The Bacterial Brain: All-atom description of a bacterial chemoreceptor array.

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Overview

- Introduction.
- Bacterial chemotaxis primer: The case of *E. coli.*
- The Bacterial Brain: A naturally-evolved, mechanical computer.
- How computation and Blue Waters can help.
- Modeling and Simulation of the chemoreceptor array.

Chemotaxis: A fundamental sensory phenomenon.

How do sperm cells find eggs?

How do white blood cells find sites of inflammation?

How do bacteria find food and avoid poisons?

Chemotaxis - Any cell motion affected by a chemical gradient, resulting in net propagation along the gradient.

Neutrophil “chasing” a bacterium.
David Rogers, Vanderbilt U. (1950’s)

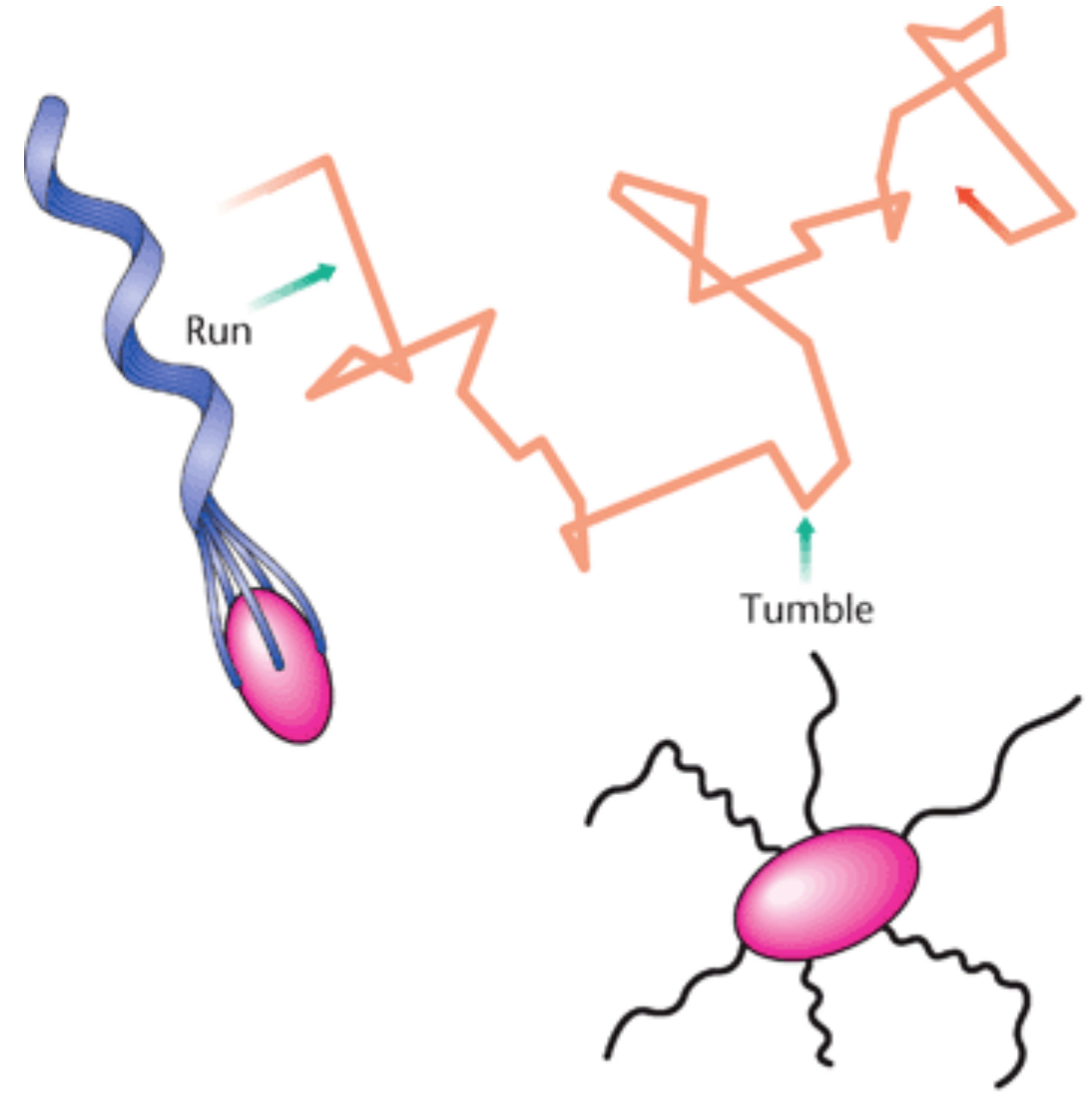
Bacterial chemotaxis primer: The case of *Escherichia coli.*

Bacteria sense a wide range of environmental chemicals.

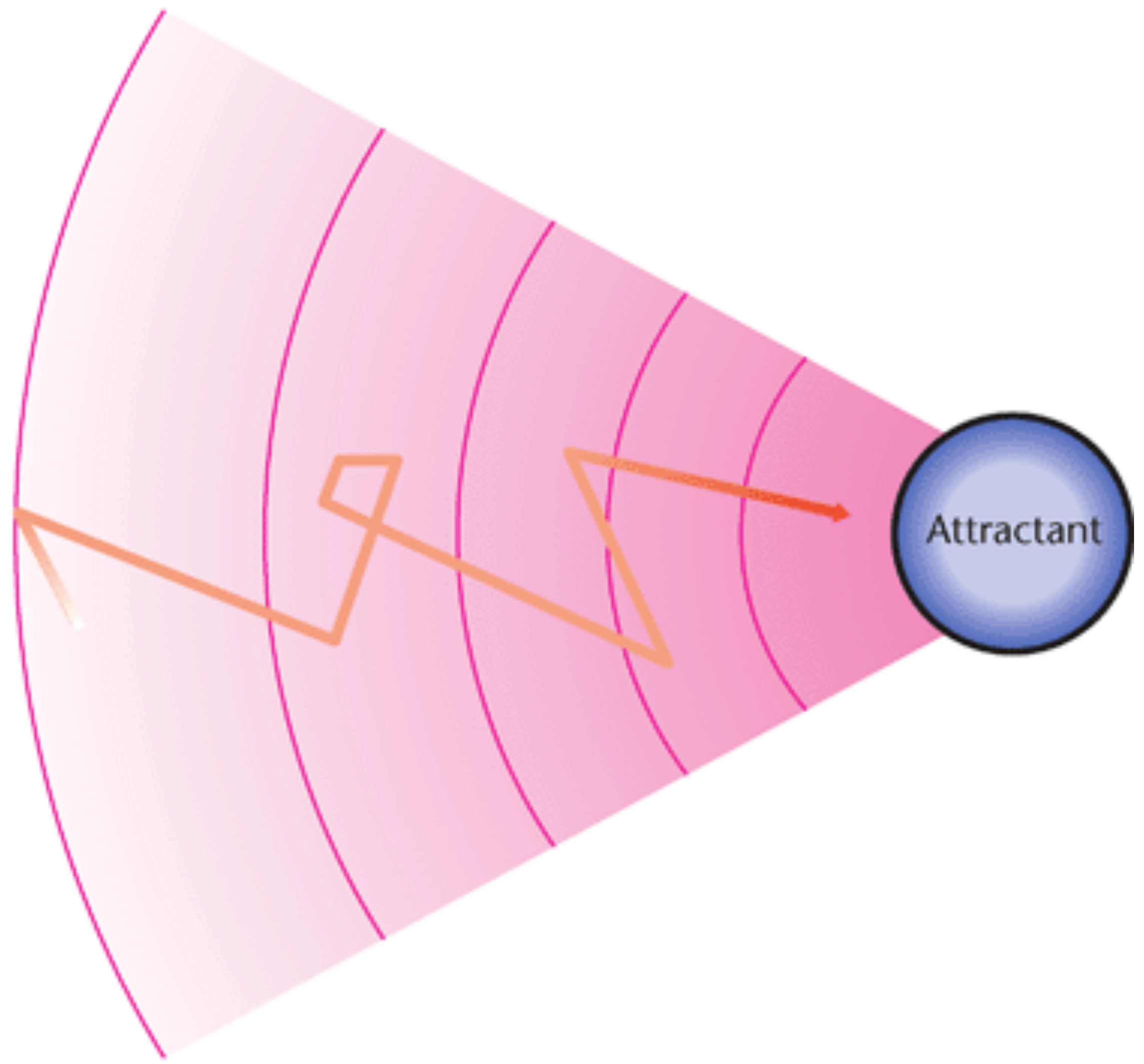
This information is used to generate motile responses that place cells in the optimal habitat.

