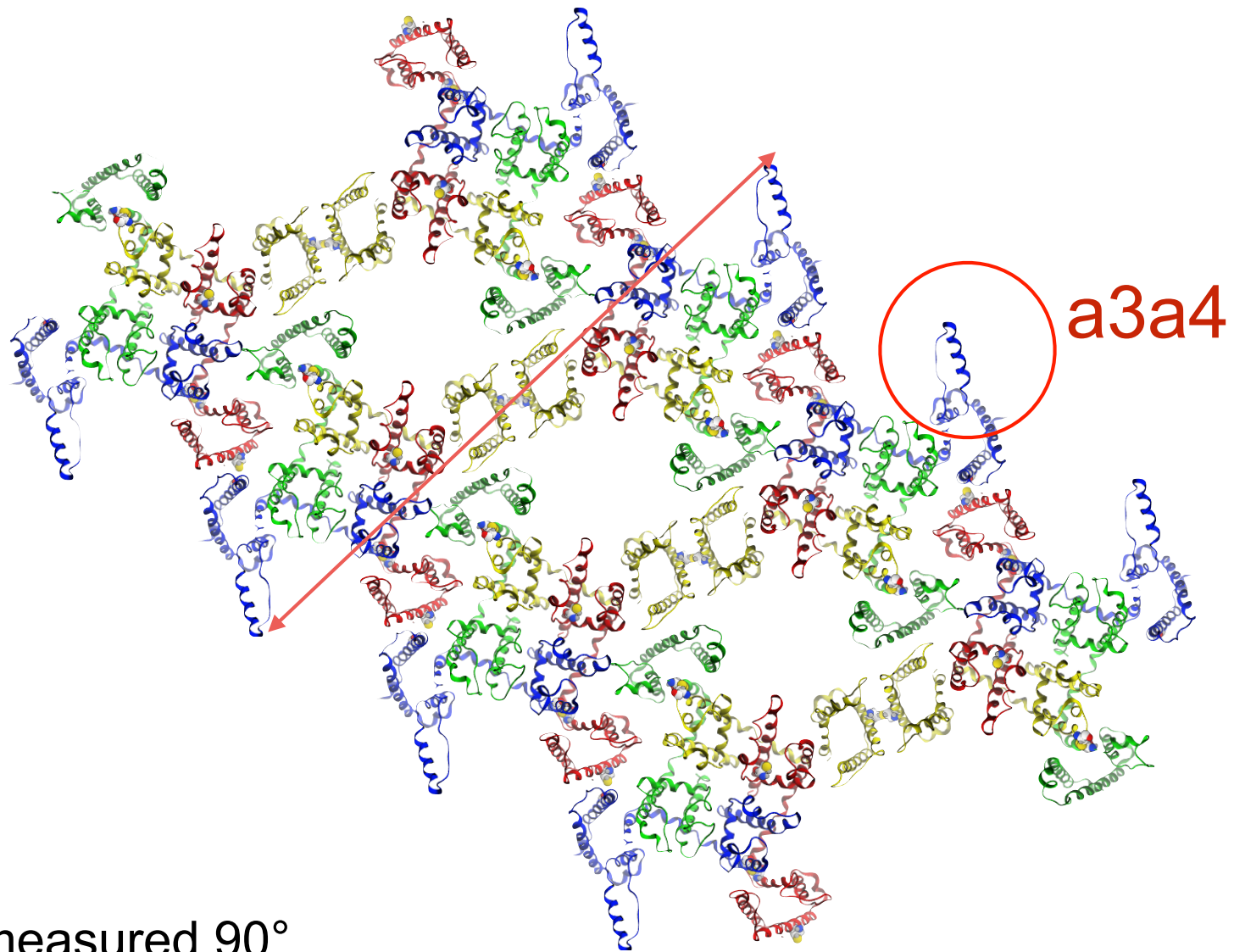


Final proposed disulfides

# Two-row model for super-quaternary structure



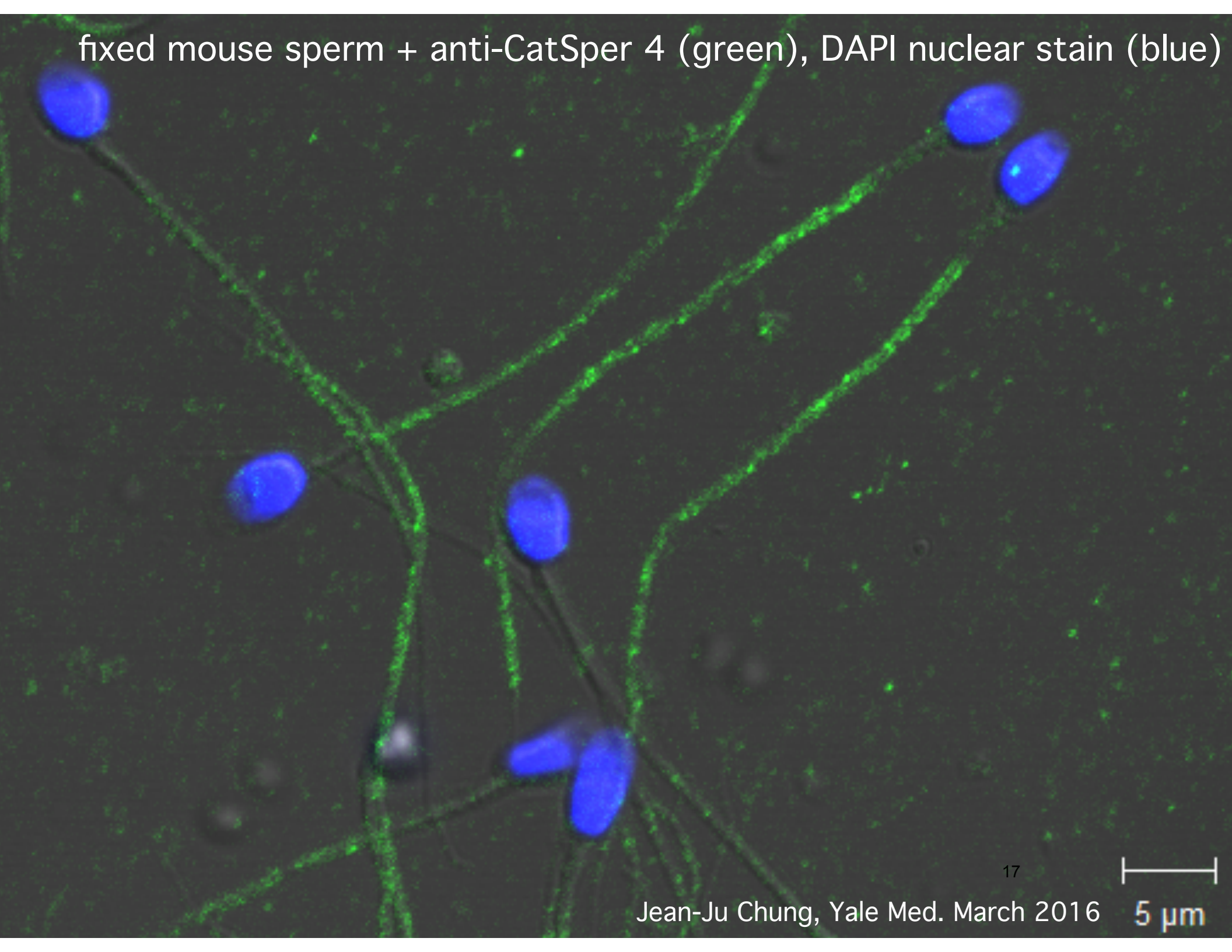
C4 N-term domain inserts between C1 and C2 N-term domains, creating a linear chain of tetramers, leaving the one remaining SH (C3 N-term domain) exposed. Single rows pair in anti-parallel fashion to give two-row model.



19.4nm measured  $90^\circ$   
from row direction,  
between a3a4 loops.

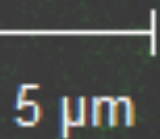


fixed mouse sperm + anti-CatSper 4 (green), DAPI nuclear stain (blue)

