

# **Investigating The Role of Ceramide In The Bam Complex**

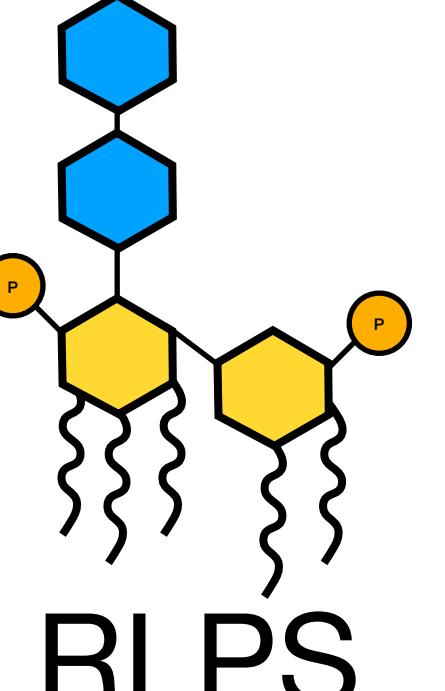
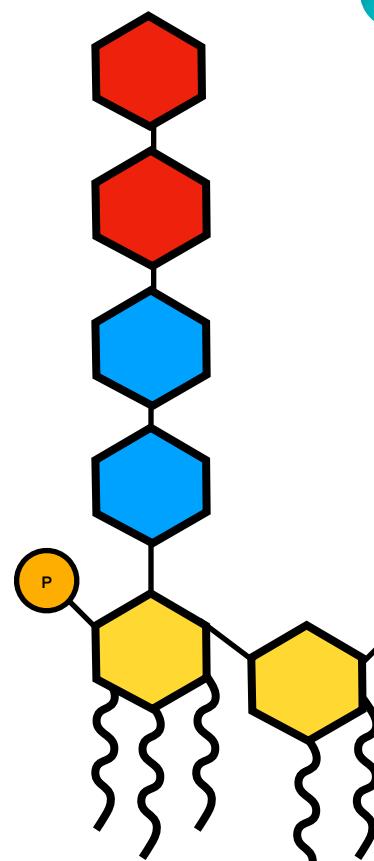
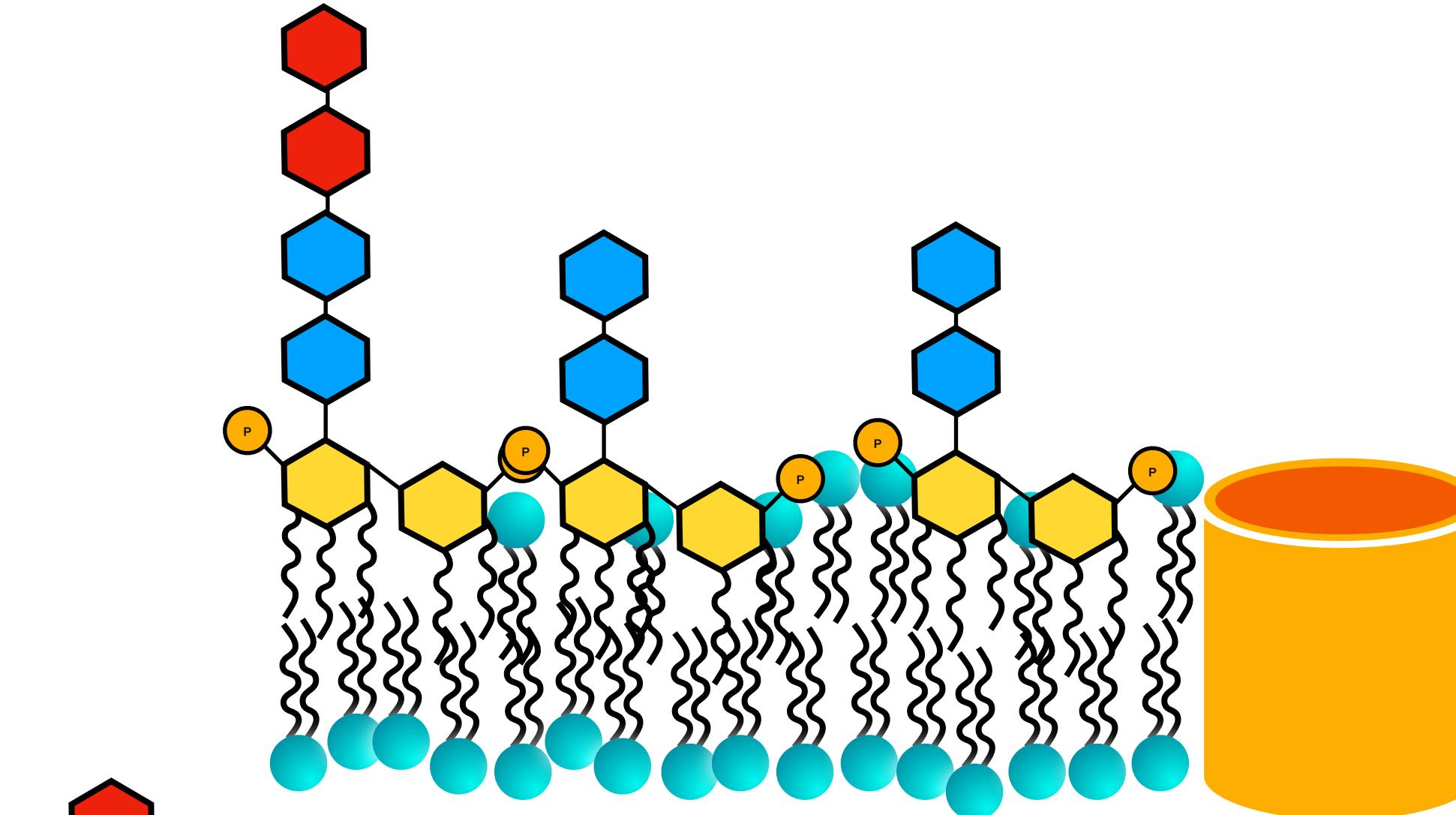
**Jahmal Ennis**

# Outline

- Background and Motivation
- Research Question
- Approach
- Results
  - Step 0: Modeling a Bacterial Membrane
  - Bacterial membranes diffuse slowly
- Summary & Future Direction

# Caulobacter crescentus outer membrane contains ceramides

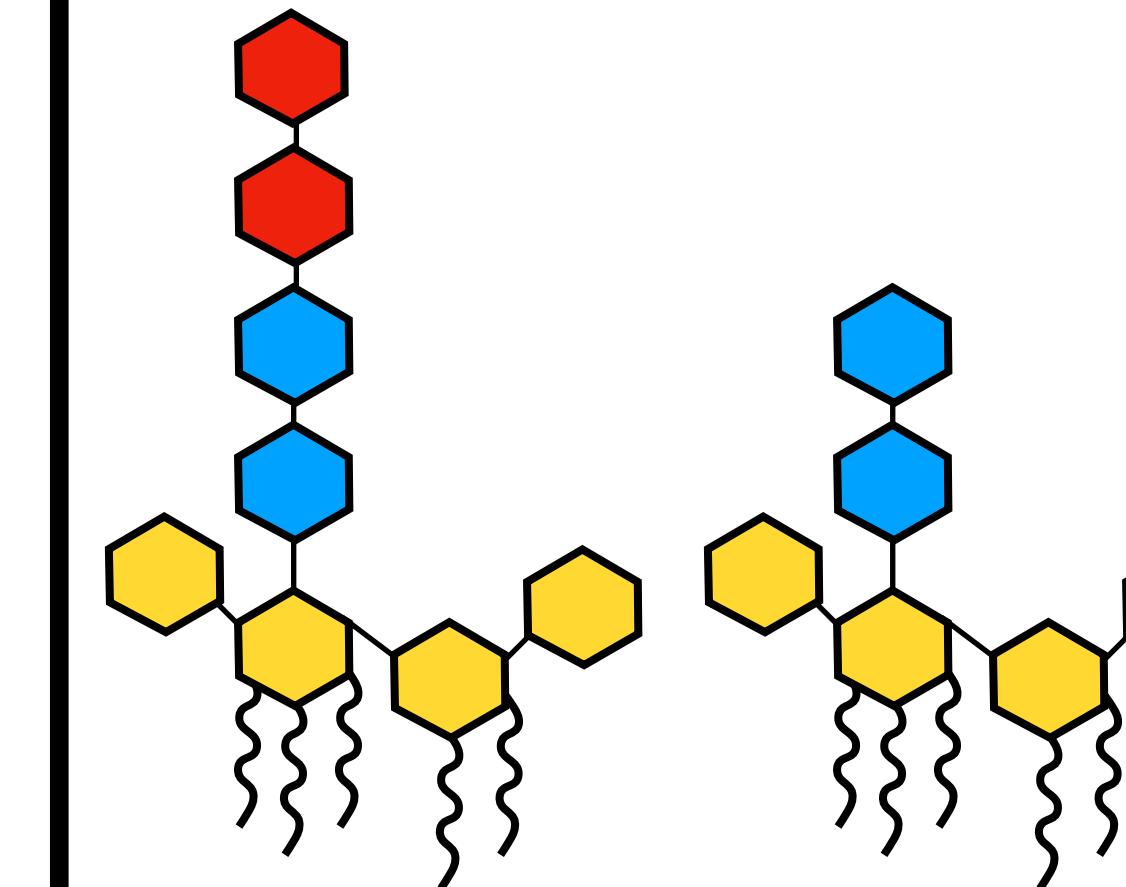
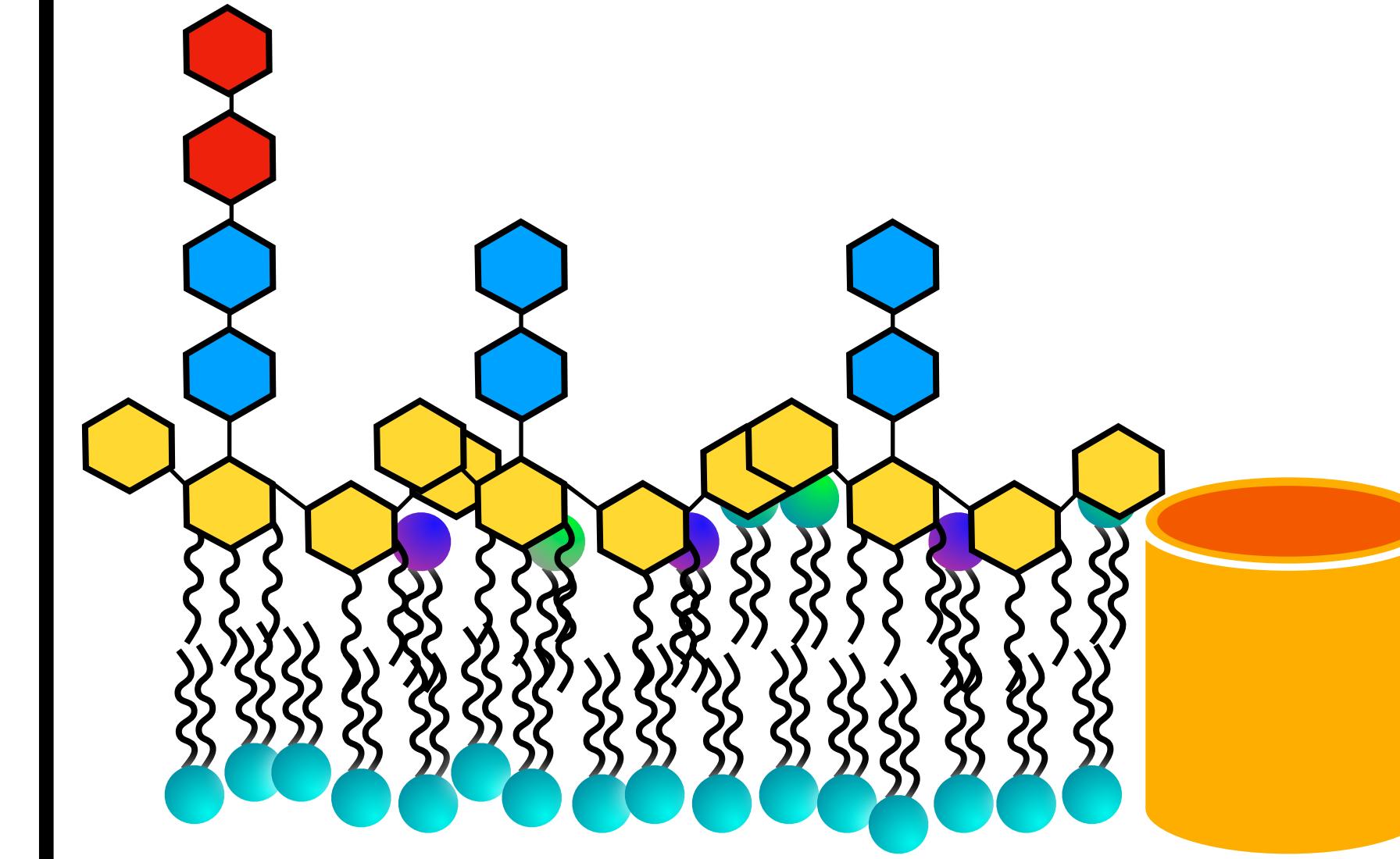
Ecoli Outer Membrane