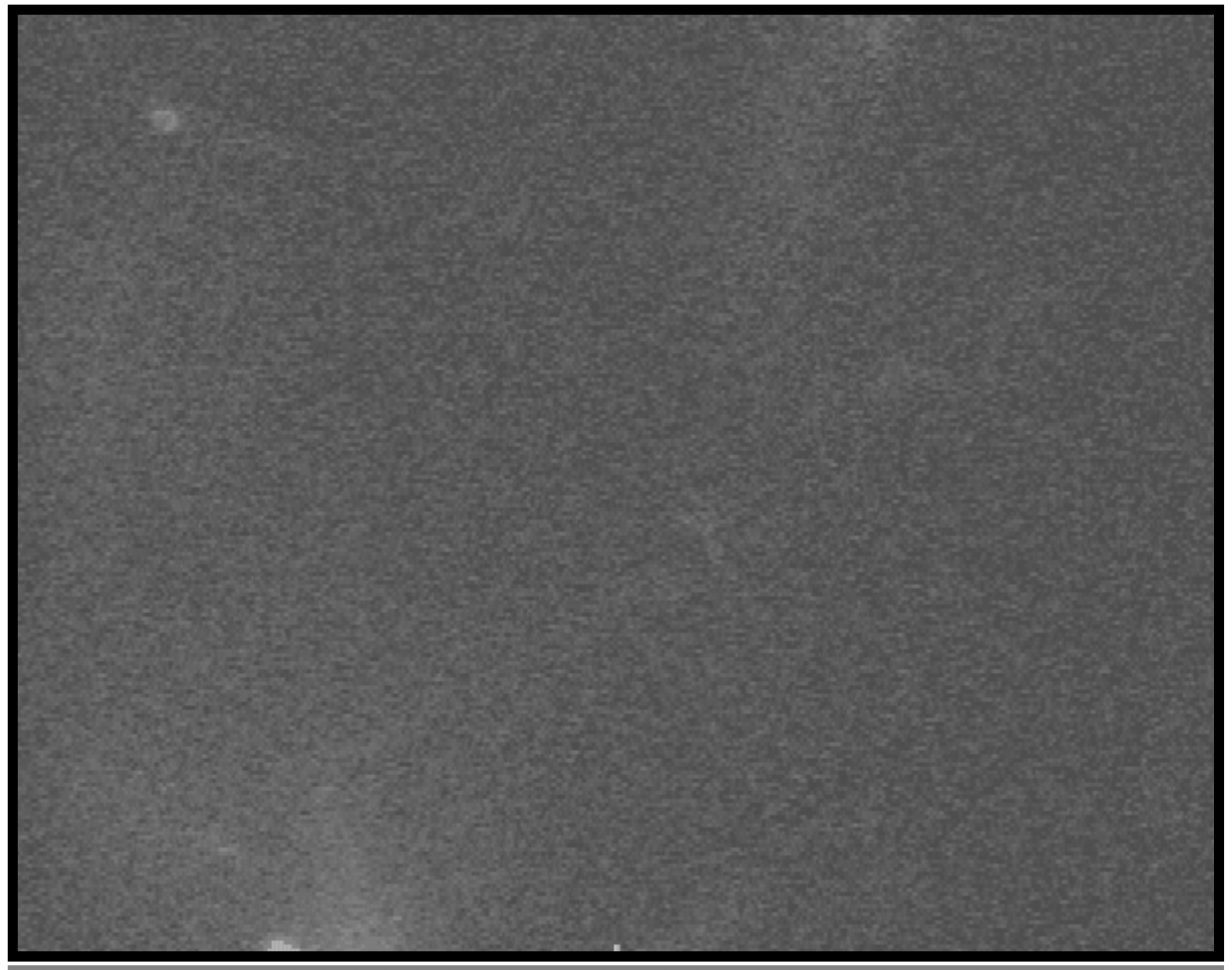
E. coli, uses a run-tumble strategy, lengthening runs in “good” directions.

No gradient = random walk



Attractant gradient = biased random walk





Free-swimming *E. coli*

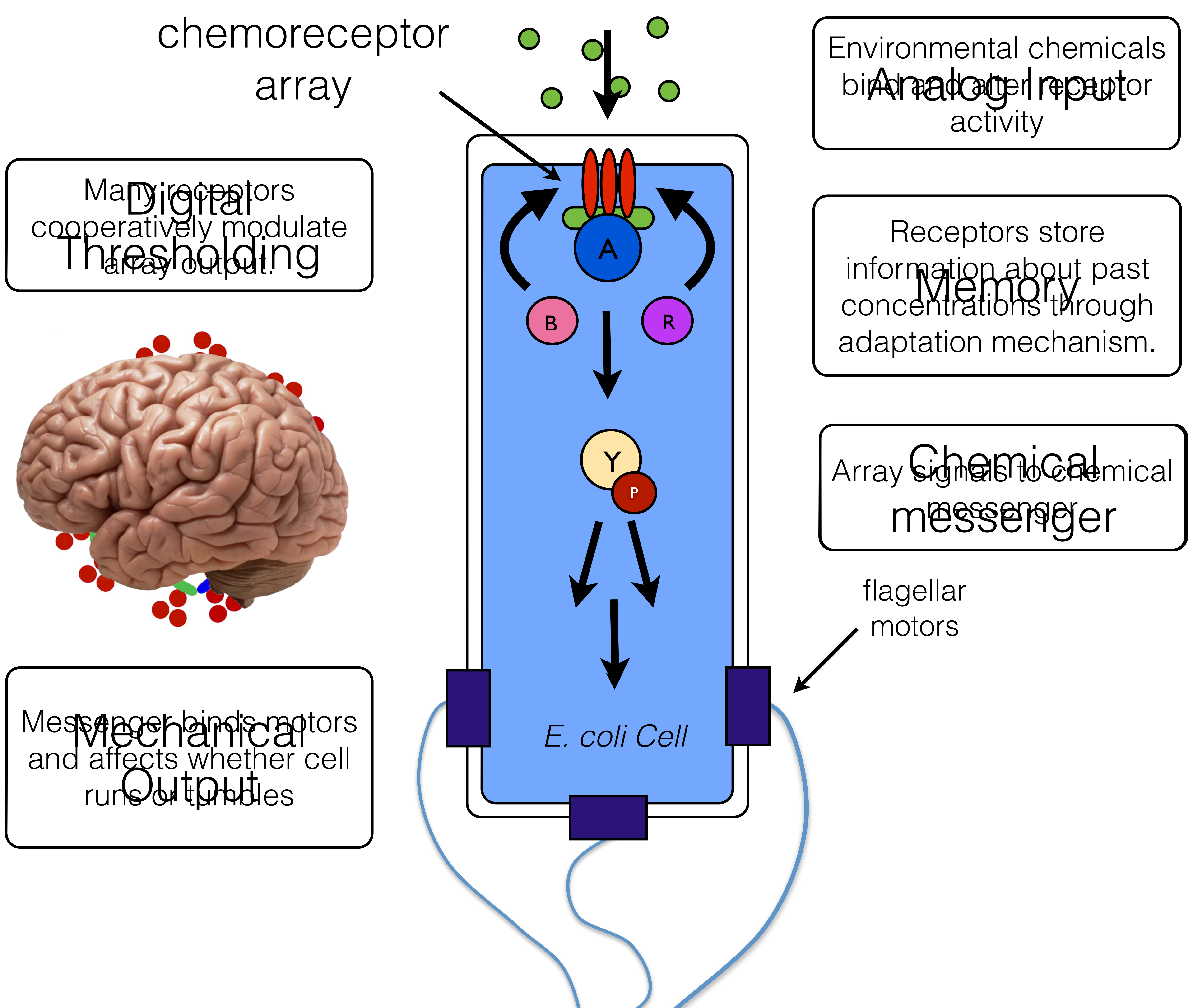
Figure: Eisenbach, Michael(Dec 2011) Bacterial Chemotaxis. In: eLS. John Wiley & Sons Ltd, Chichester. http://www.els.net

Video: Berg, Howard (Harvard U.) http://www.rowland.harvard.edu/labs/bacteria/movies/ecoli.php

To tumble, or not to tumble?