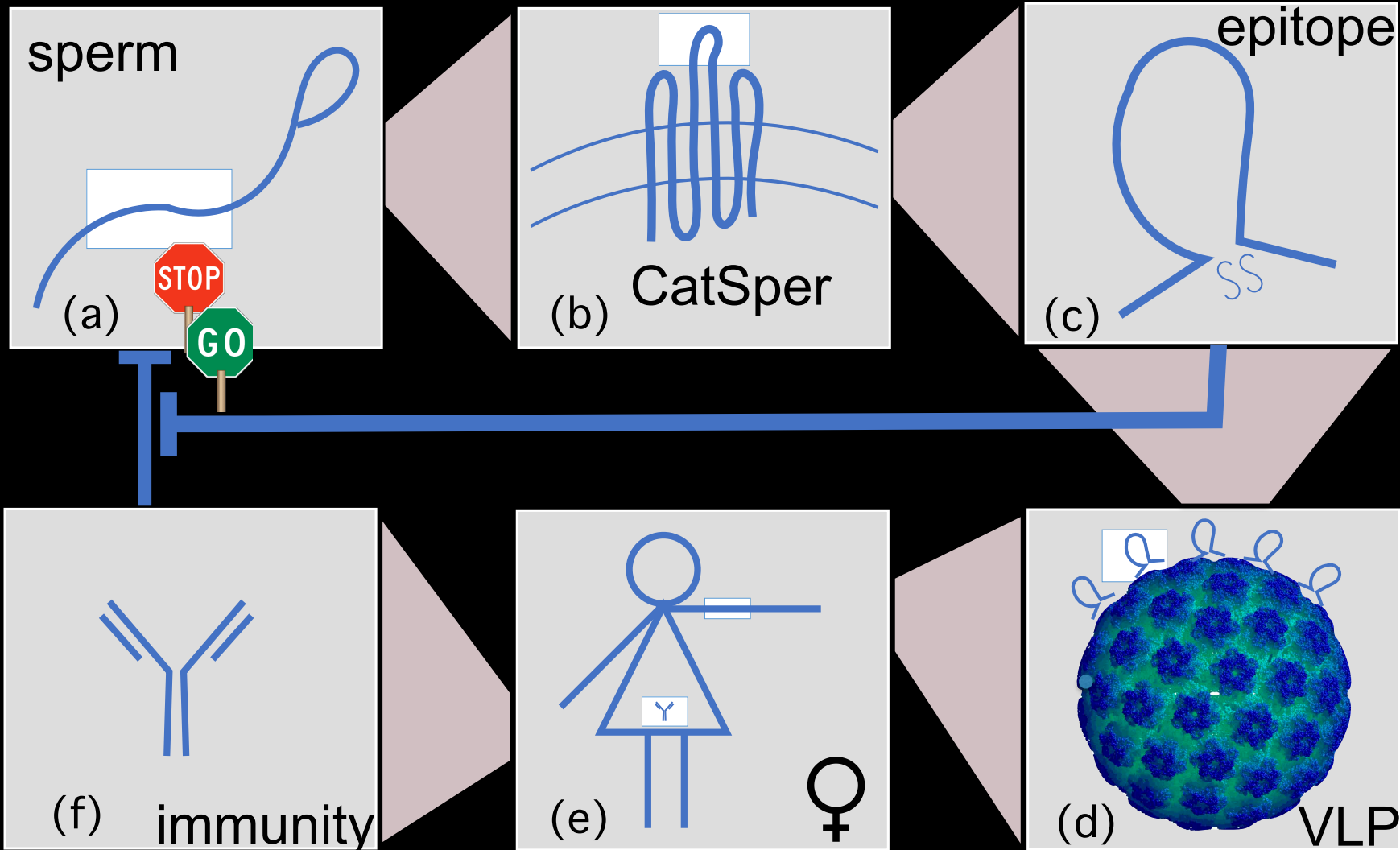


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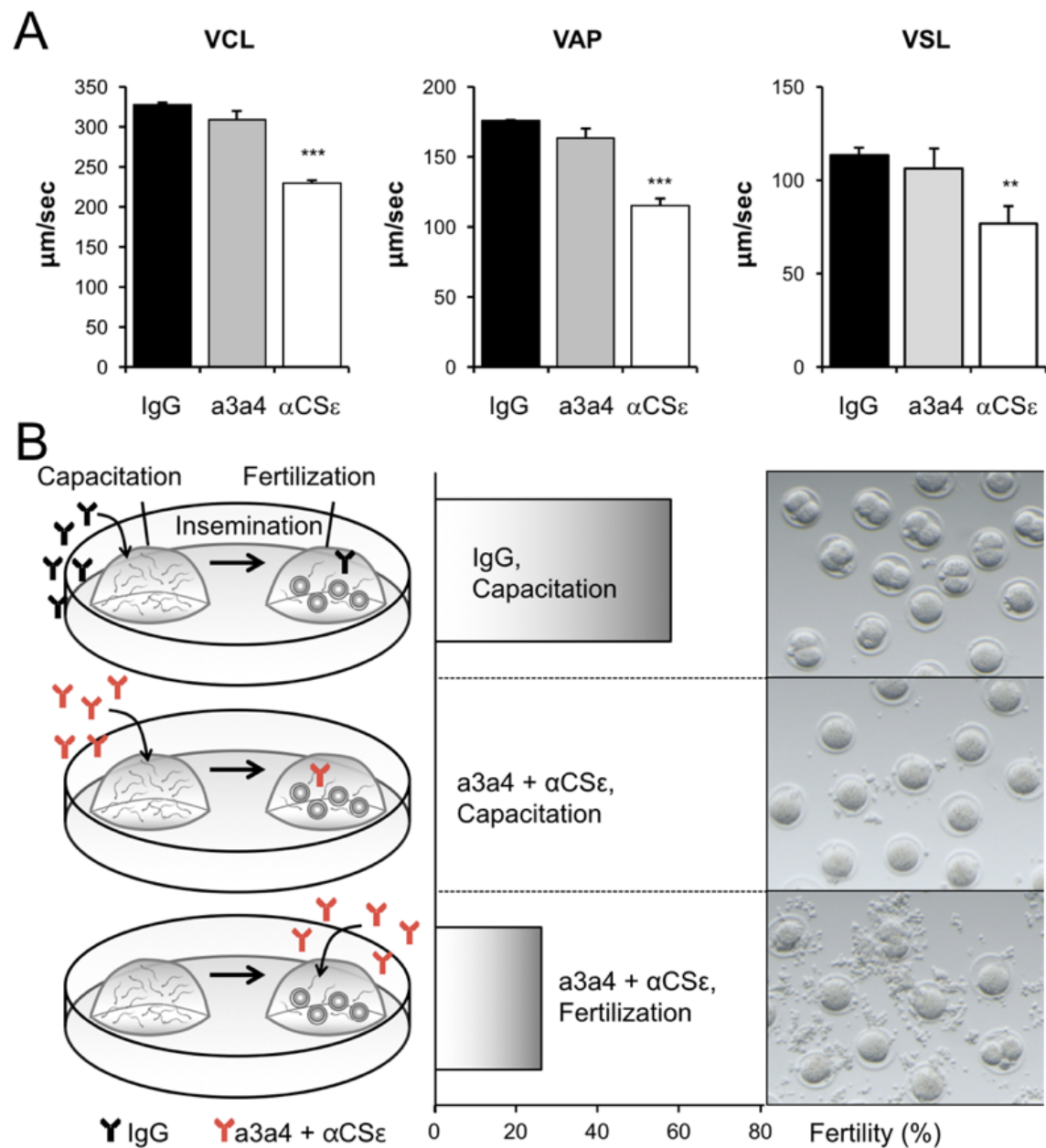
Jean-Ju Chung, Yale Med. March 2016