Phospholipids

Protein<sup>3</sup>

Caulobacter Outer Membrane

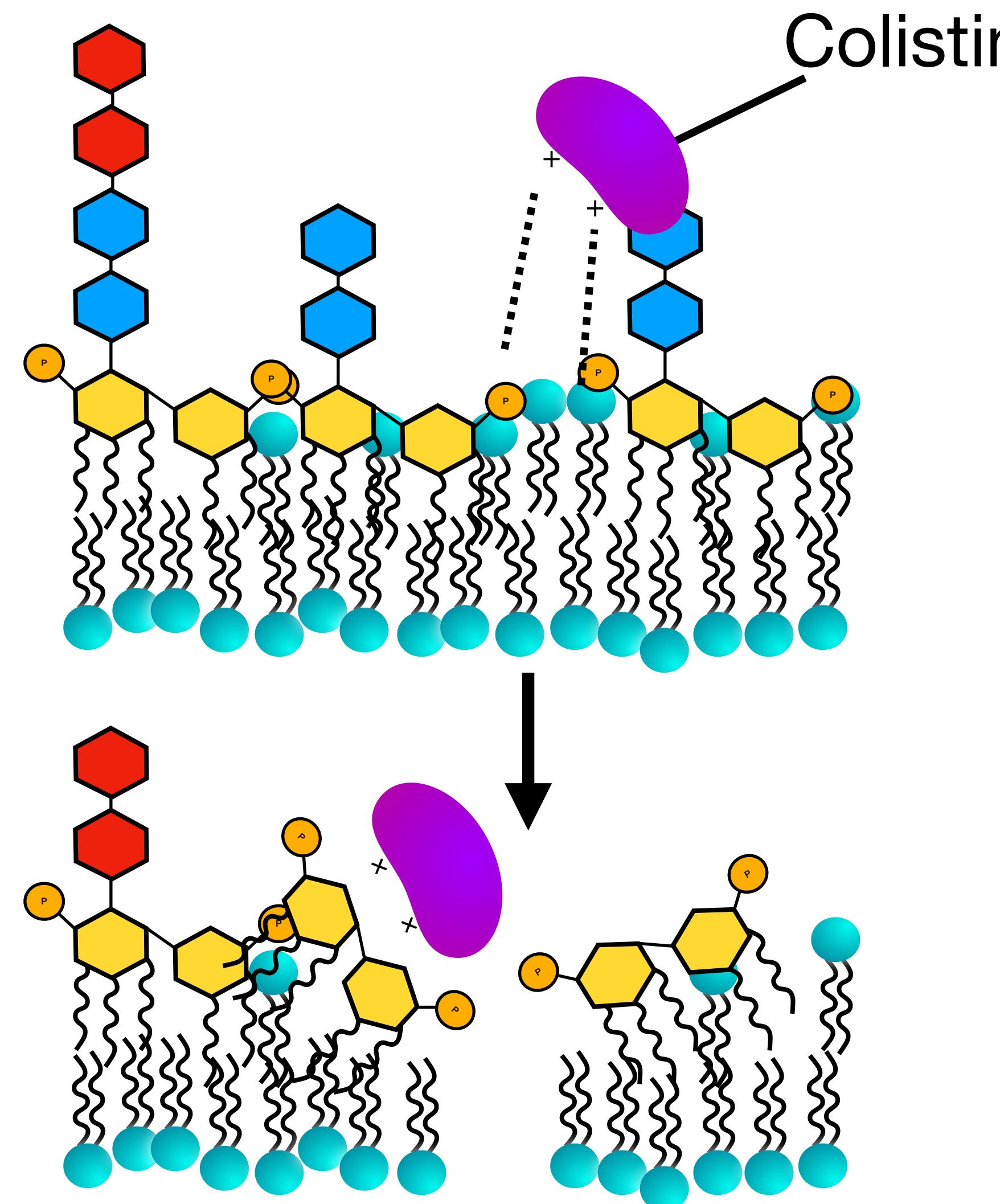


Anionic  
+

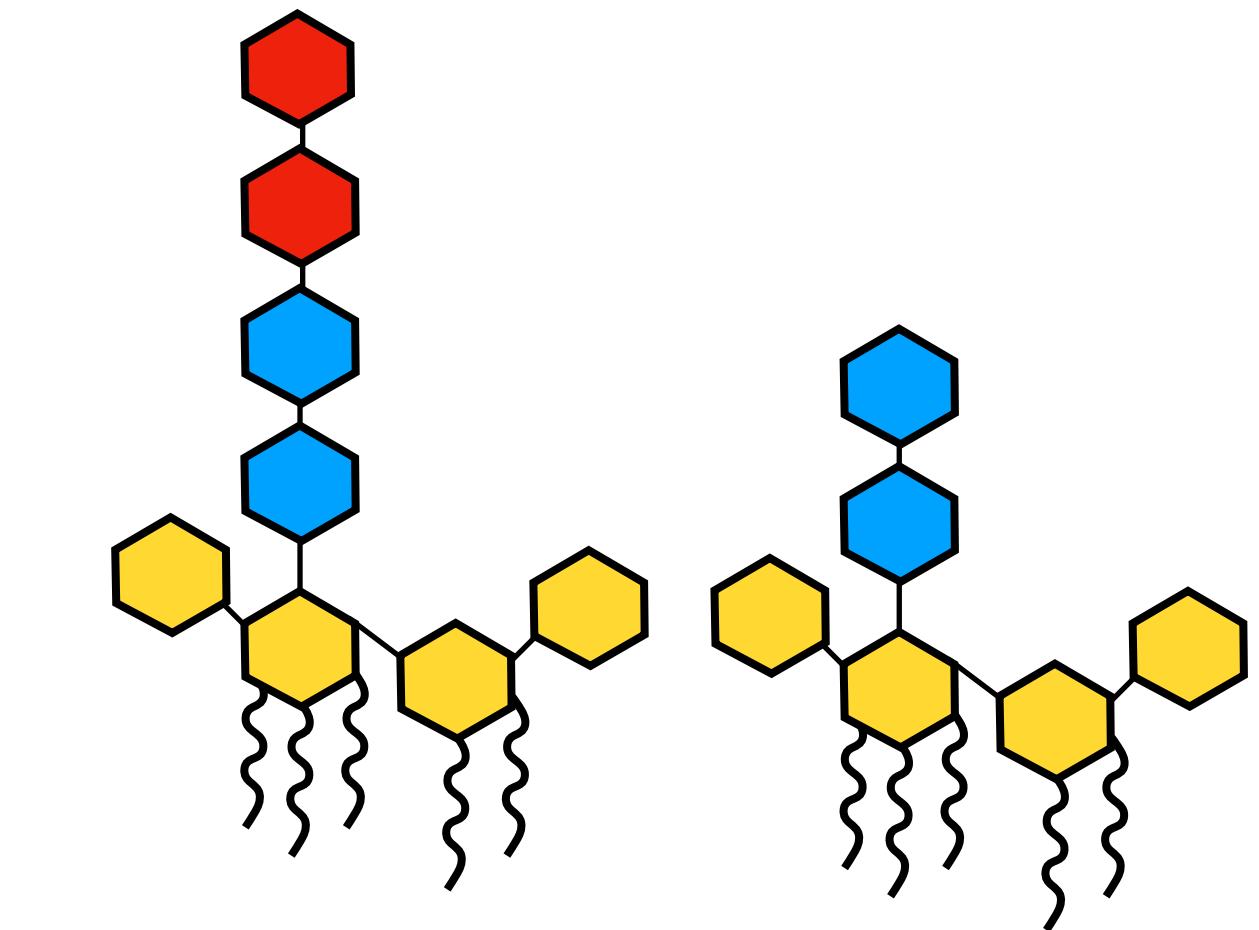
Neutral  
Ceramides

# Mechanisms for antibiotic susceptibility in E. coli

## Polymixin Disruption in E. coli



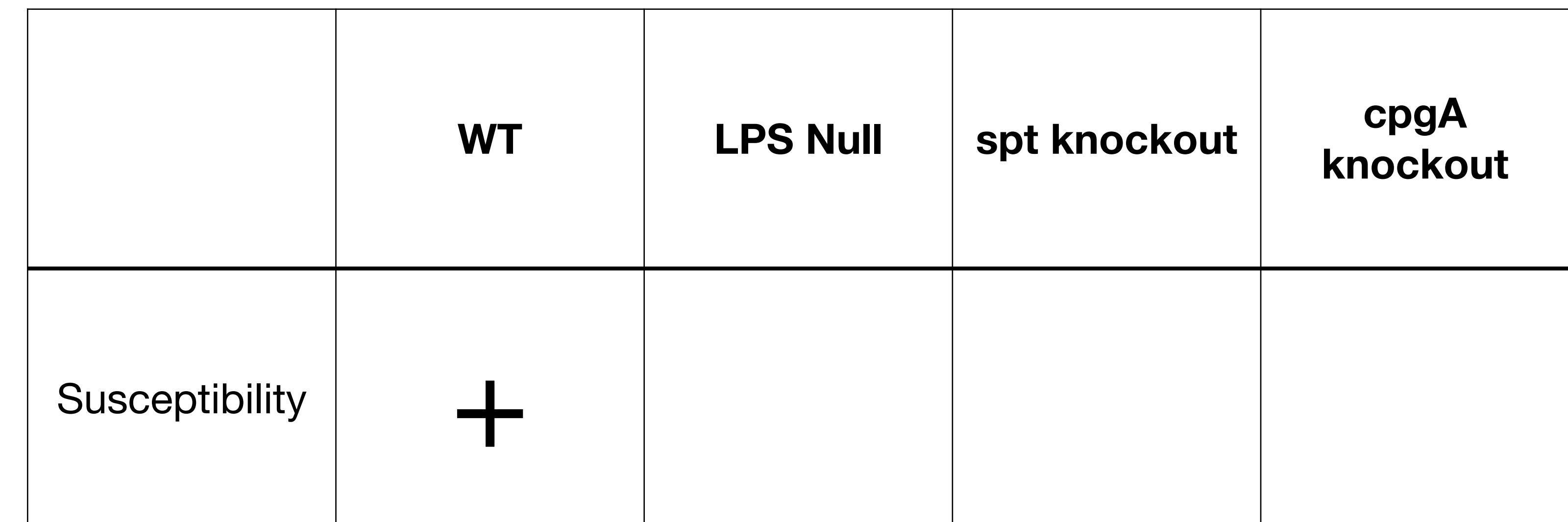
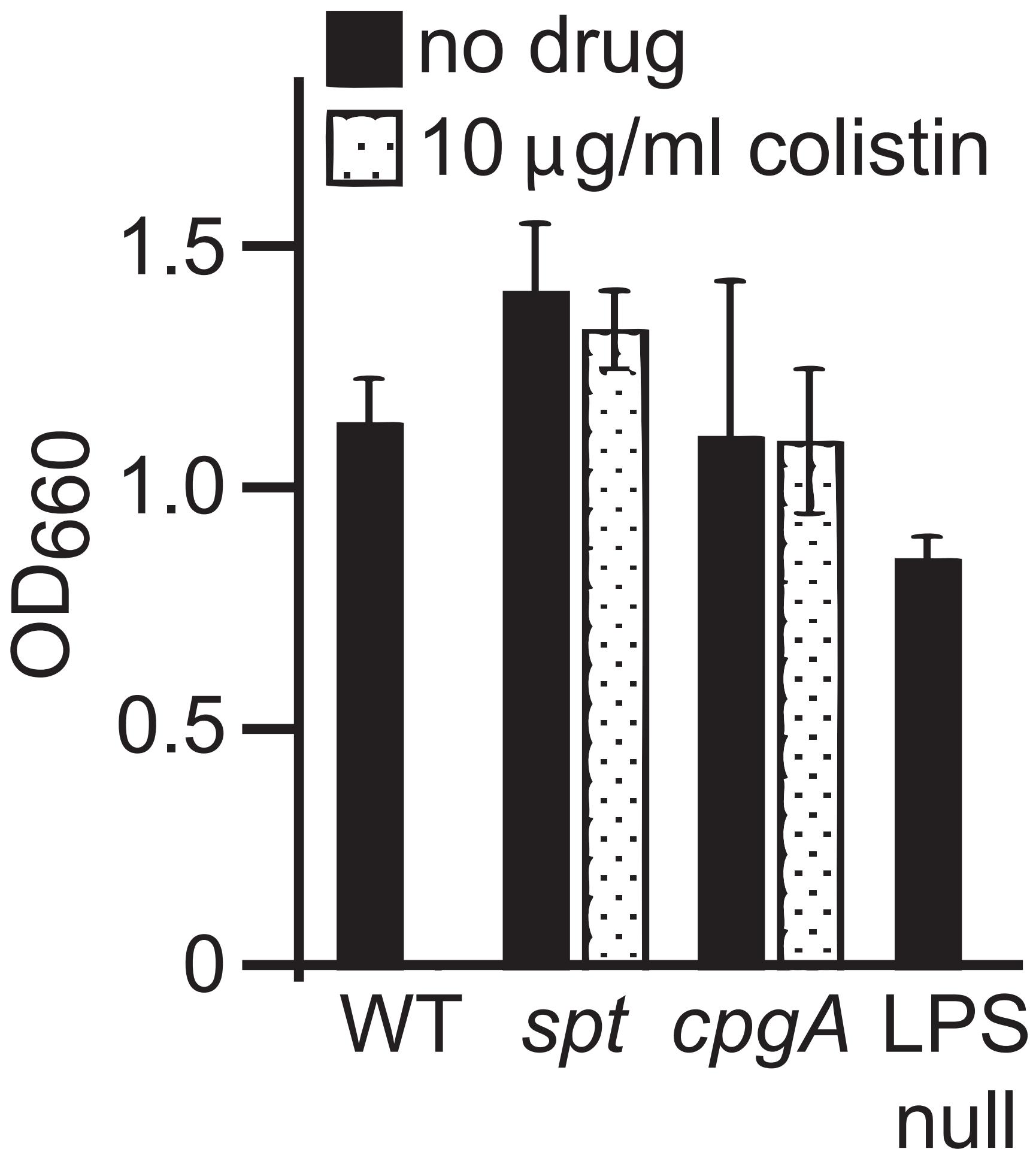
## Caulobacter Lipids