The *E. coli* chemotactic network.

Most thoroughly studied sensory signal transduction system in biology…



The network is sophisticated!

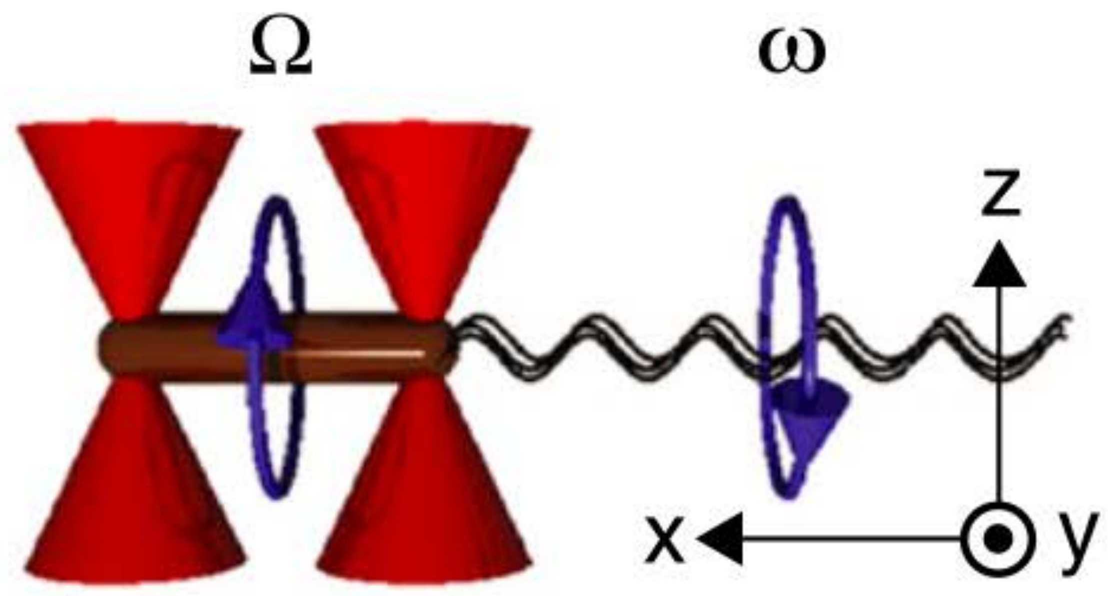
(I) ***Ultra-Sensitivity*** - Detects gradients as little as three molecule change per cell volume.

(II) ***High Gain*** - Cells can amplify stimuli over 50-fold.

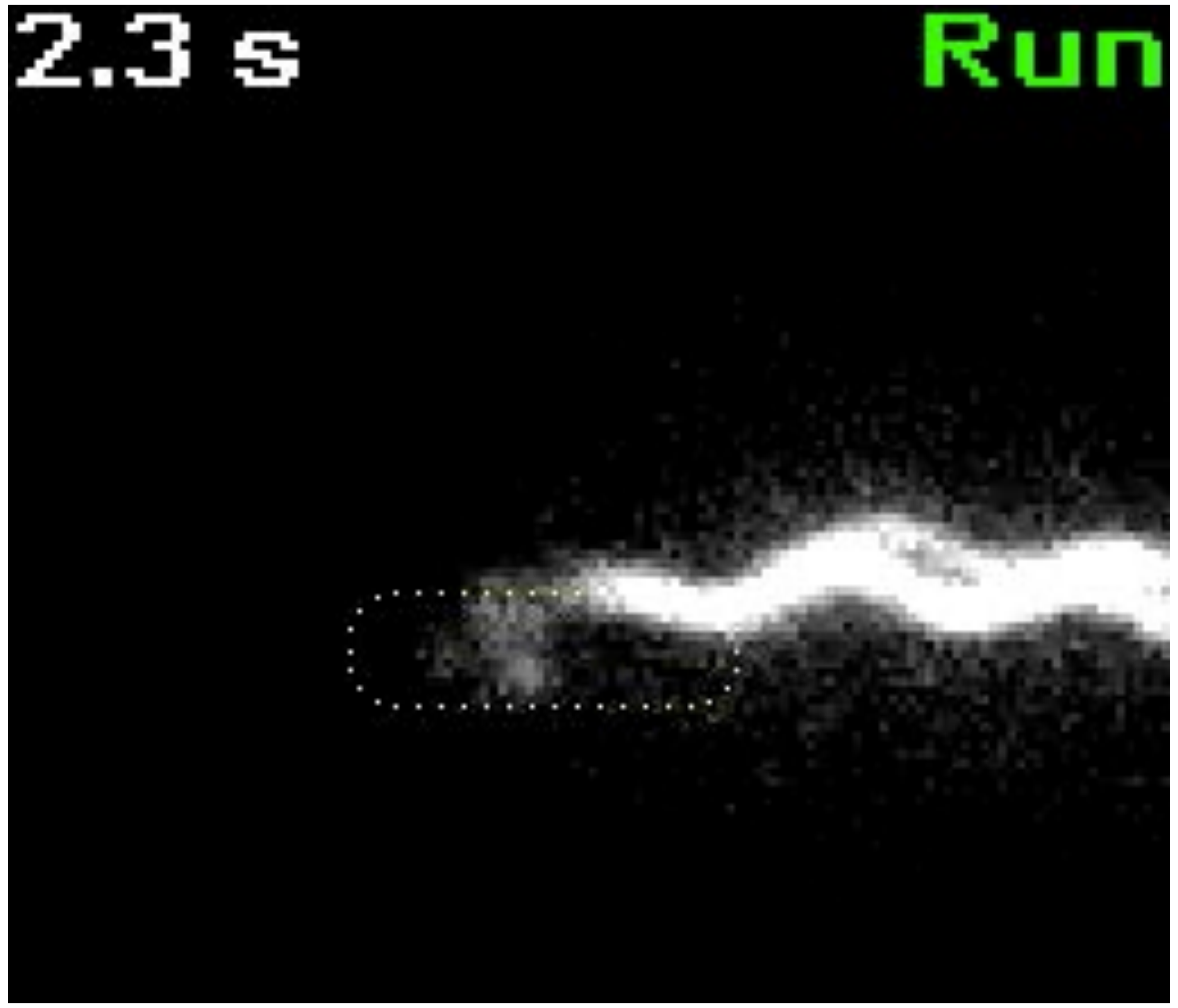
(III) ***Precise adaptation*** - Extends the range of concentrations that can be discriminated to five orders of magnitude.

Experiements and quantitative modeling studies point to receptor clustering within the array to explain enhanced signaling features.