5 μm



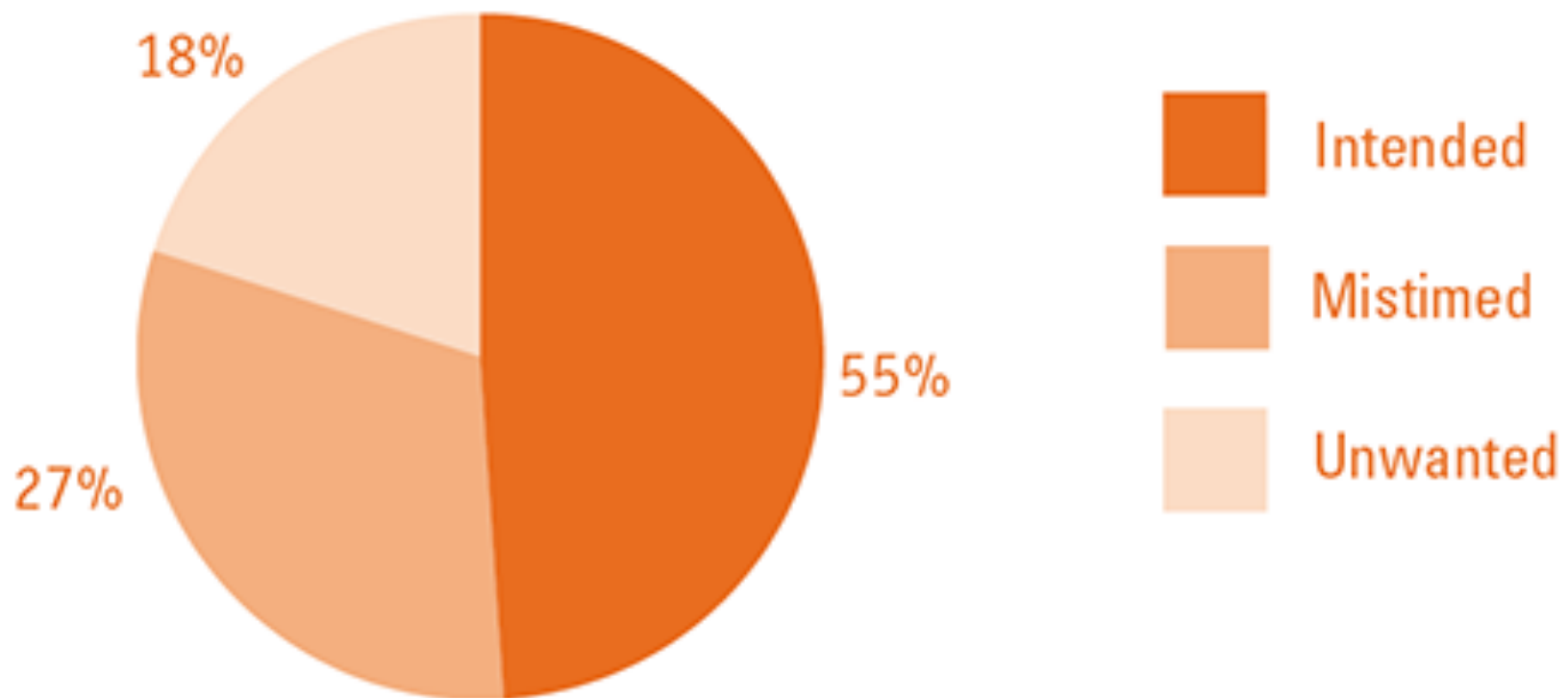




**Figure 6.** Inhibition of motility and fertility by CatSper antibodies against extracellular epitopes of CatSper 1 (a3a4) and epsilon (CS $\epsilon$ ). (A) Motility analysis of capacitated mouse sperm in the presence of 20  $\mu$ g/ml antibodies by CASA. (B) *In vitro* fertilization performed with COC eggs. Antibodies are treated either during sperm capacitation or fertilization at 20  $\mu$ g/ml (*unpublished*).

## Pregnancies by Intention Status

Nearly half of U.S. pregnancies are unintended.