SLPS

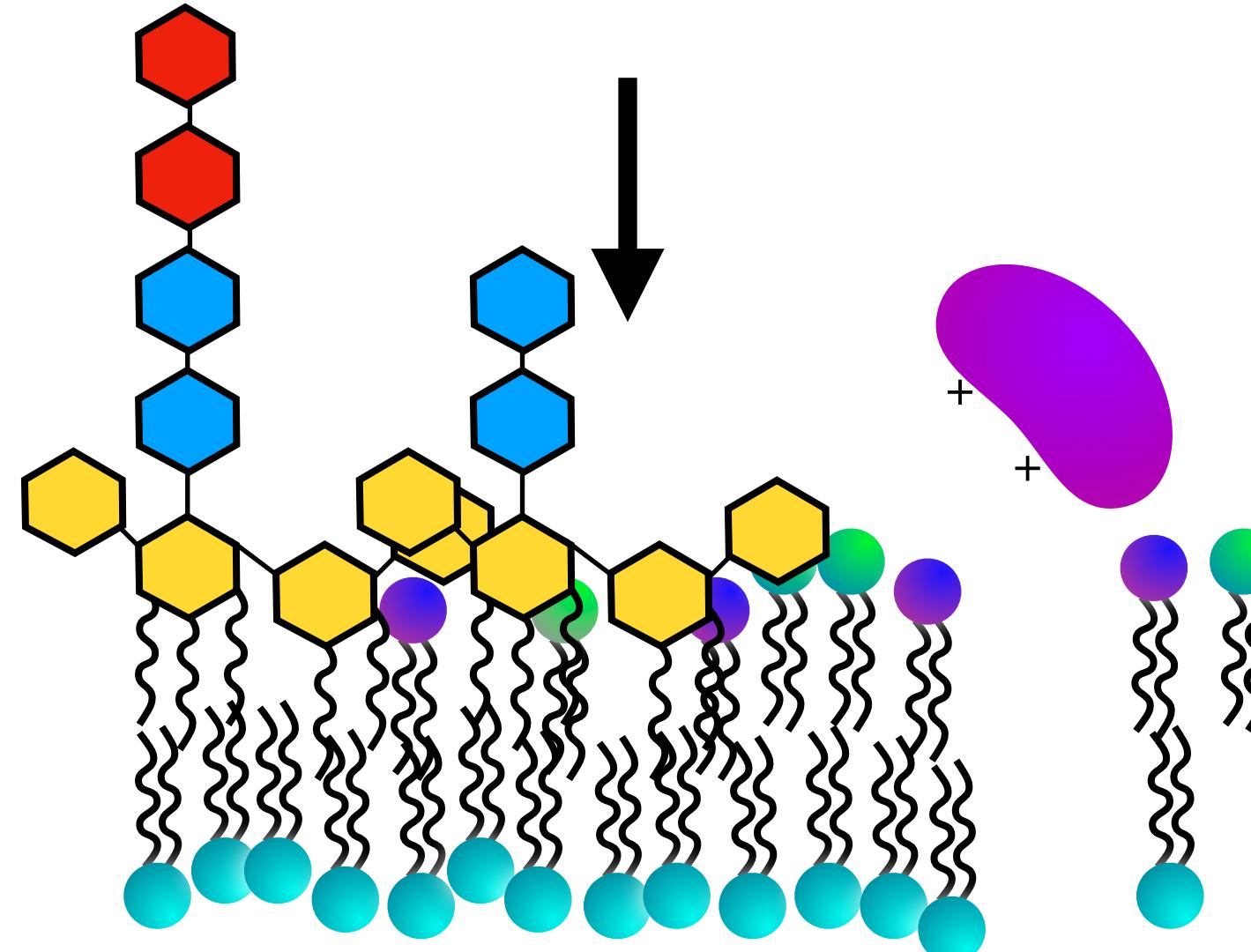
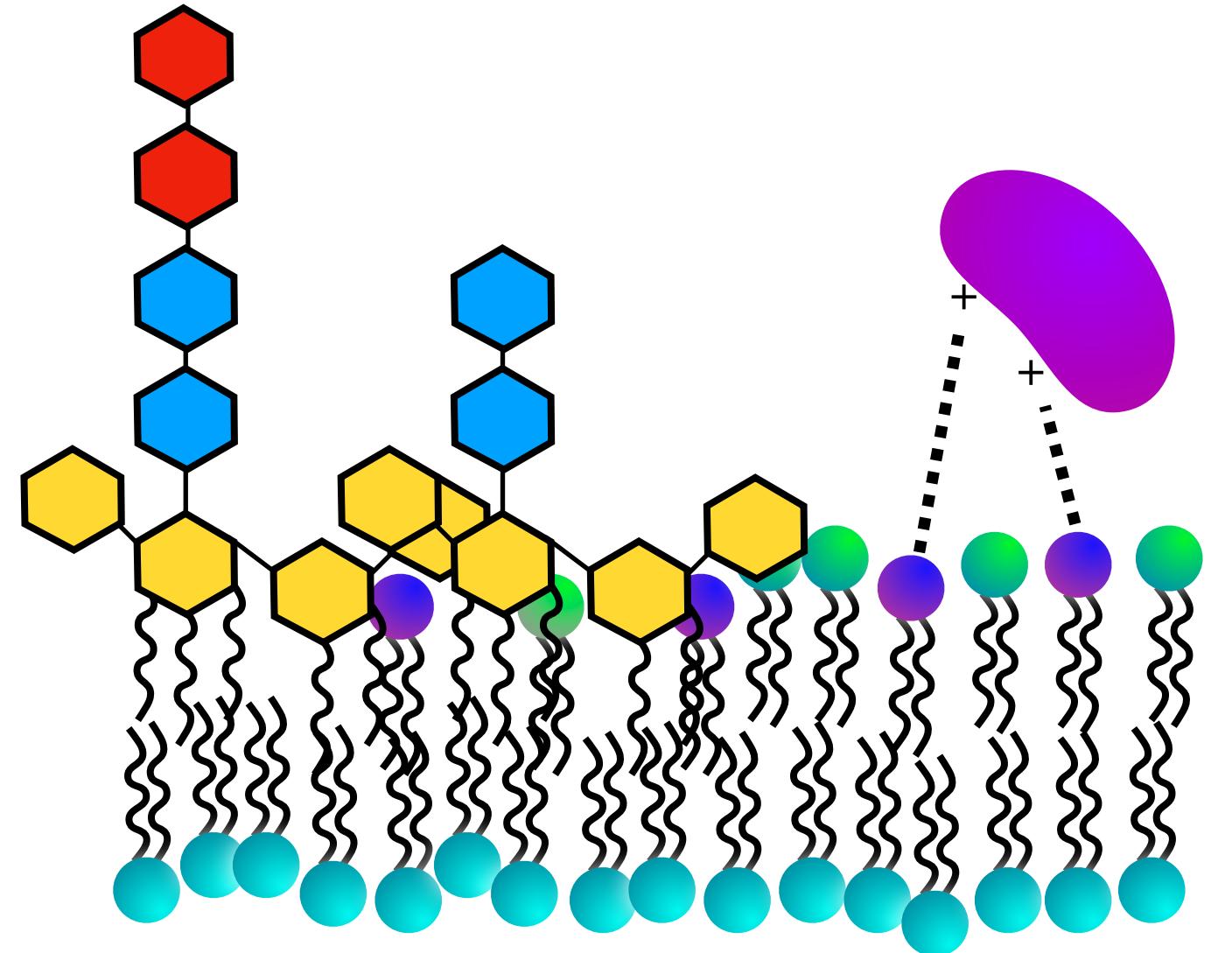
RLPS

# Sphingolipids affect antibiotic susceptibility

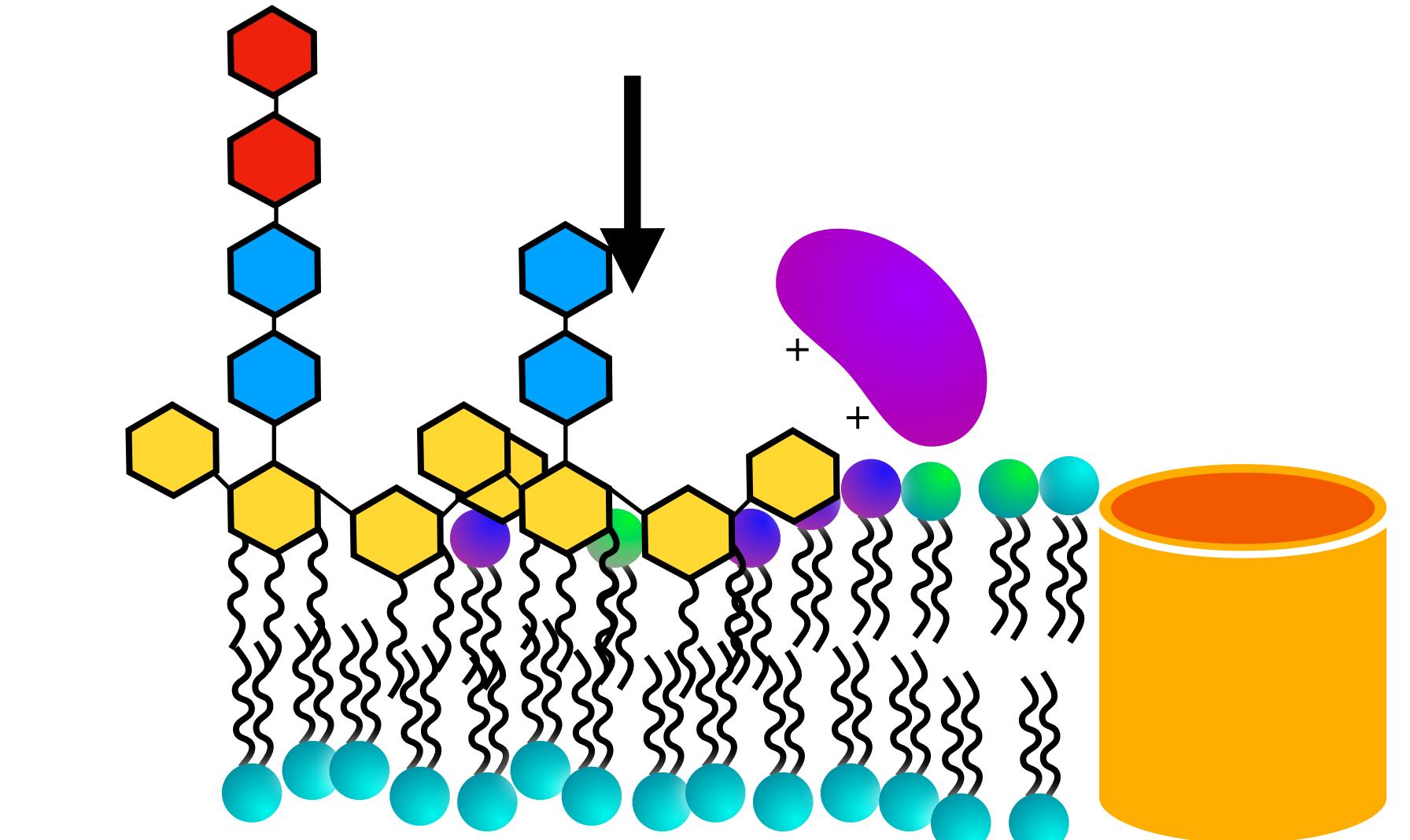
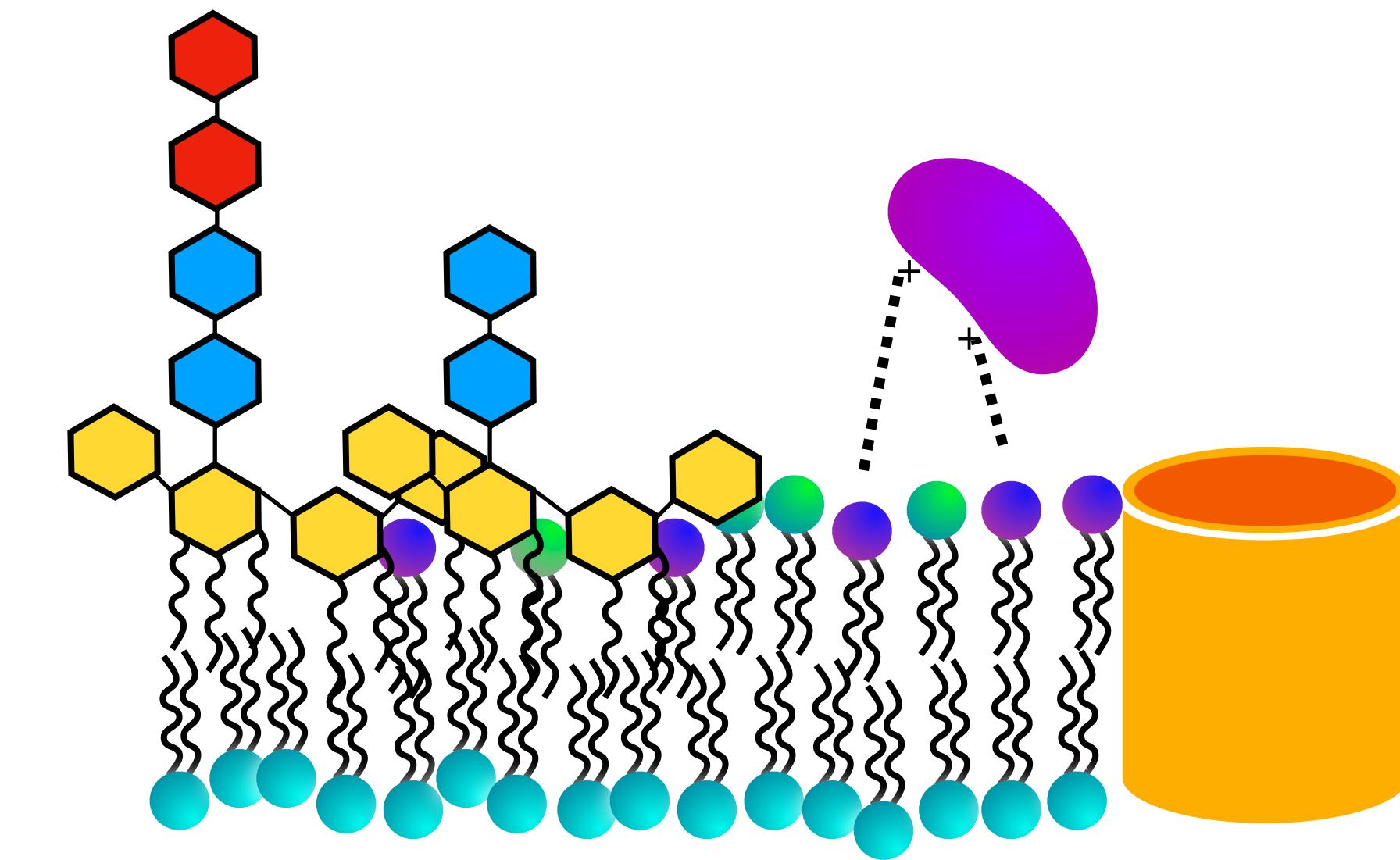