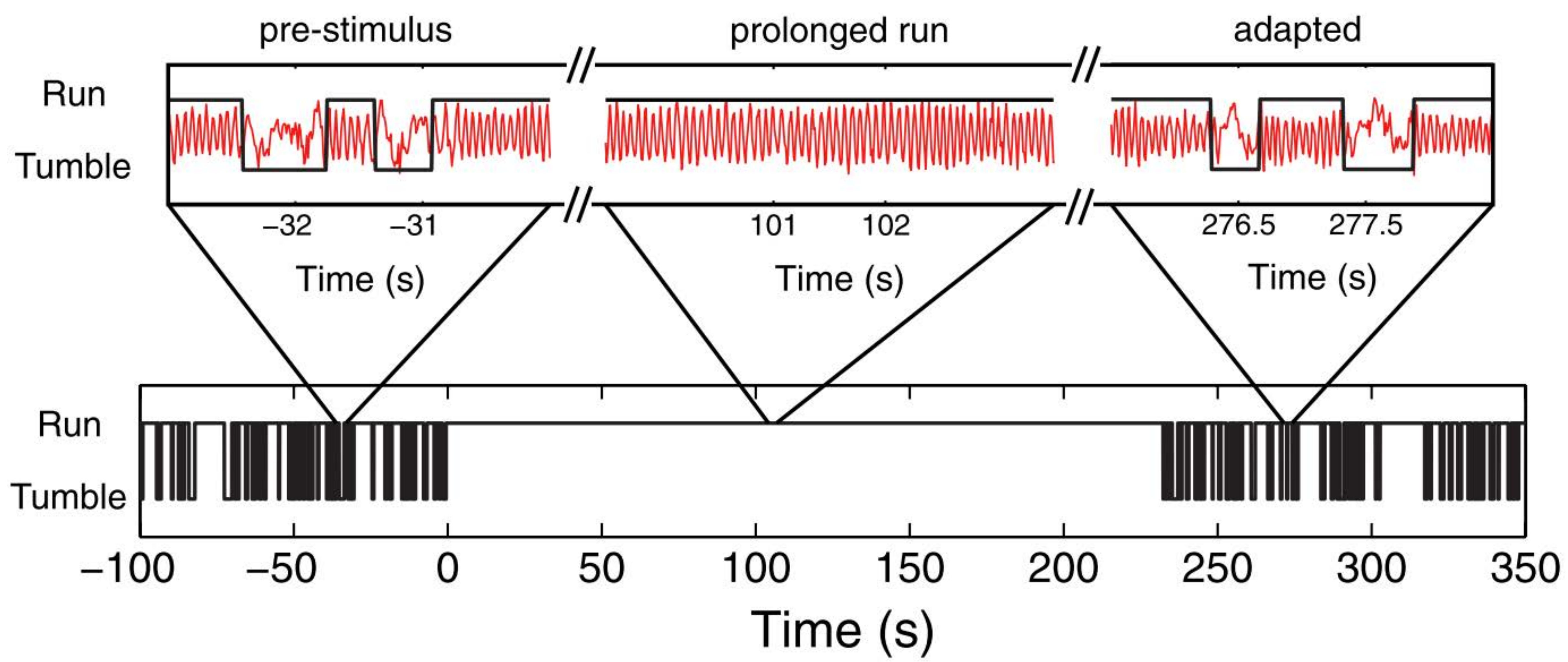
Recent work from Chemla Group.



Optical traps characterize swimming behavior of single cells.



Individual run-tumble events are tracked revealing “abruptness” of adaptation.

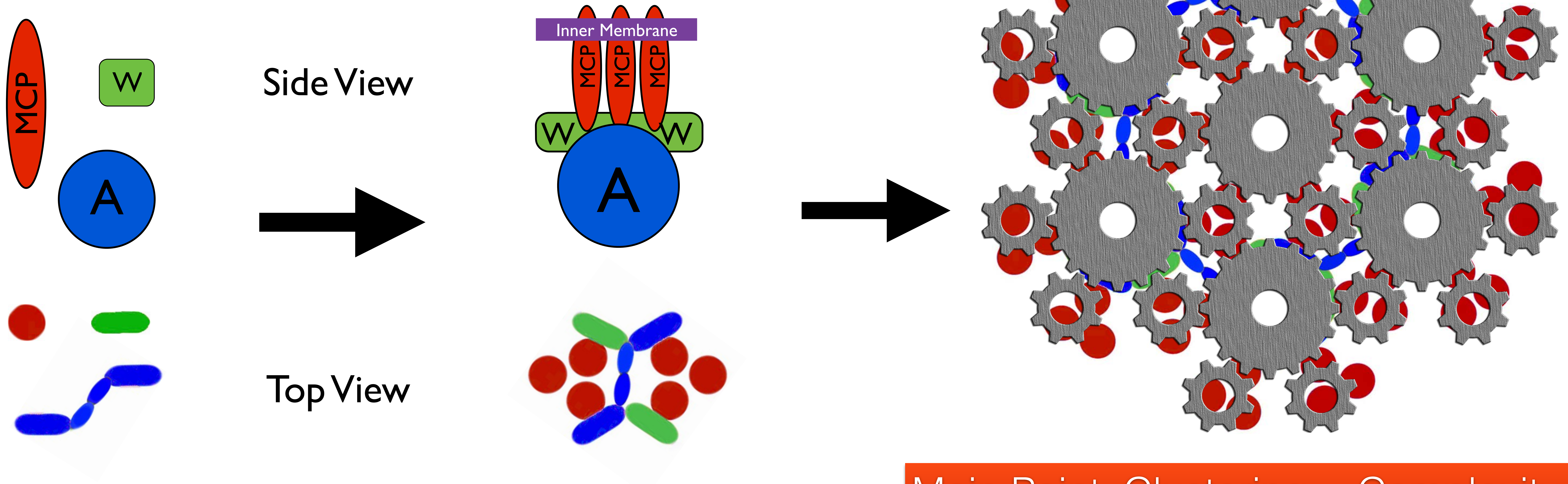


Mathematical modeling and stochastic simulations suggest this characteristic adaptation response originates from clusters of interacting receptors.

Min, T. L. et. al. *Chemotactic adaptation kinetics of individual Escherichia coli cells*. Proc. Natl. Acad. Sci. USA (2012),109(25), 9869–74.
Mears, P. et. al. *Escherichia coli swimming is robust against variations in flagellar number.* eLife (2014), 1–18.

The Bacterial Brain: A naturally-evolved, mechanical computer.

Thousands of copies of Methyl-accepting chemotaxis proteins + histidine kinases + adaptor proteins cluster to form the chemoreceptor array.