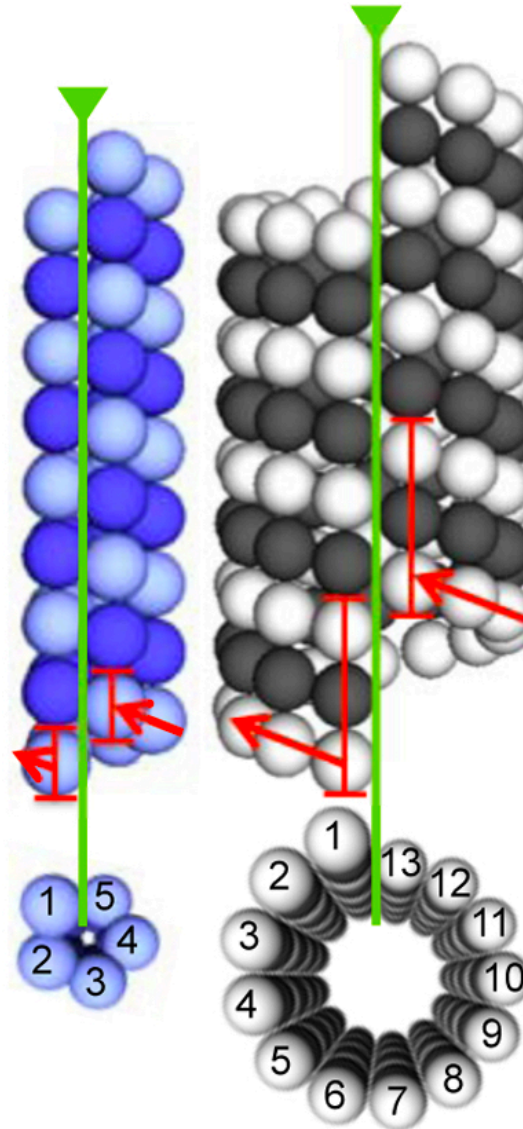
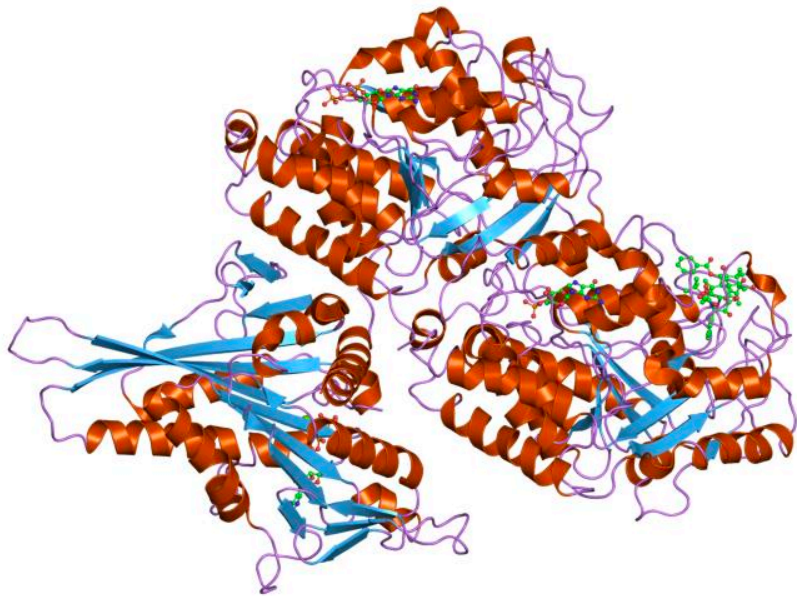


[www.guttmacher.org](http://www.guttmacher.org)