# Two possible mechanisms for antibiotic susceptibility in *C.crescentus*

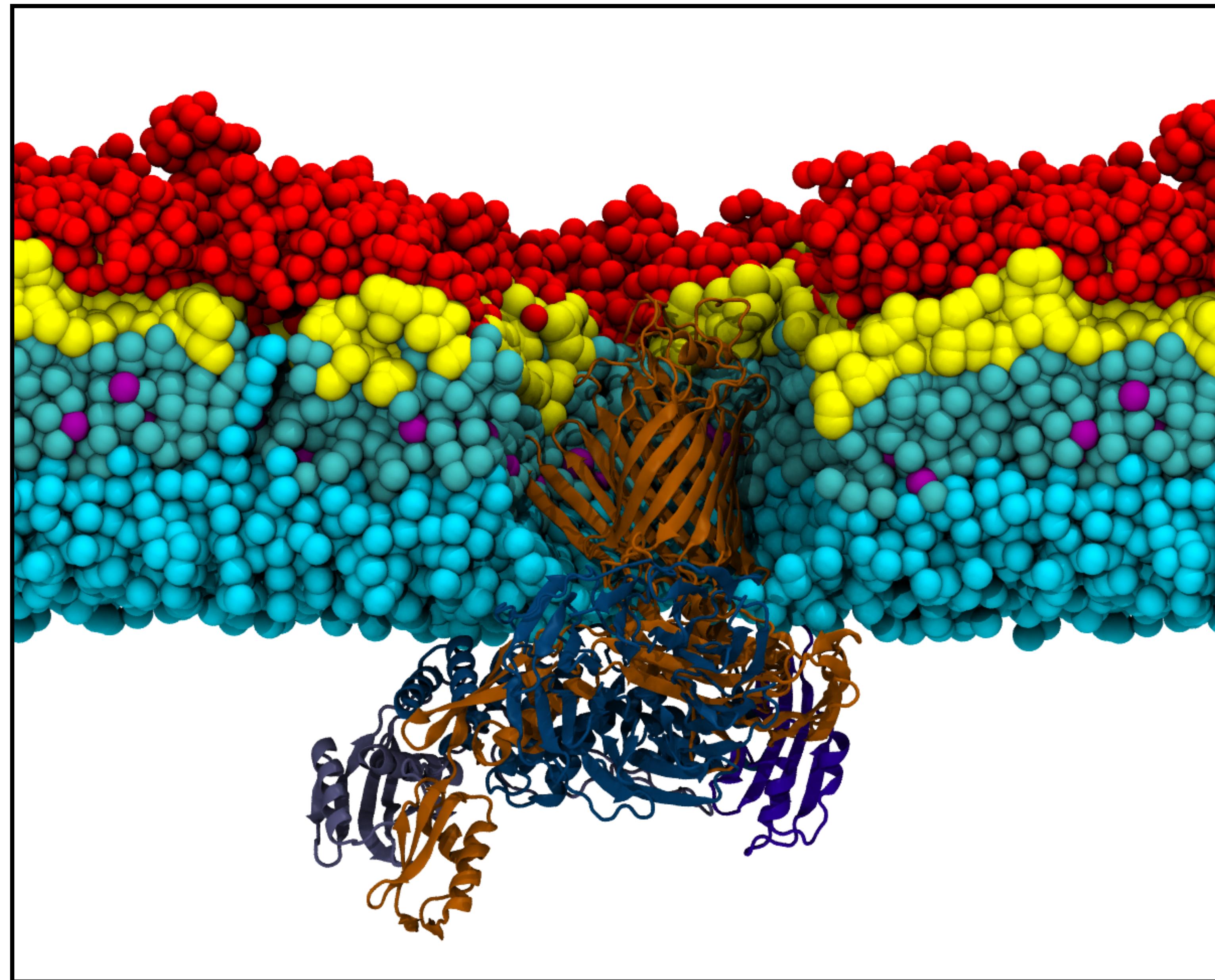
## Sphingolipid Extraction



## Sphingolipid binding with protein



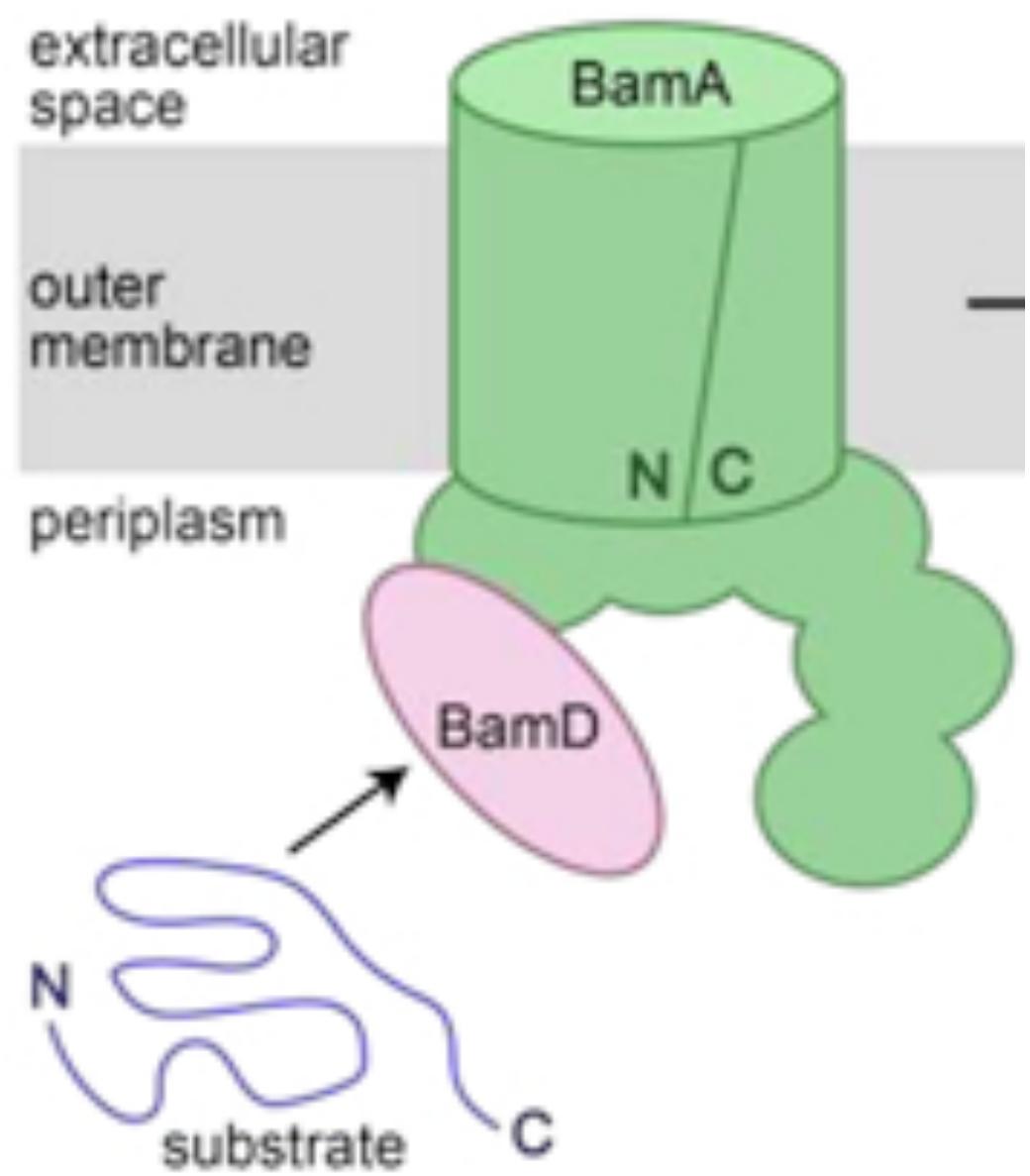
# Other possible mechanism: Interference with BamA function



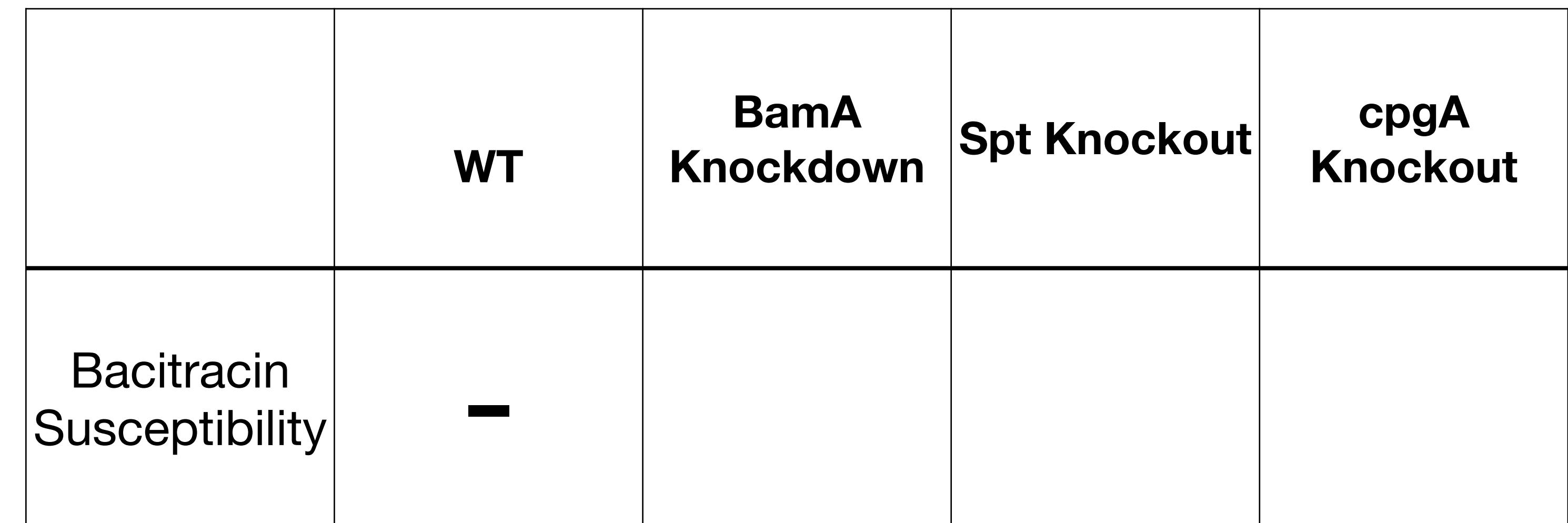
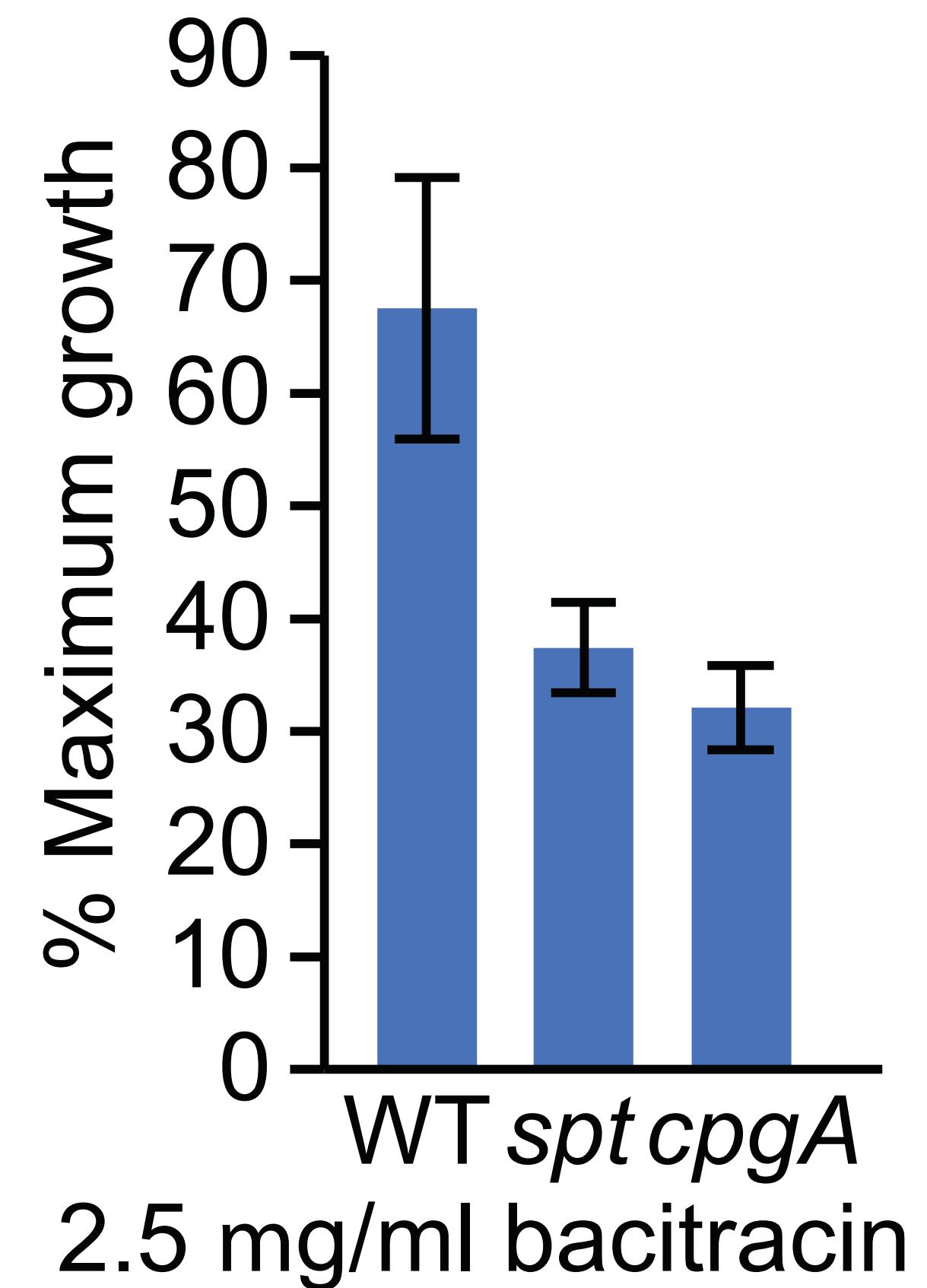
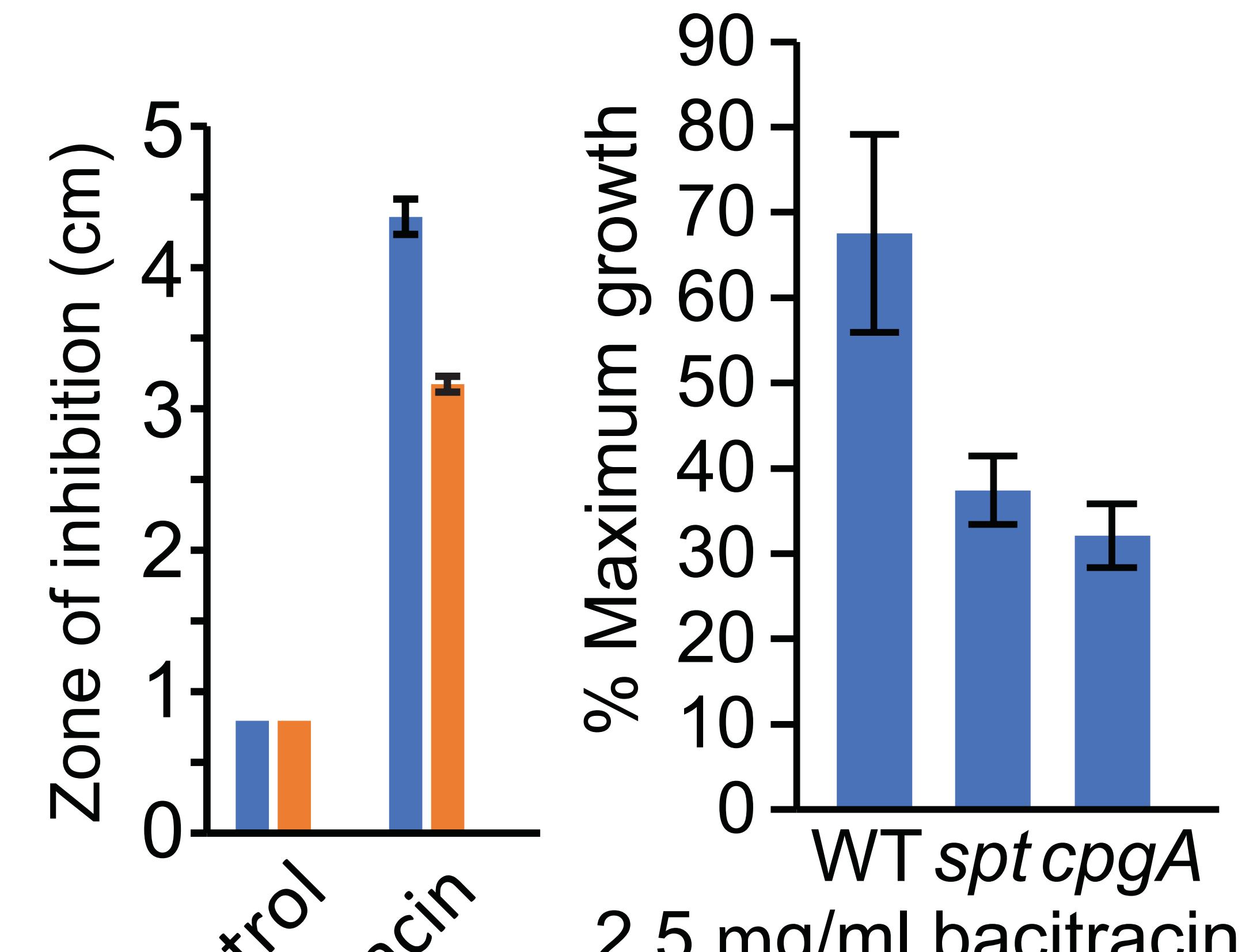
# BamA folds proteins in the outer membrane