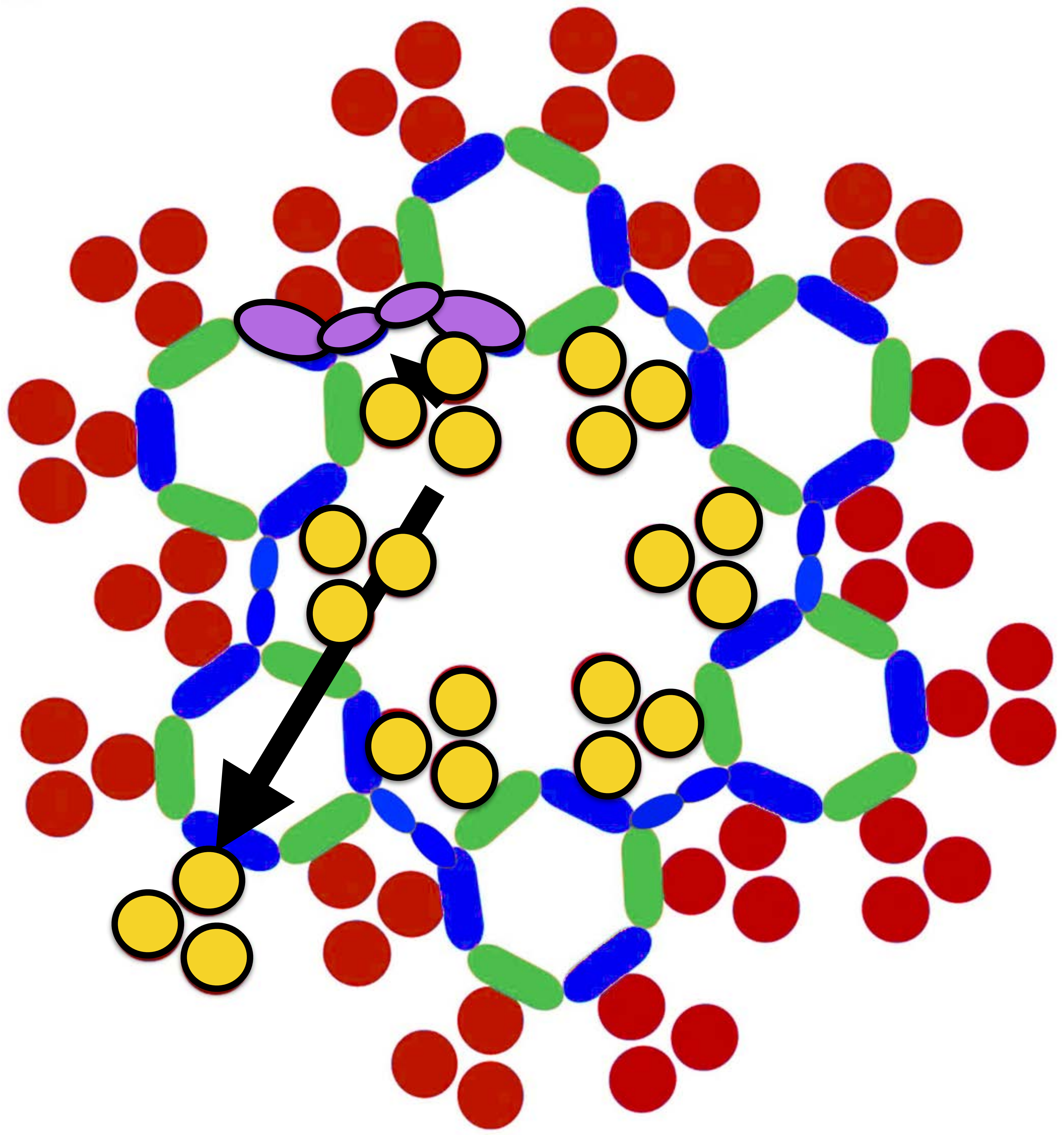


Main Point: Clustering = Complexity & Irreducibility

Average sized array contains *~15,000 proteins* with surface area *~250x250 nm**2.*

Briegel, A. et. al. *Structure of bacterial cytoplasmic chemoreceptor arrays and implications for chemotactic signaling.* eLife (2014).

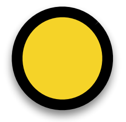
The Bacterial Brain: A naturally-evolved, mechanical computer.

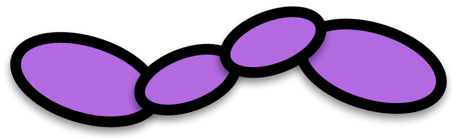


Key problem: What are the ***molecular origins*** of the enhanced information processing and distinctive signaling features of bacterial chemotaxis?

How does clustering lead to robust and cooperative signal transduction?

How does clustering lead to the characteristic signal regulation seen in Chemla experiments?

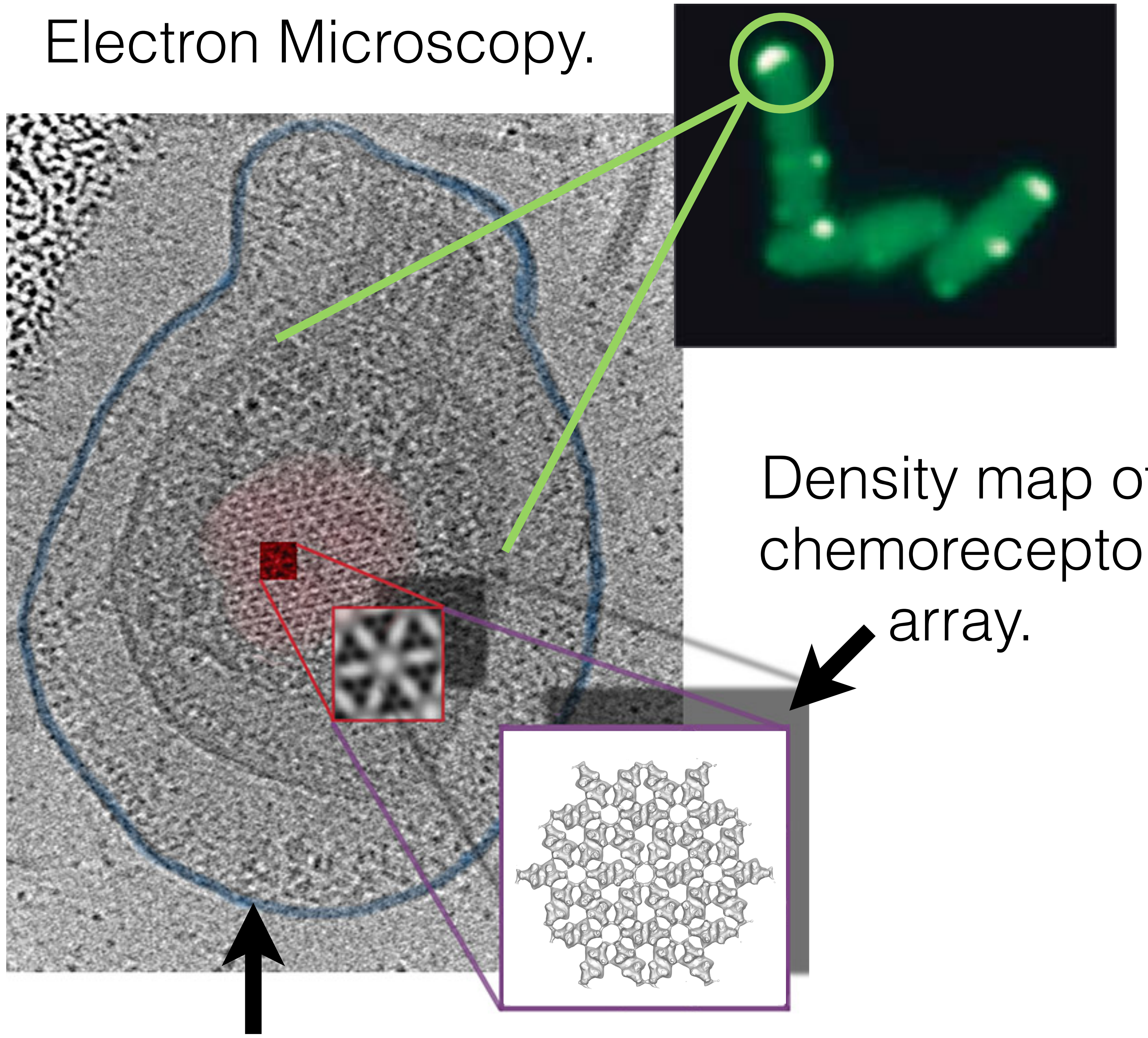

“activated” chemoreceptor


“activated” CheA

The Bacterial Brain: A naturally-evolved, mechanical computer.

Experiments have provided a lot of structural data…

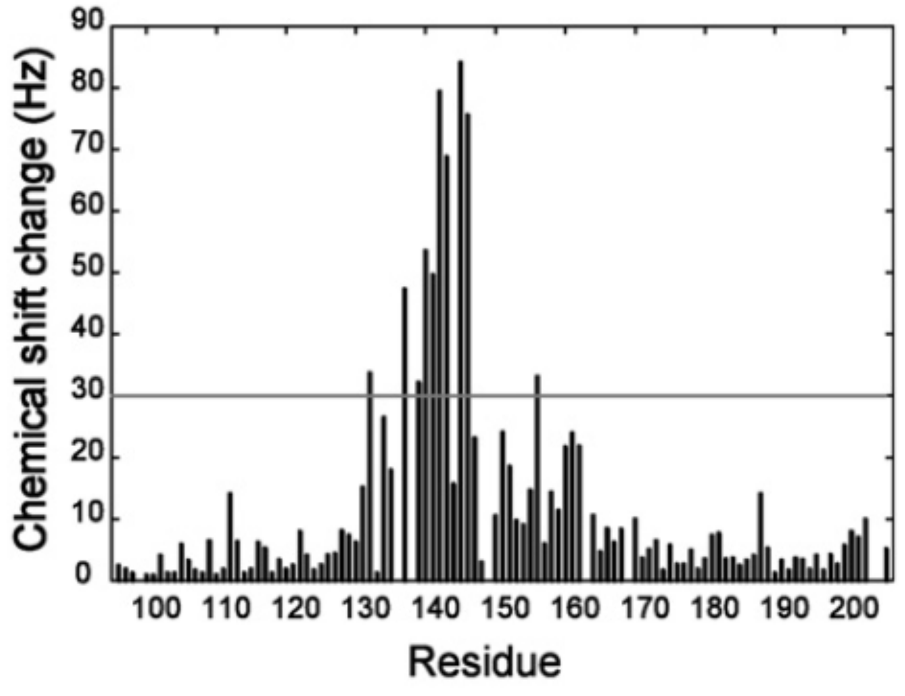
Electron Microscopy.