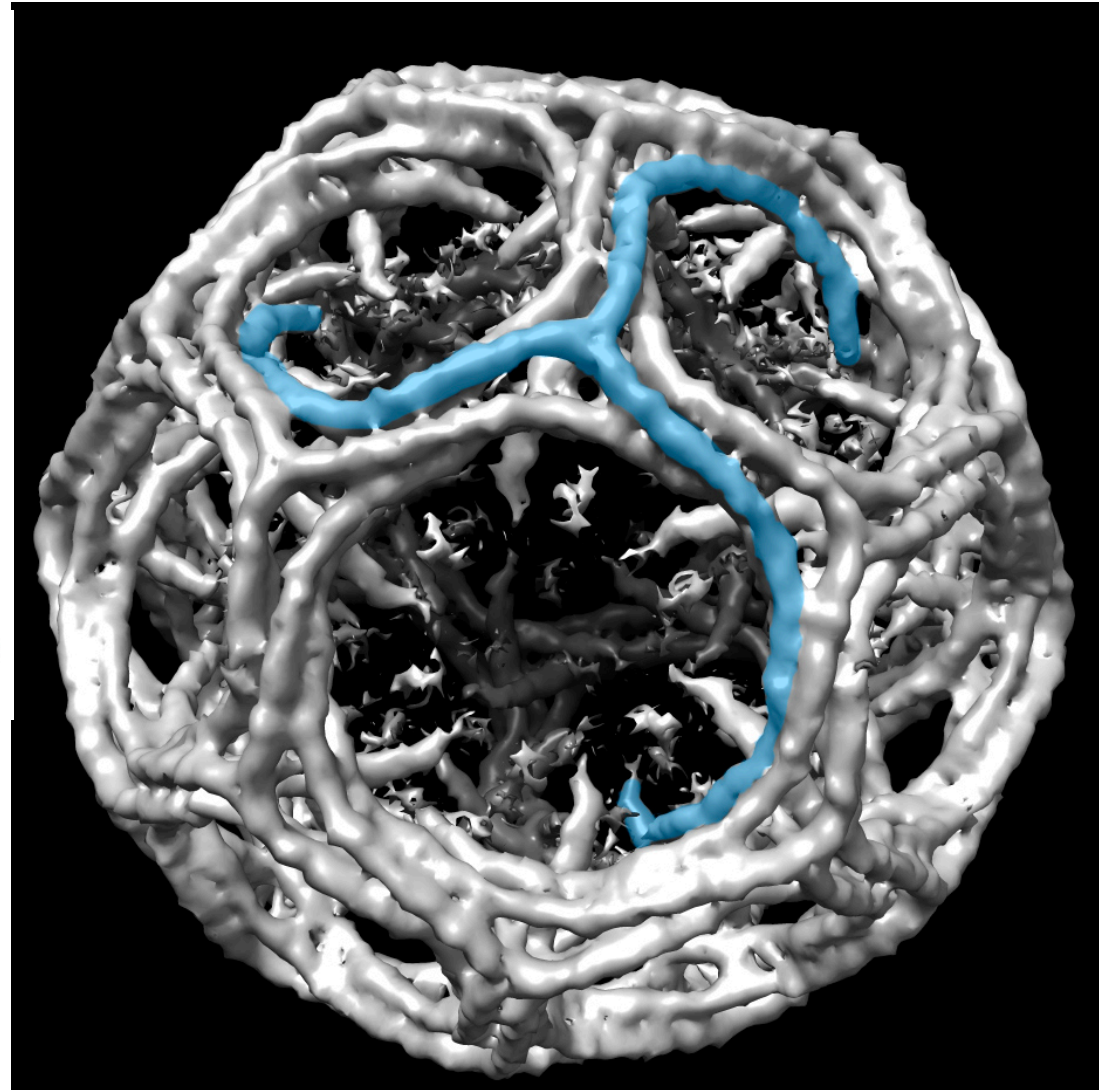
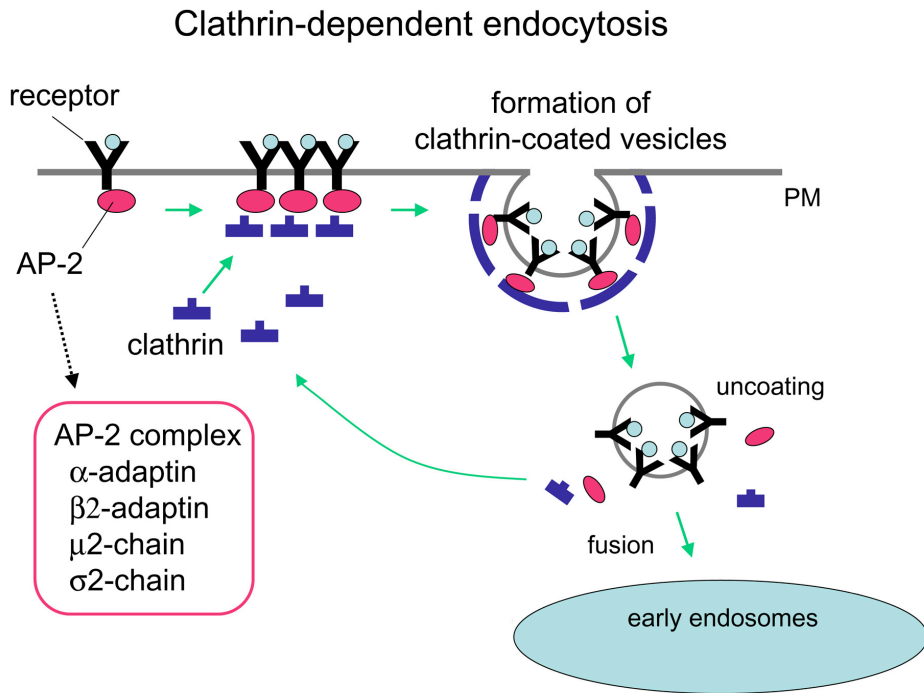
# Other superquaternary structures