(a)

Lateral gate closed



# Ceramide's may modulate BamA Function



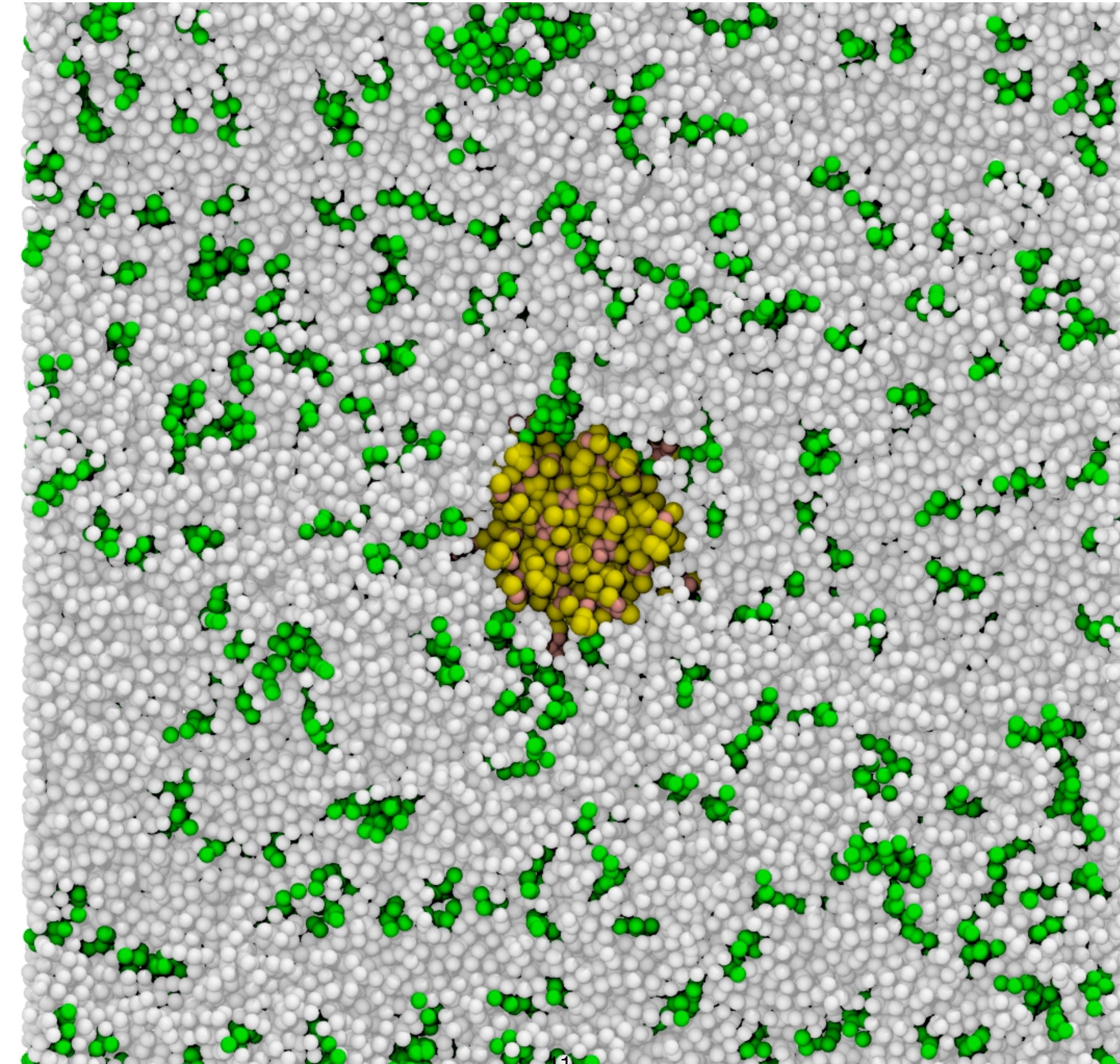
**Research Question: Does  
Ceramide Bind to BamA?**