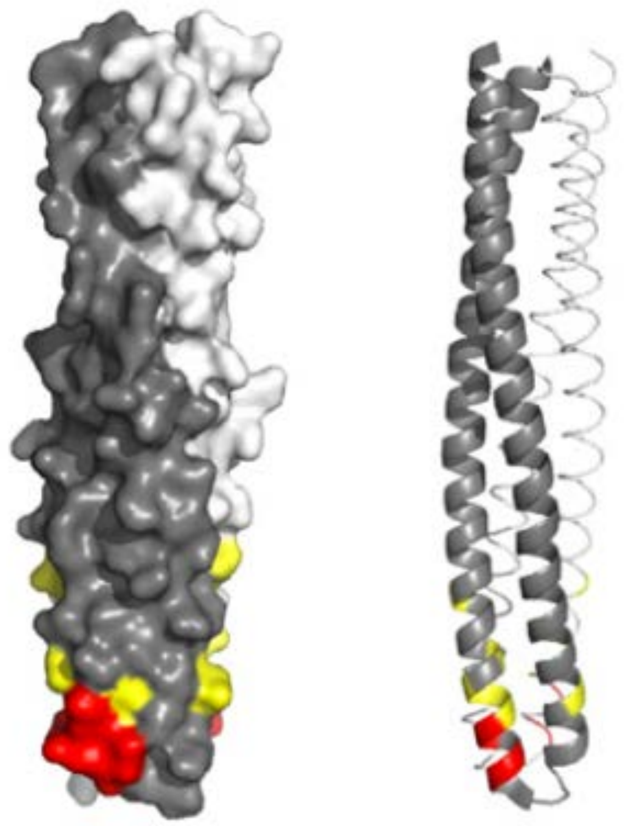


Density map of chemoreceptor array.

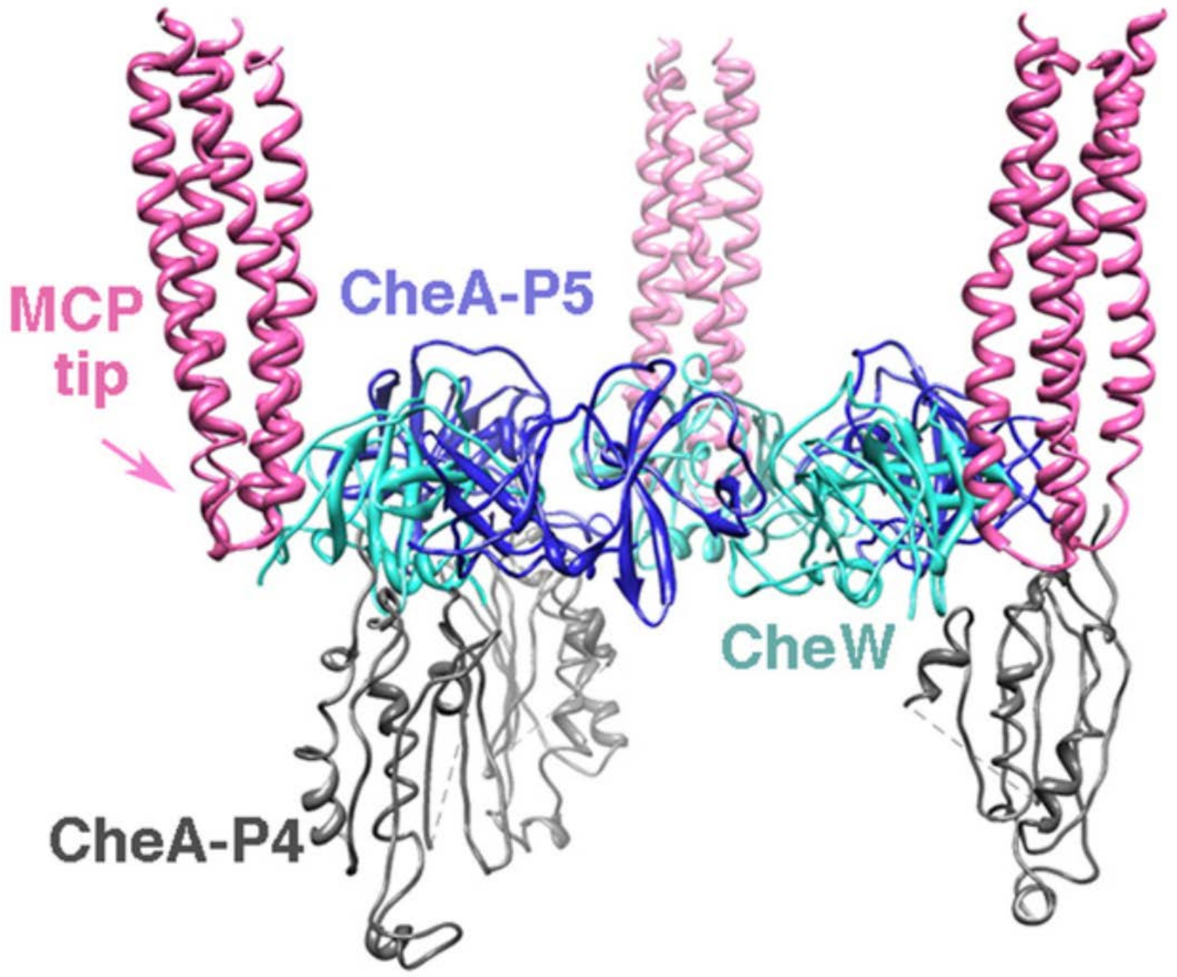
Tomographic slice through top of a bacterial cell.

Biochemical methods.





X-ray crystallography.



Such techniques alone do not currently provide the resolution needed to track structural changes essential to the array's computational ability

Vu, A. et. al. *The receptor-CheW binding interface in bacterial chemotaxis.* J. Mol. Bio. (2012), 415(4), 759–67.

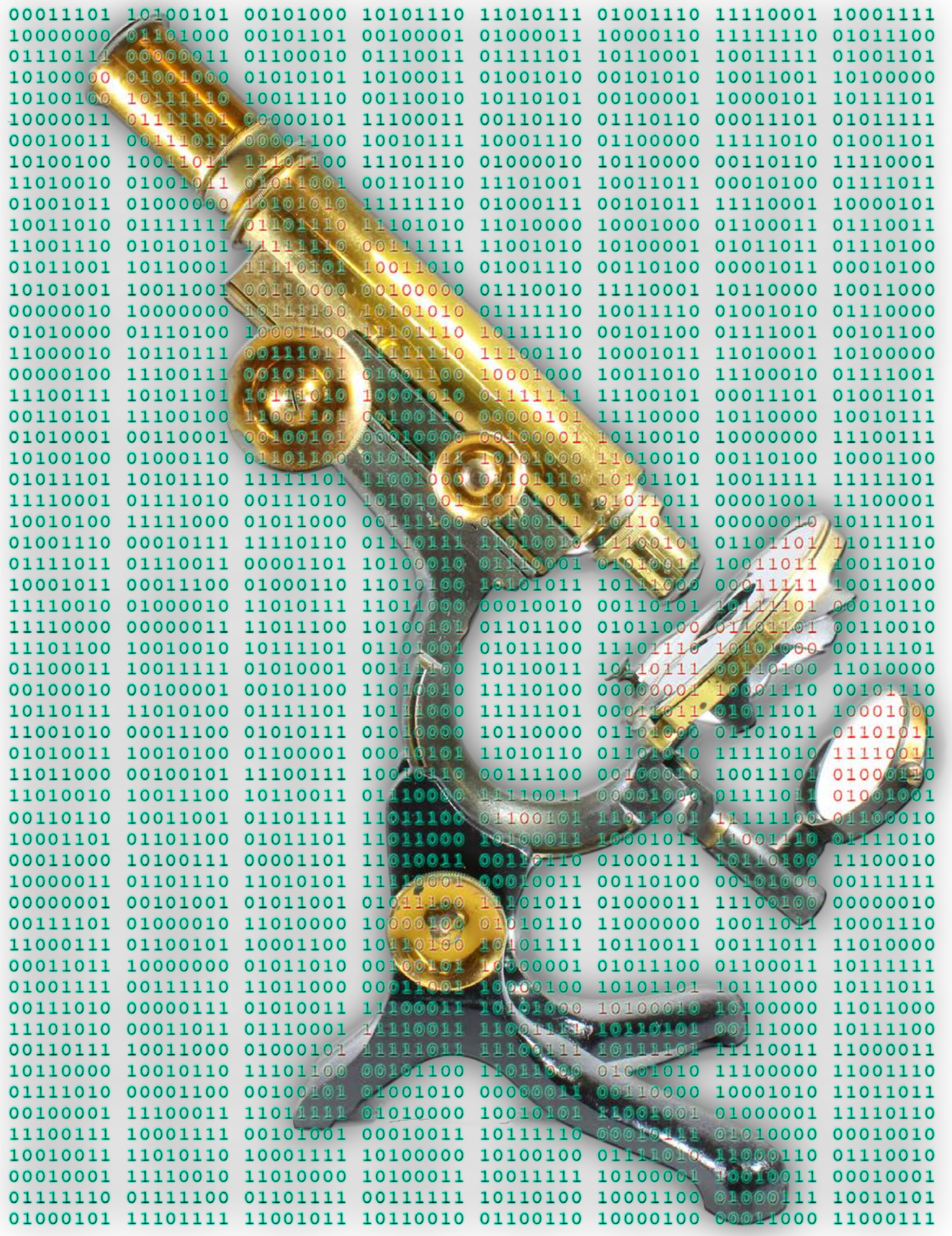
Sourjik, V., Armitage, J.P., *Spatial organziation in bacterial chemotaxis.* EMBO J. (2010), 29(16): 2724–2733.