- Actin
- tubulin
- clathrin
- capsids

# Tubulin

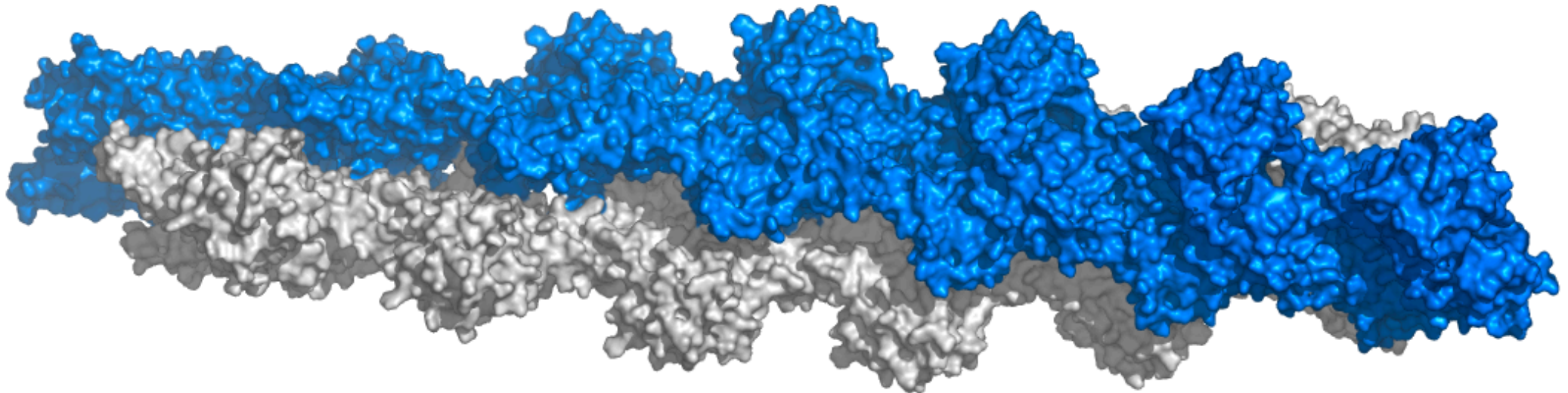
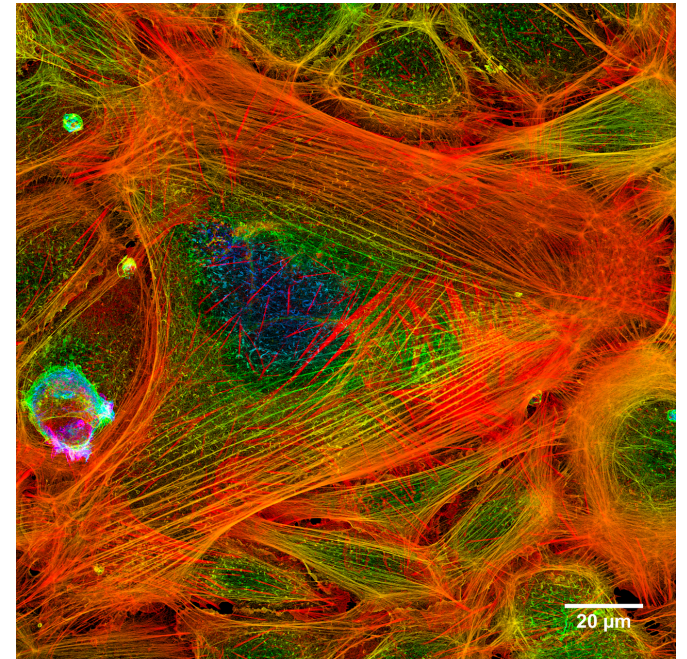
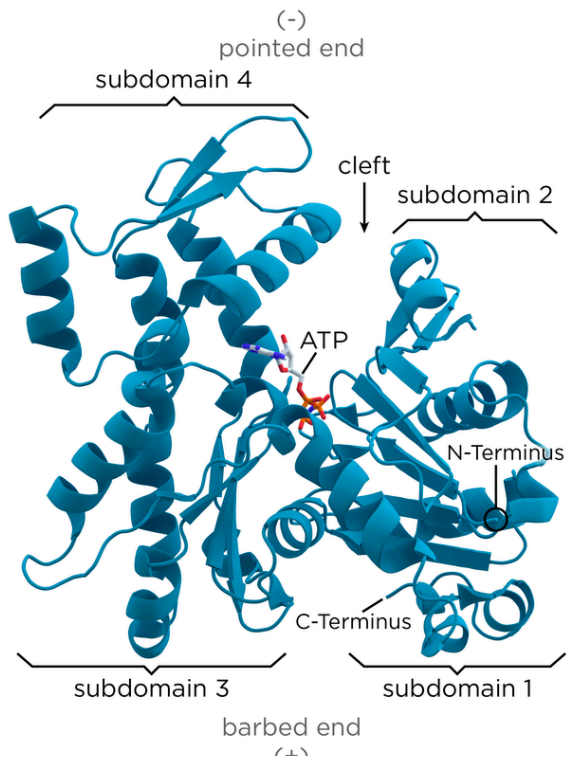