# How do we study nanoscale events at high resolution?

85:15 POPE:POPG

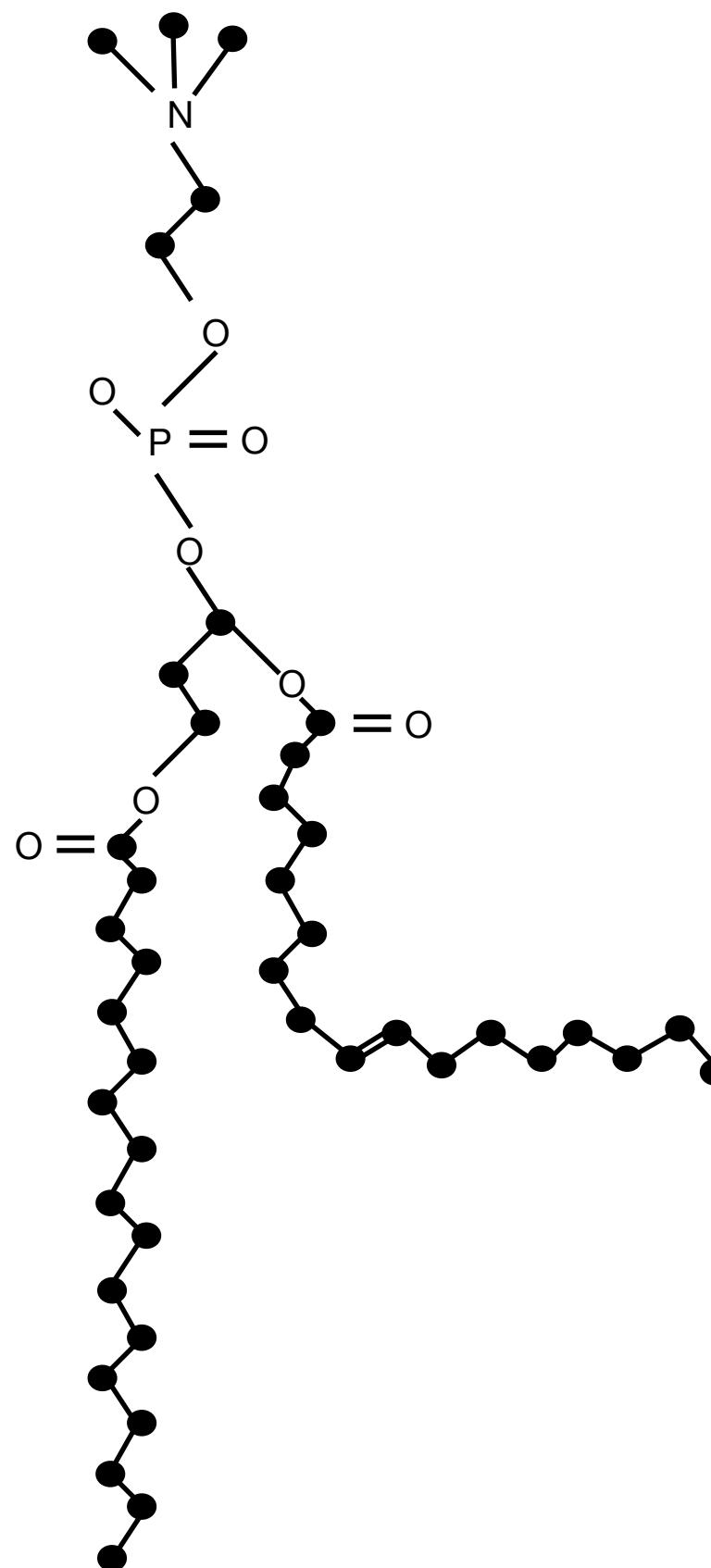
Neutral

Anionic



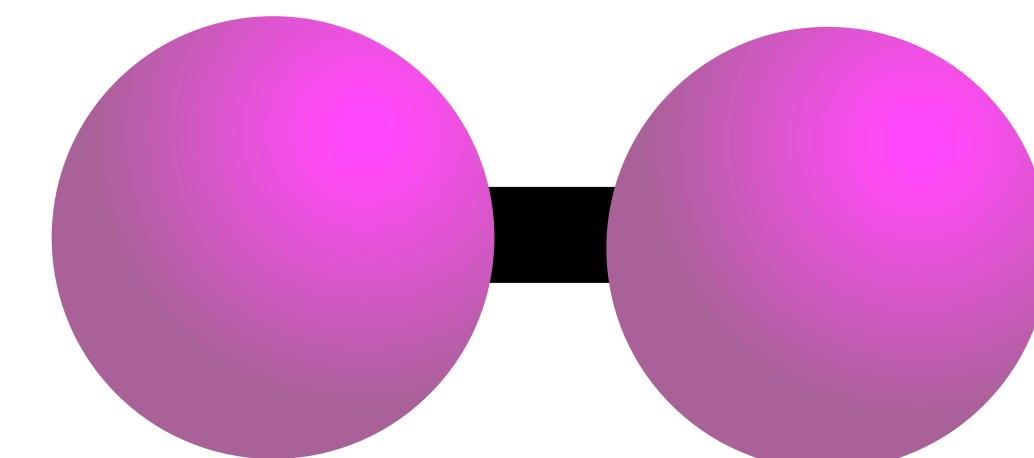
# Molecular dynamics

Structure

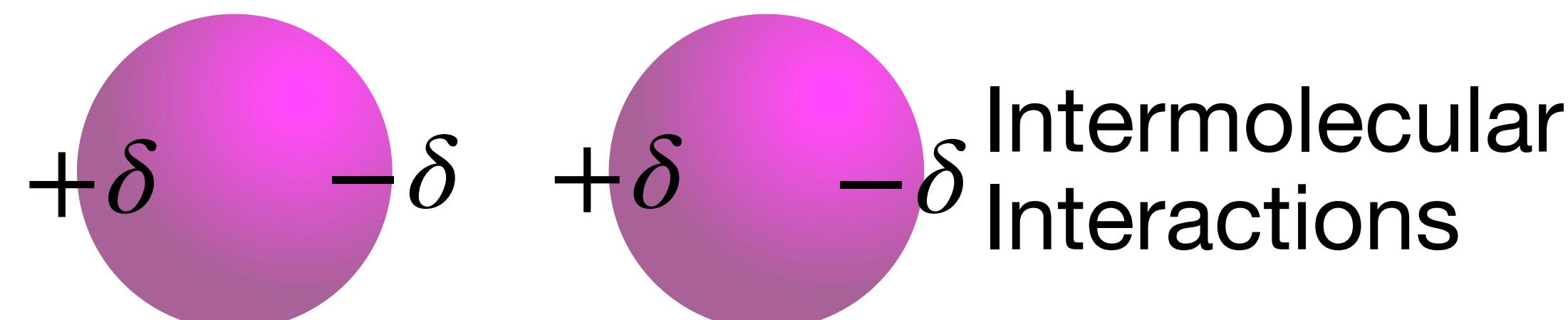


Define  
Interactions for  
all atoms

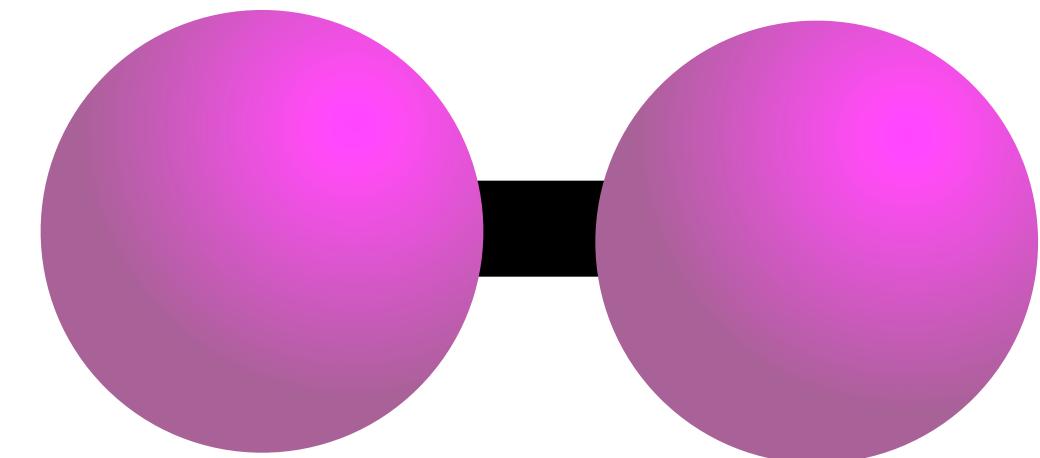
Integrate over  
equations of  
motion



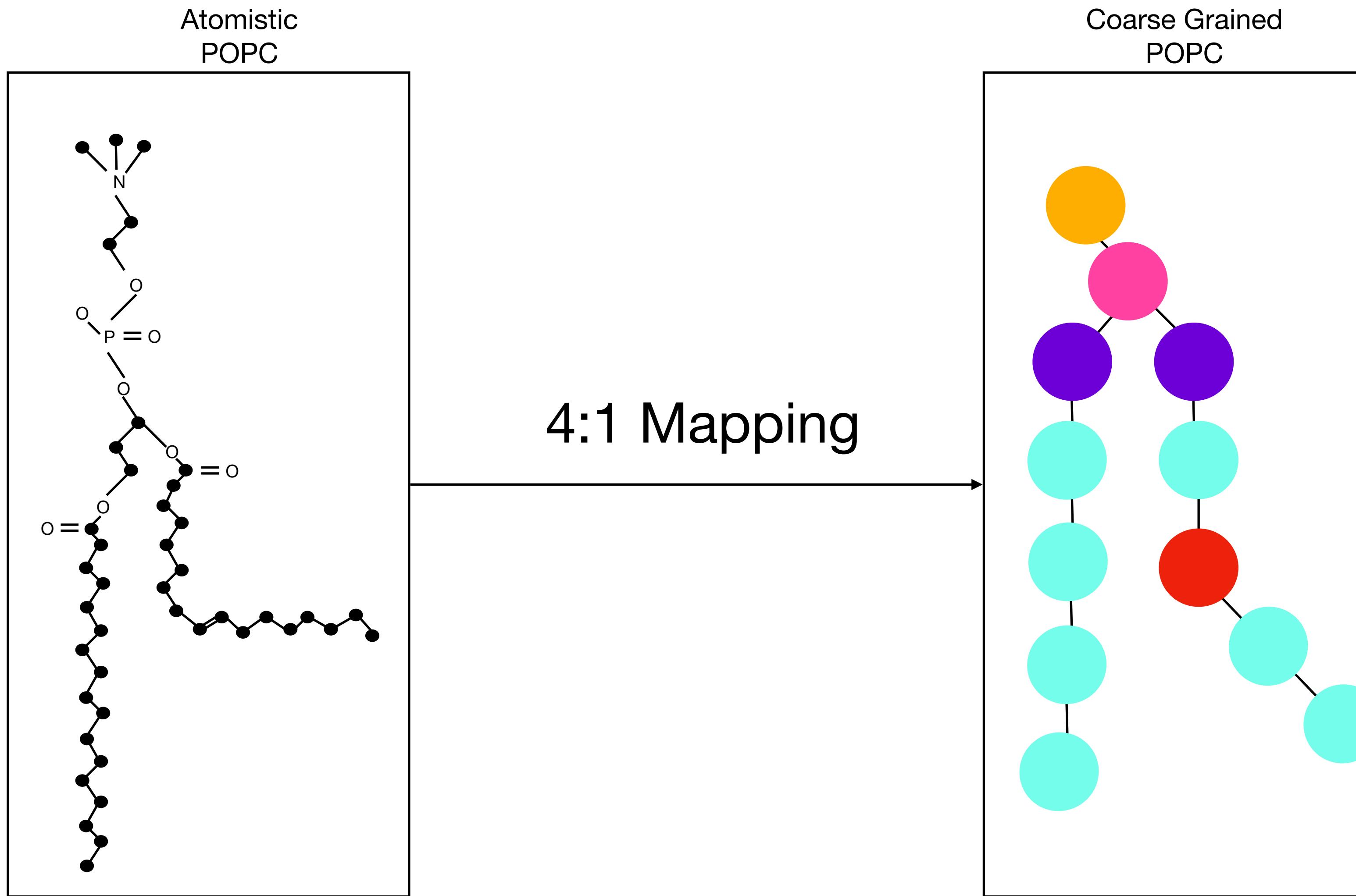
Intramolecular  
Interactions



Intermolecular  
Interactions



# Coarse-grained molecular dynamics

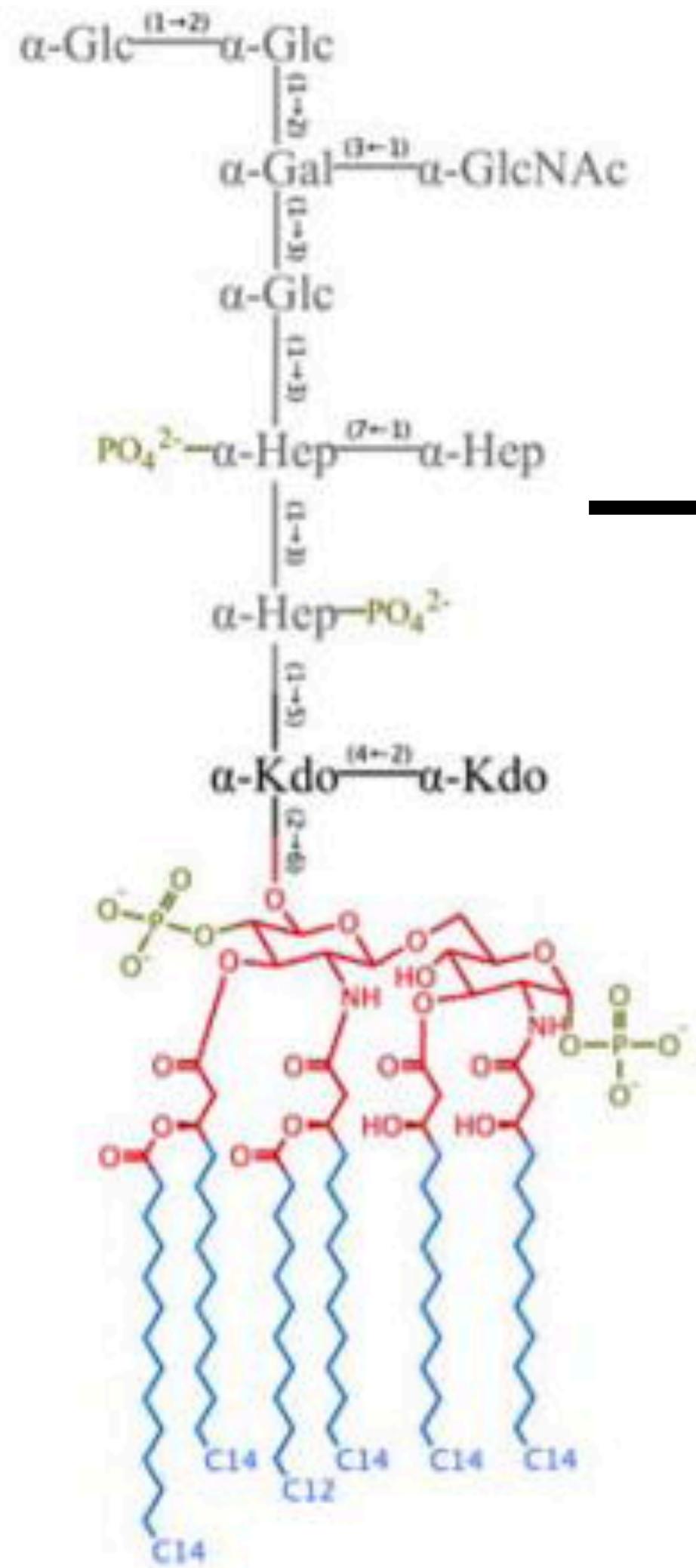


# Outline

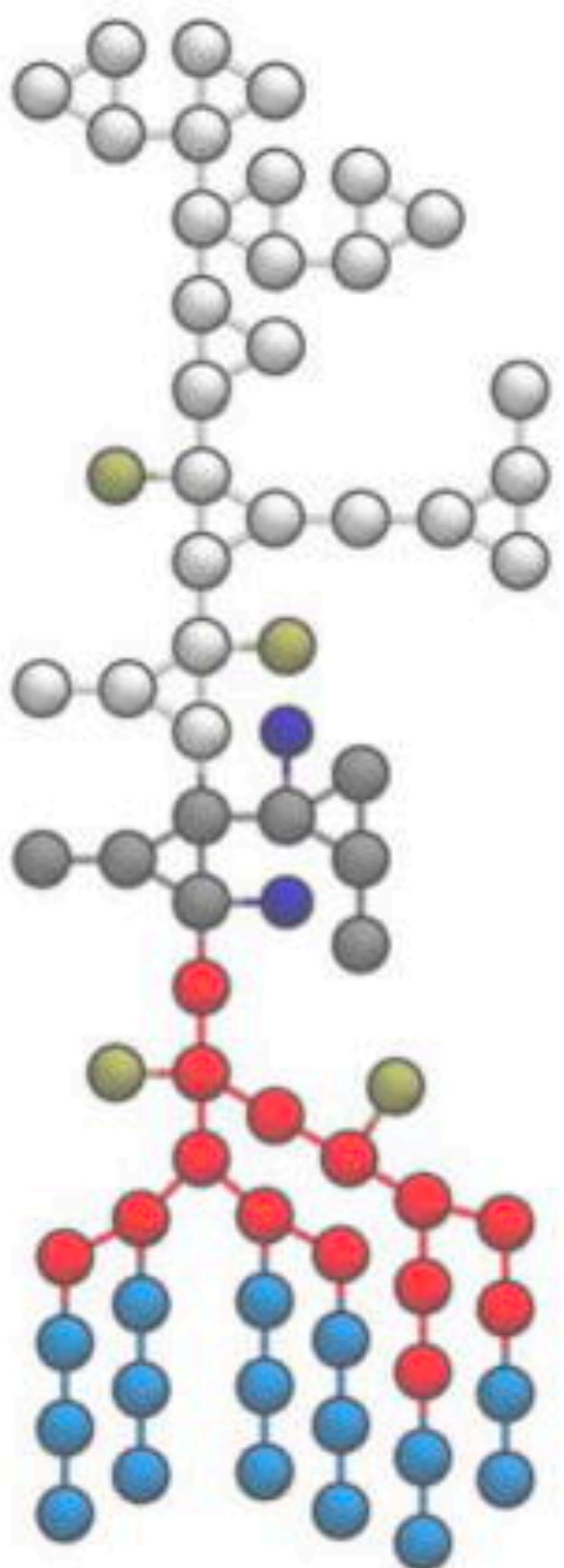
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# Developing a model for *C.Crescentus* membranes