Briegel, A. et. al. *Universal architecture of bacterial chemoreceptor arrays.* Proc. Natl. Acad. Sci. USA (2009), 106:17181-17186

Briegel, A. et. al. *Bacterial chemoreceptor arrays are hexagonally packed trimers of receptor dimers networked by rings of kinase and coupling proteins.* Proc. Natl. Acad. Sci. USA (2012). 109:3766–3771

Why Computation?

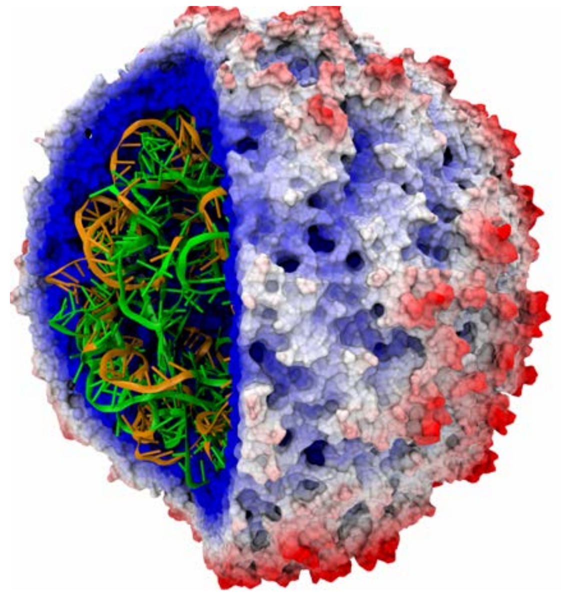
Molecular dynamics (MD) simulations provide an excellent tool to investigate the *structural and dynamical properties* of biomolecules.

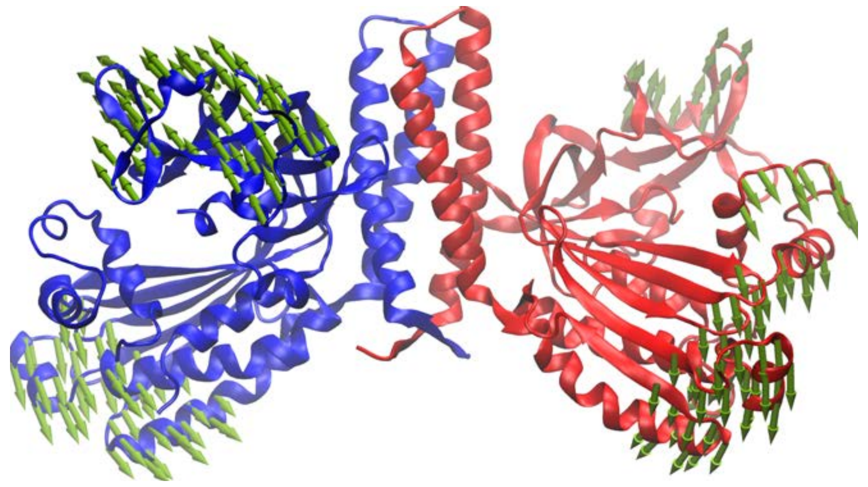


The "Computational Microscope"

Analysis of MD trajectories allow the extraction of *important physical information* from the complex dynamics of biomolecules.

Electrostatic potential of STMV capsid.