# Clathrin

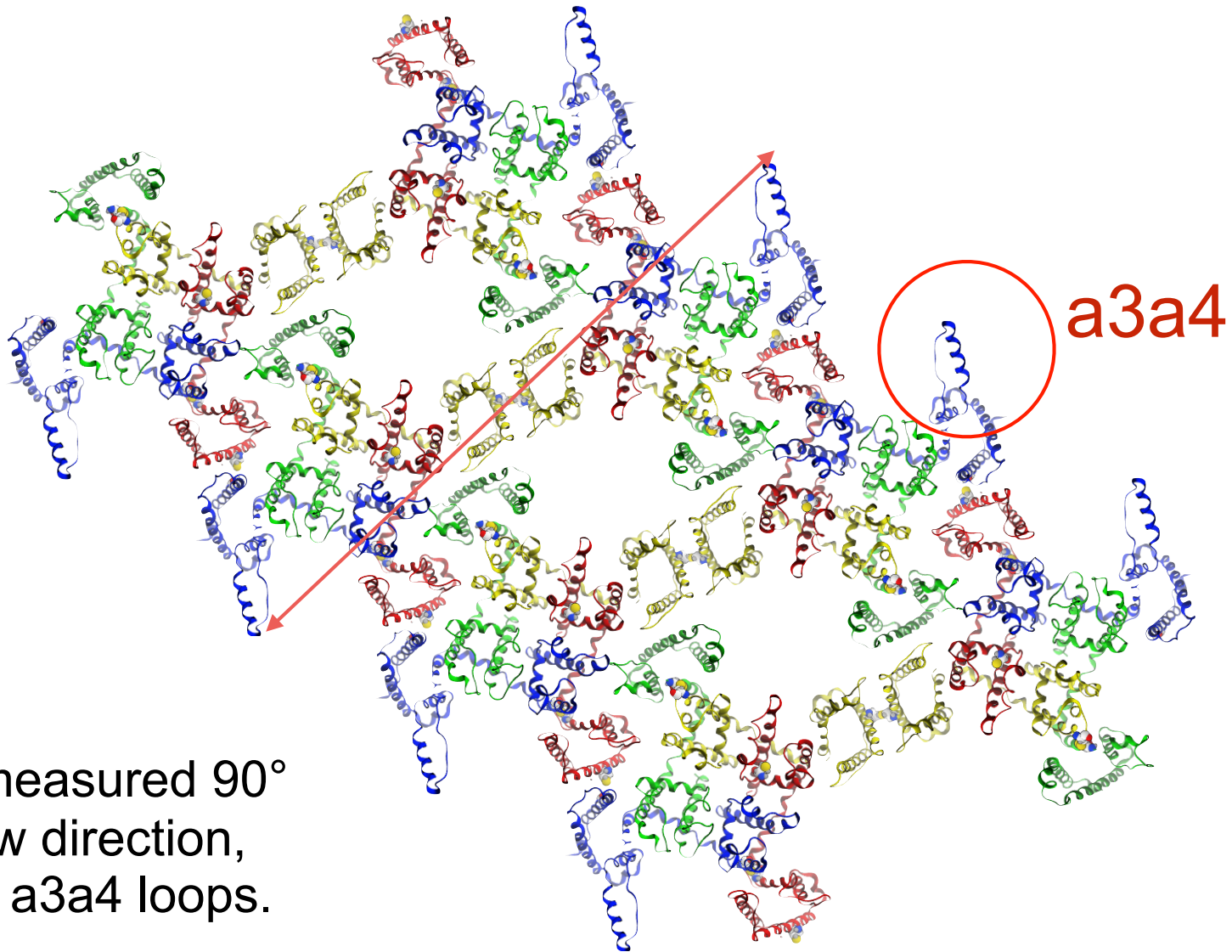




# Actin



Two-row Catsper displays C4 N-term domain. If it self-associates, then anit-parallel rows can stack,



19.4nm measured  $90^\circ$   
from row direction,  
between a3a4 loops.



# 10.1 SWISS-MODEL

automated homology modeling

SWISS-MODEL operates in 3 modes

First approach mode

needs to find a template with at least 25% identity

Alignment mode

You provide an alignment of target to template.

Project mode

You provide a DeepView project file containing multiple templates and a model. Useful for “second pass” modeling.

# SWISS-MODEL Algorithm

**4 steps:** template selection -- alignment -- model building-- evaluation

## 1) Template selection

Template batches for target domains.

## 2) target/template alignment

≤5 templates/batch.

High RMSD templates removed.

Placement of indels optimized. <--- *Take note.*

Islands moved to flanks. (*What is an “island”*)

## 3) Model building

Core coordinates averaged, weighted by sequence similarity to target.

Loop ensemble created using *Constraint Space Programming*.

Flanking residues added in if no good loop found.

If no good loop found or length > 10, then a *database search* is done.

Side chains built iso-sterically. <--- *Is this always possible?*

Uses backbone dependent rotamer library for side chains.

Scores H-bonds, SS-bonds

Energy minimization used only to *regularize* structure. <--- *What does regularize mean?*

## 4) Evaluation

WhatCheck -- atomic mean force potential -- used to identify problem areas.

Alignment is seen as the main source of modeling errors. <--- *my experience exactly*