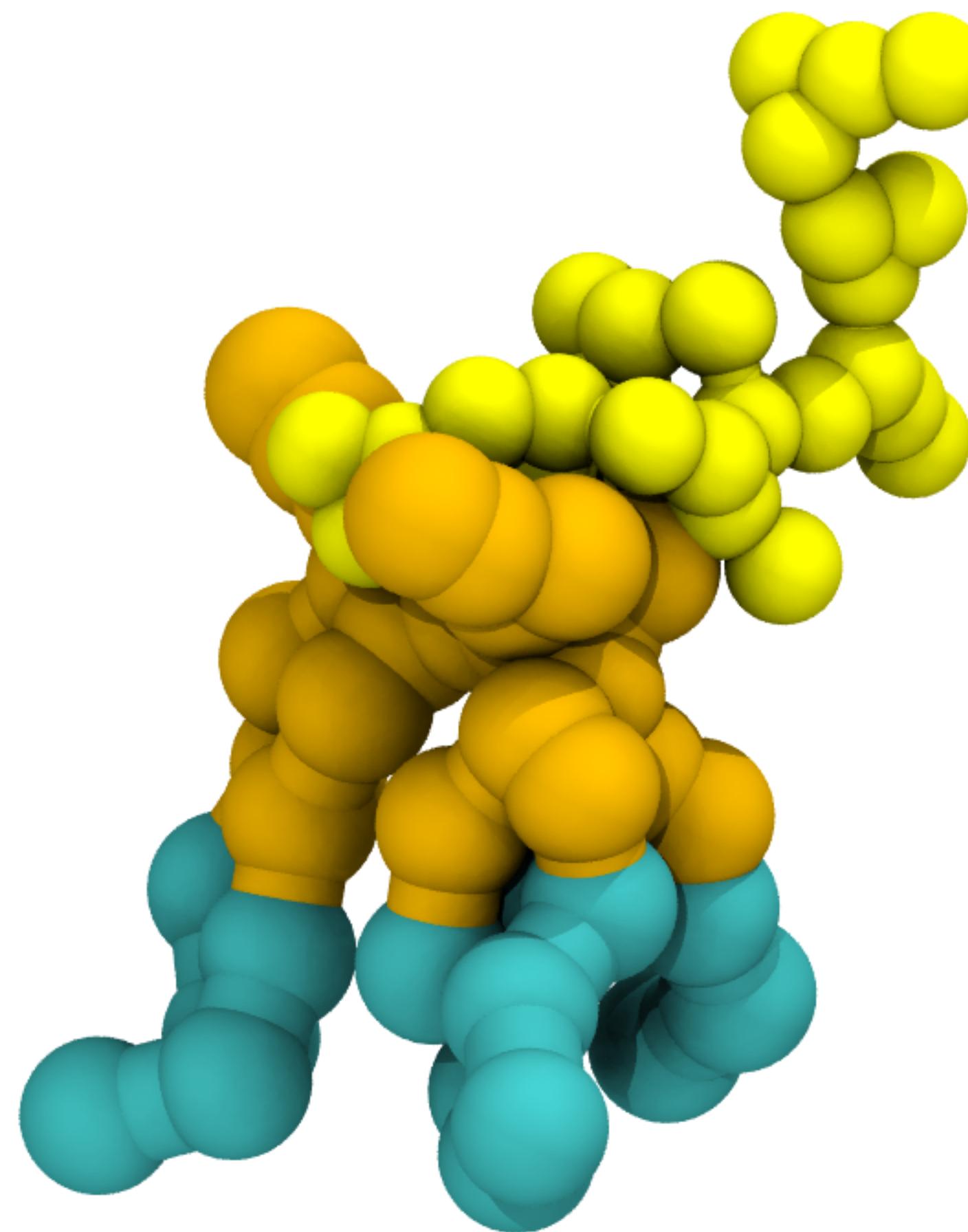
Ecoli LPS



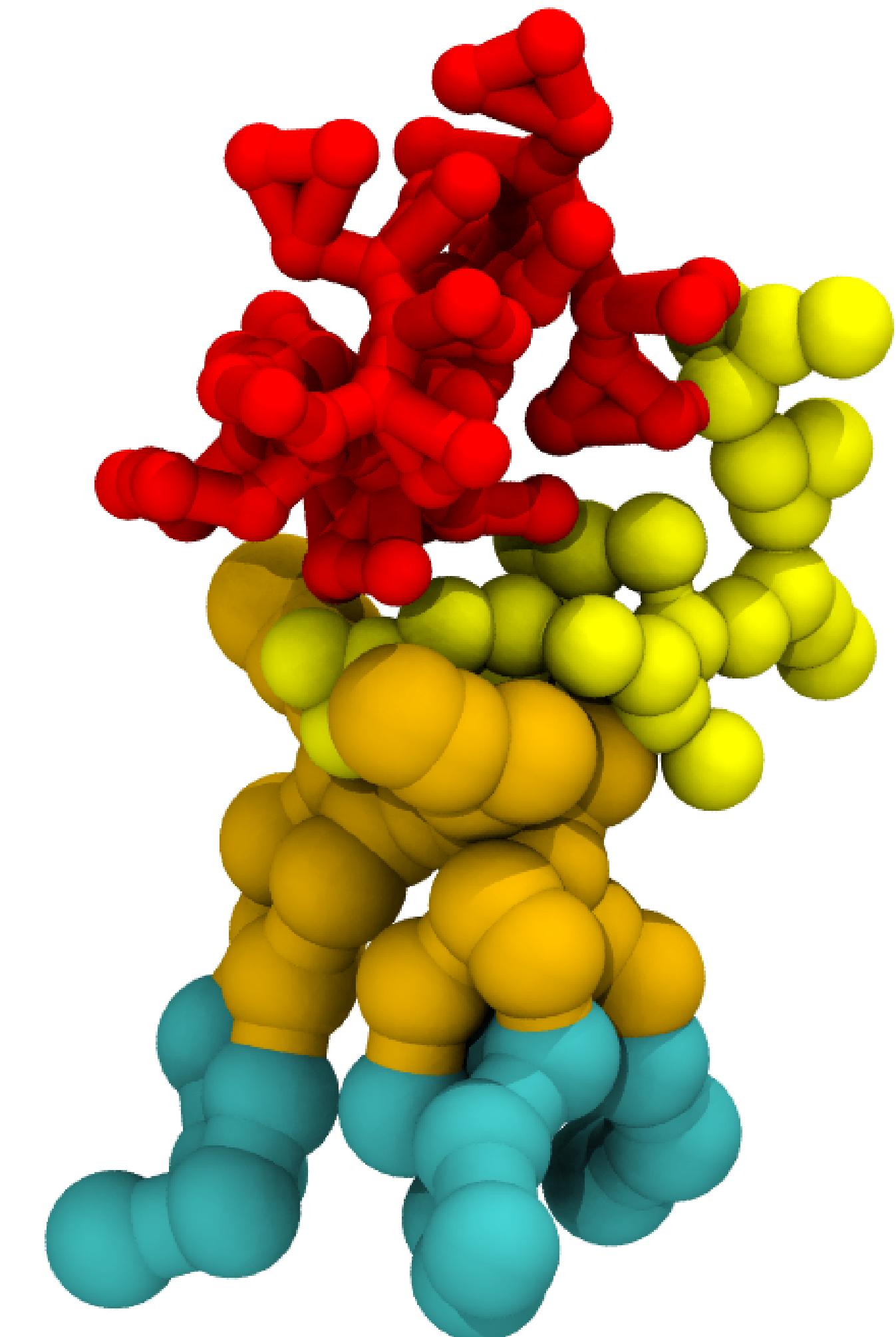
Coarse-Grained Ecoli LPS



Coarse-Grained  
Caulobacter RLPS

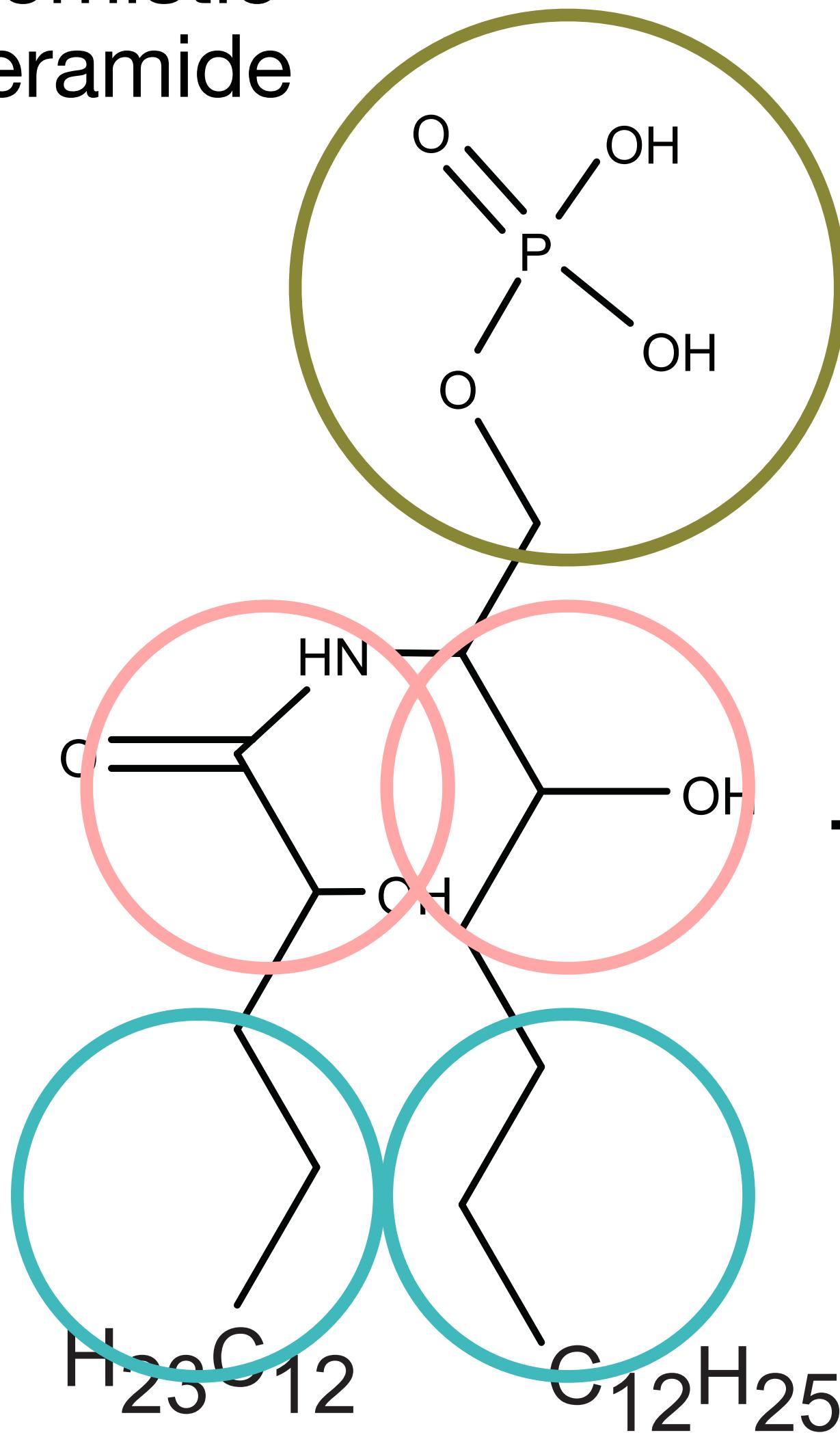


Coarse-Grained  
Caulobacter SLPS

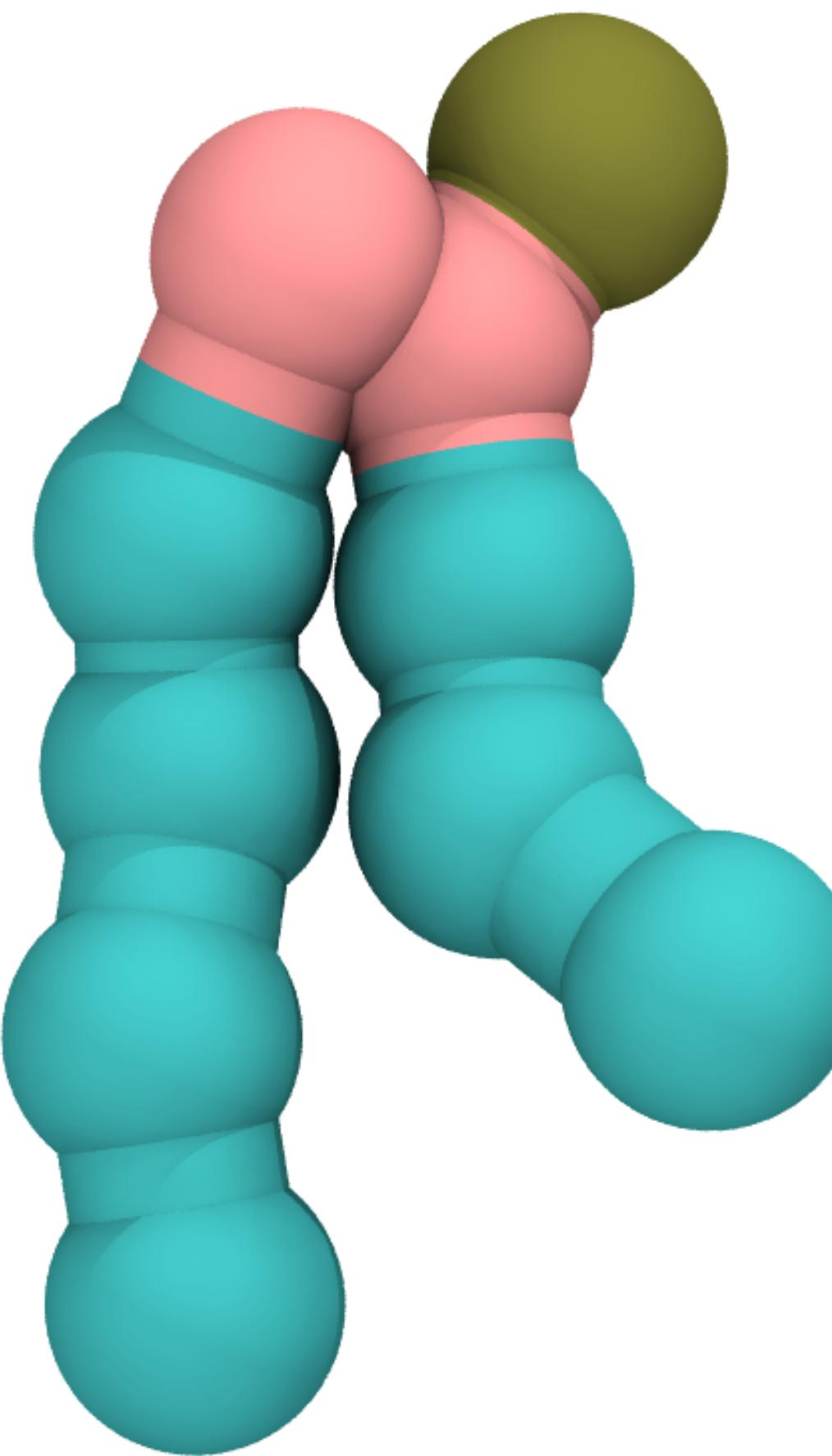


# Developing a model for *C.crescentus* membranes

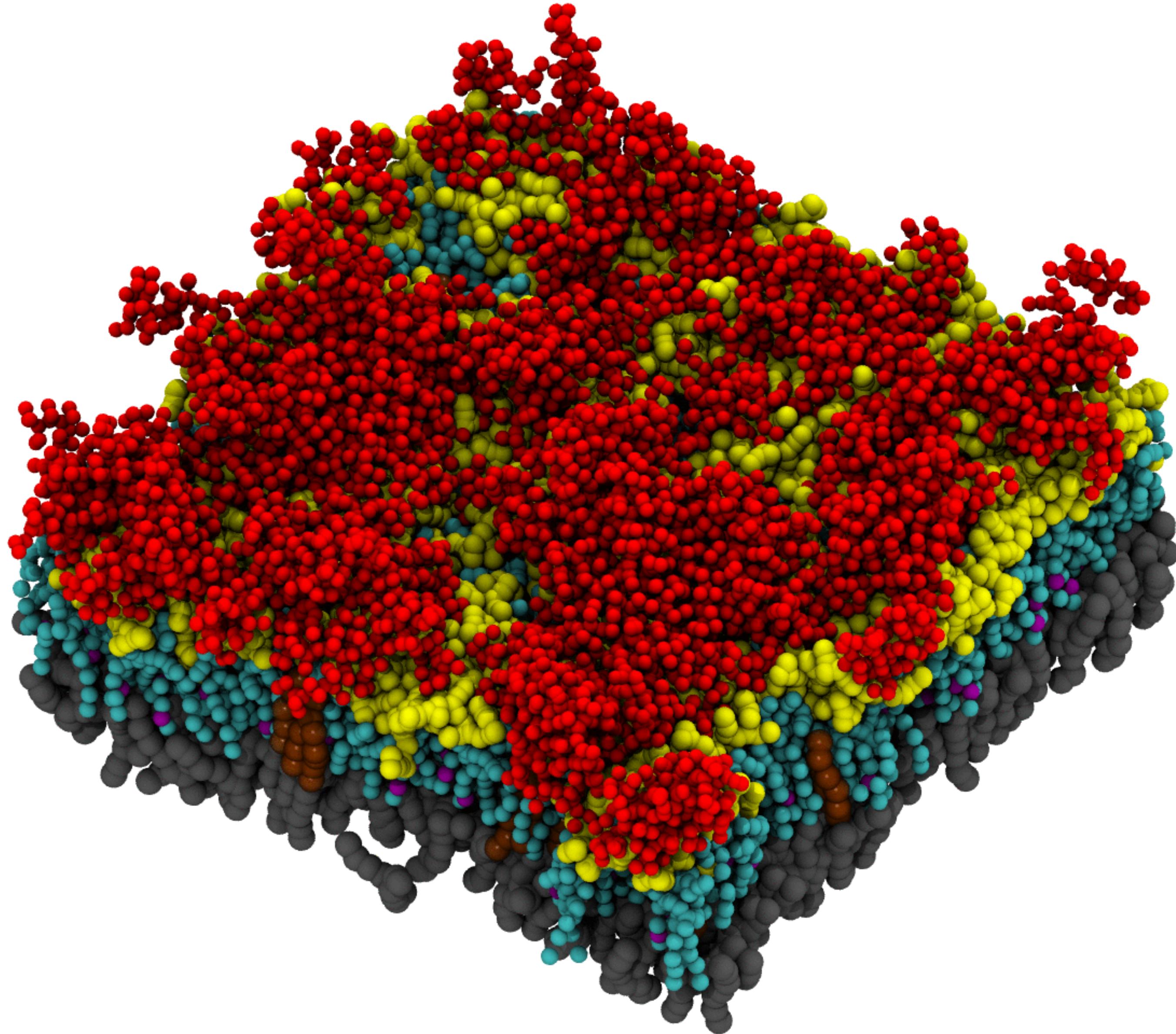
Atomistic  
Ceramide



Coarse-grained  
Ceramide



# C.crescentus membrane model

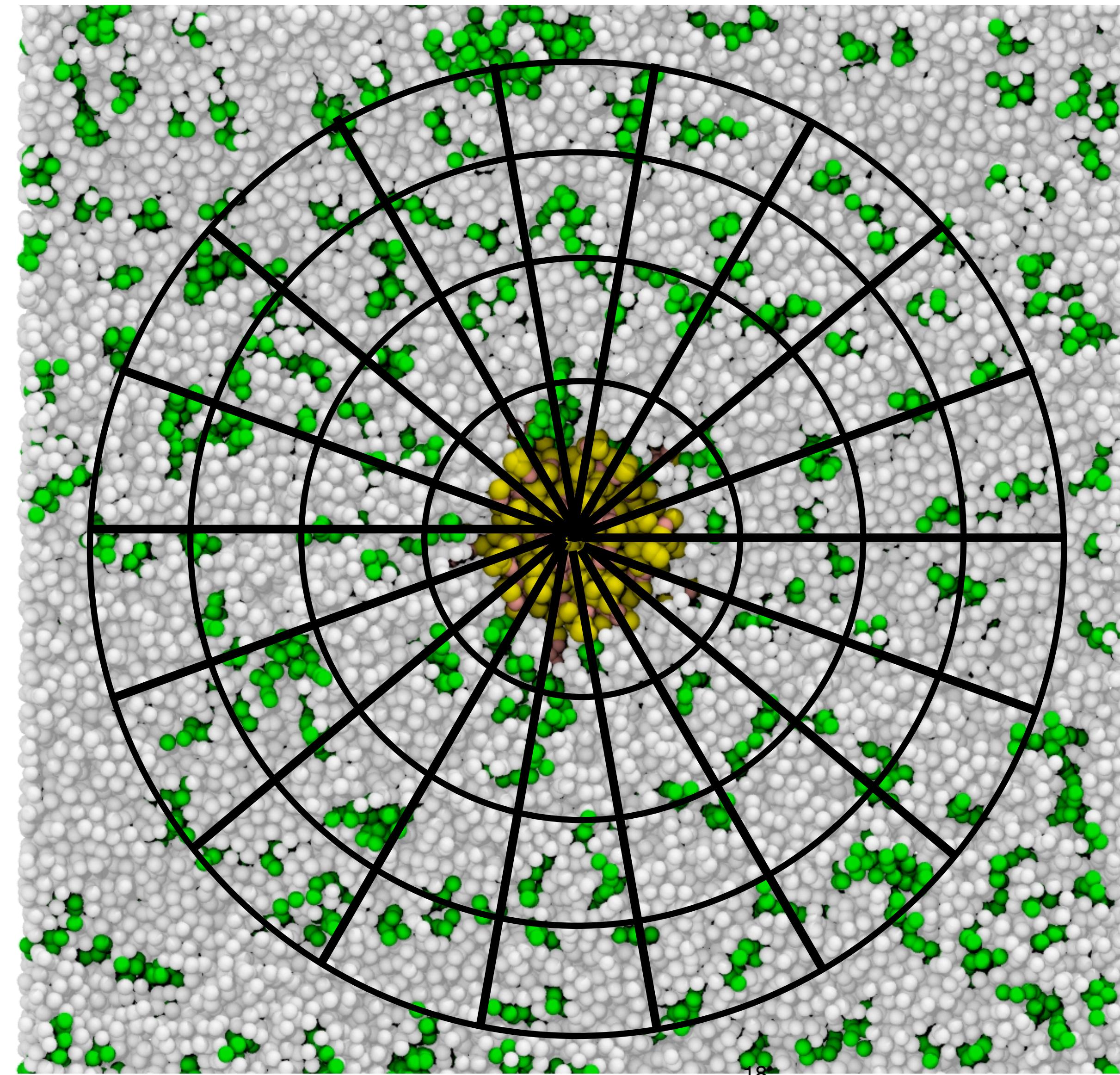


# Can we now study lipid binding?

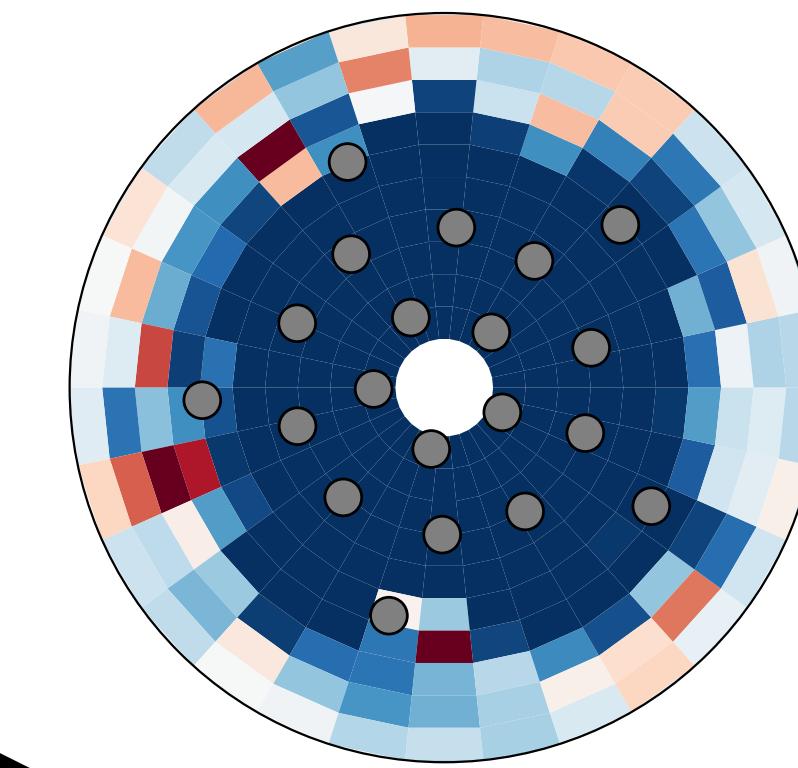
85:15 POPE:POPG

Neutral

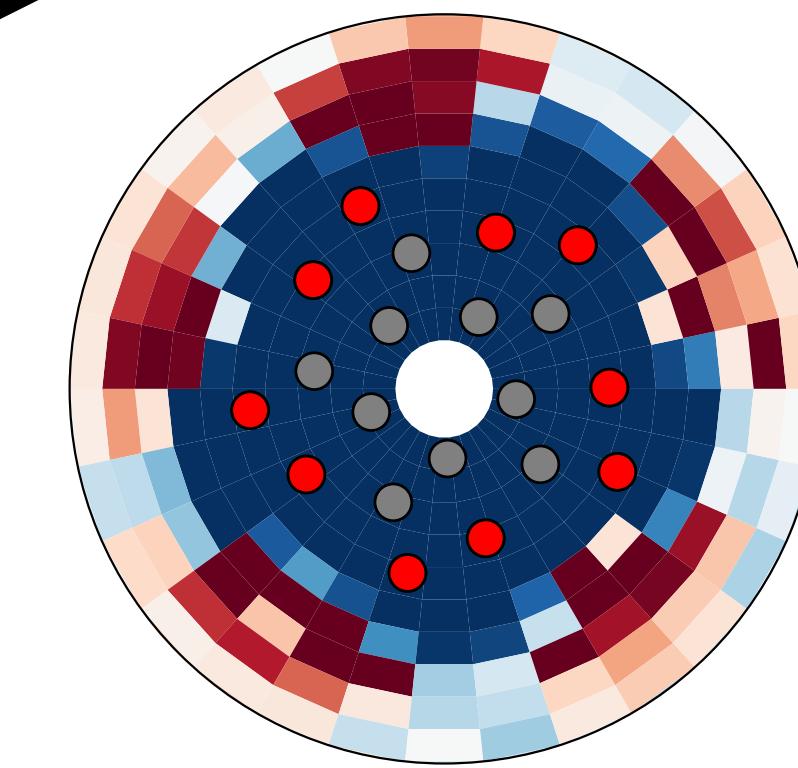
Anionic



PG Density

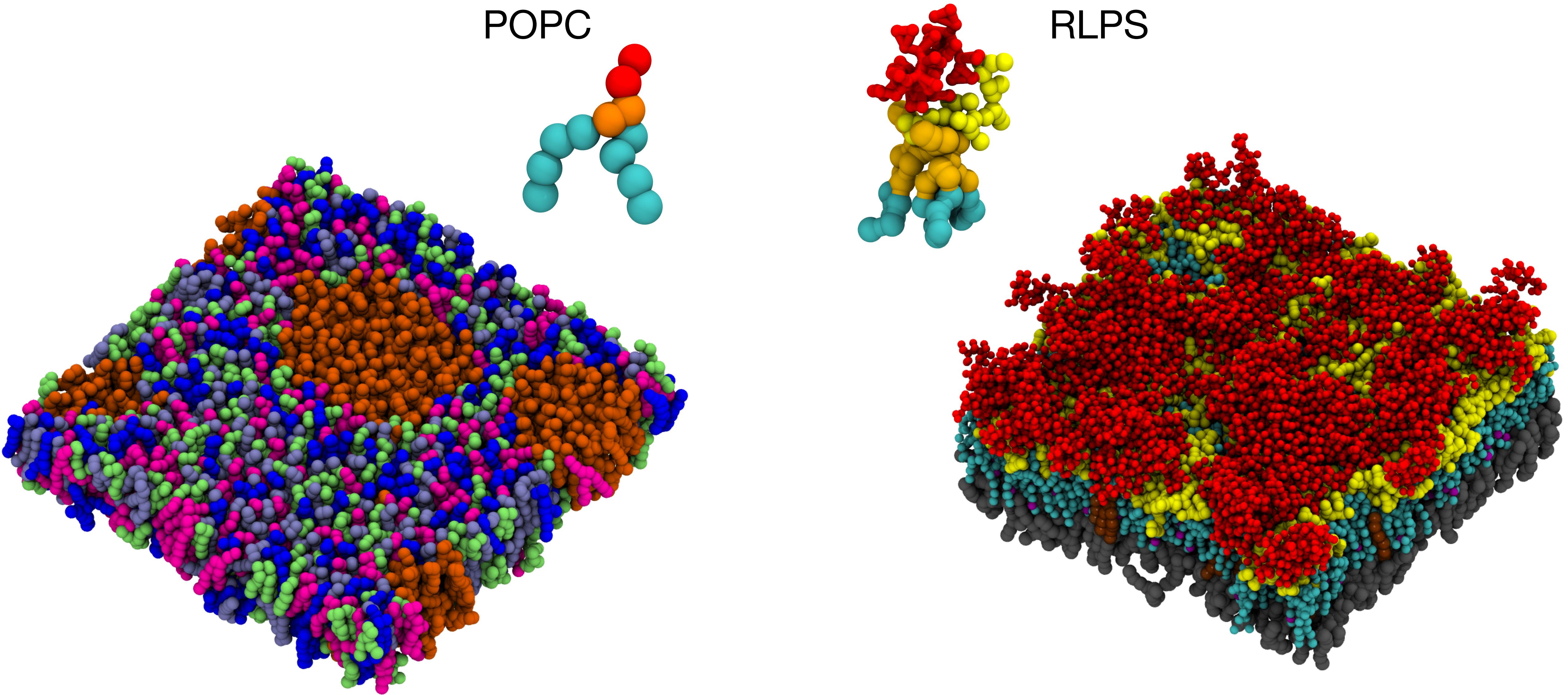


Symmetric Leaflets



Few PG      Many PG

Maybe, bacterial membranes diffuses on long time scales, is this a problem for studying lipid binding?



# Summary & Future Directions

- Summary
  - Developed a Caulobacter membrane model
- Future Directions
  - Long coarse-grained simulations to identify time scale of ceramide diffusion in bacterial membranes
  - Develop a homology model for Caulobacter BamA
  - Simulate BamA in Caulobacter membrane
  - Calculate ceramide density around BamA

# Acknowledgments



## PI

- Dr. Grace Brannigan
- Dr. Eric Klein

## Lab Members

- Jesse Sandberg
- Ezry St. Iago-McRae
- Connor Pitman
- Regina Salzer

## Former Lab Members

- Anushriya Subedy

# Questions?