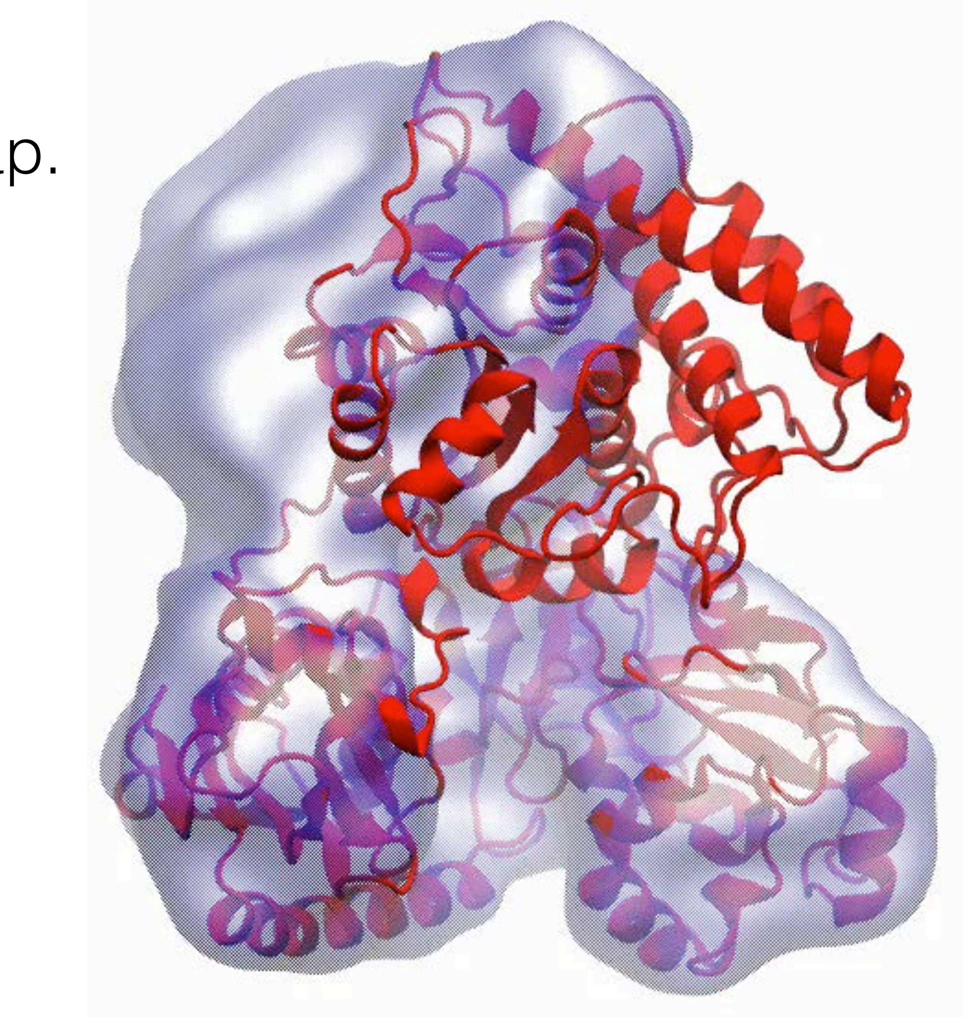




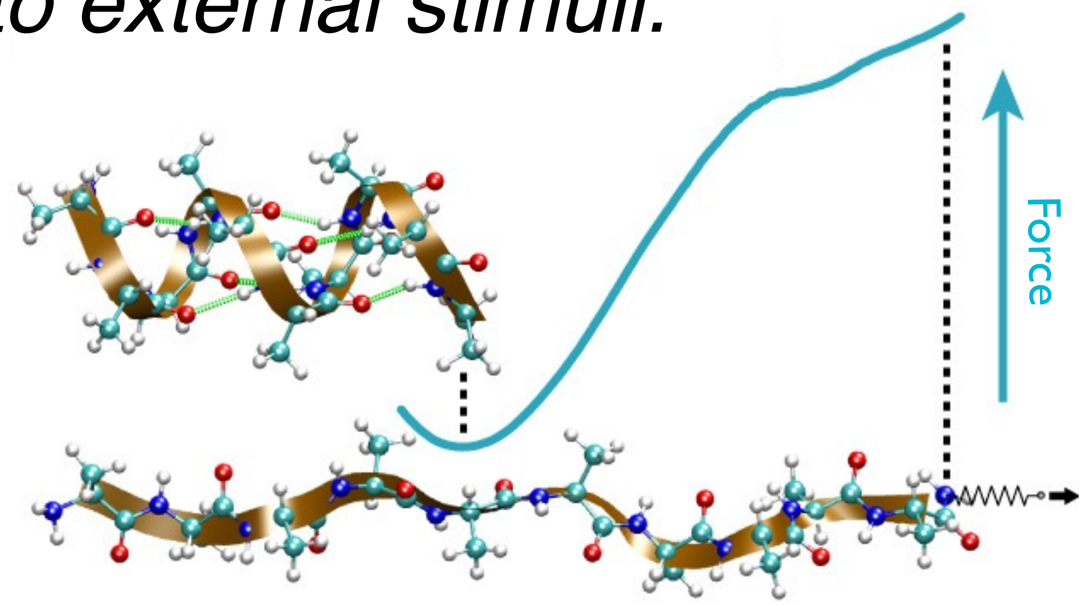
Principal Component Analysis (PCA) of CheA protein.

Computational modeling can ***integrate multi-scale experimental data*** into single model.

MDFF fitting of X-Ray structure into Cryo-EM map.



Steered MD provides the ability to systematically exert forces on biomolecules to investigate ***responses to external stimuli.***



"Steered" stretching of deca-alanine.

Until recently it has been impractical to investigate such a large, multi-protein complex using available computational techniques and facilities….

Why Blue Waters?

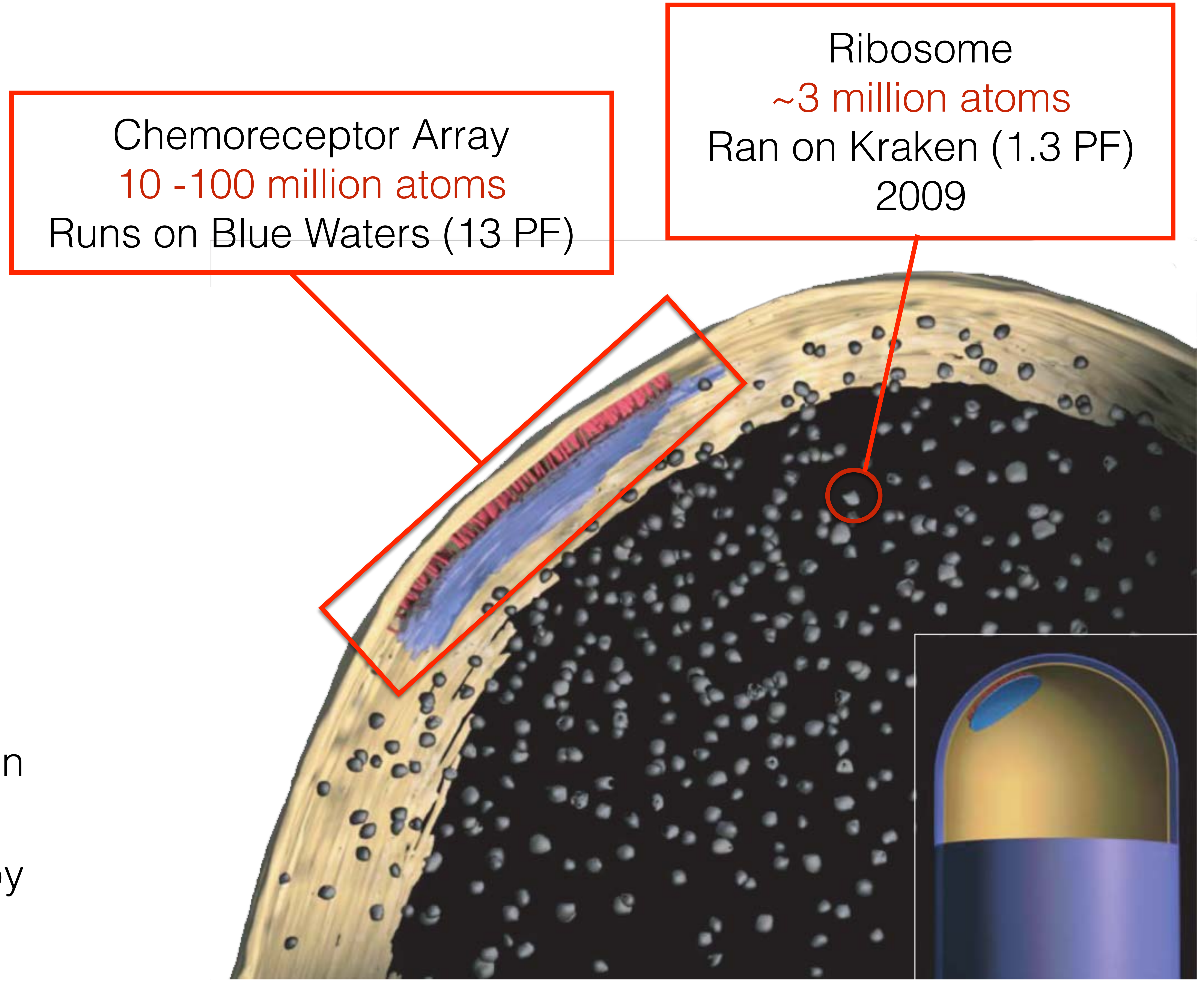
The Chemoreceptor array is a ***petascale*** system…

The array is ***necessarily*** large…

- The array's computational ability emerges from the collective interactions of its many parts.
- Faithfully representing the irreducible nature of the native array structure is essential.

…And needs ***all-atom*** detail.

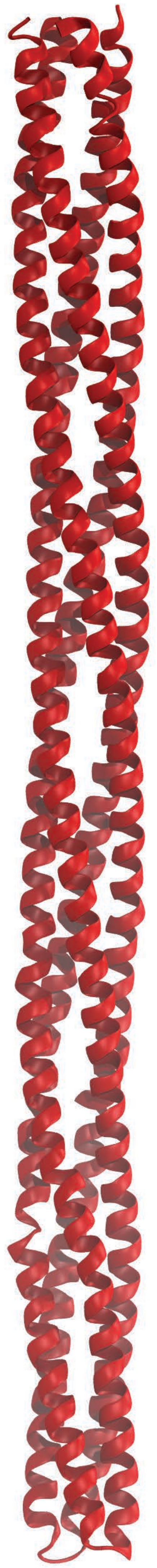
- Experiments point to minute, allosteric rearrangements (i.e., not large-scale changes in structure).
- Mechanisms are subtle and could be missed by CG model.

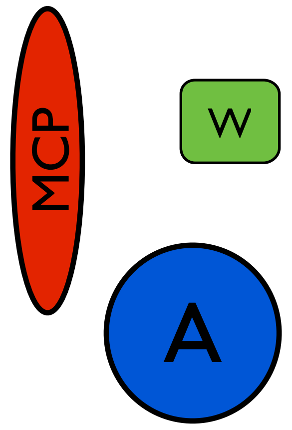


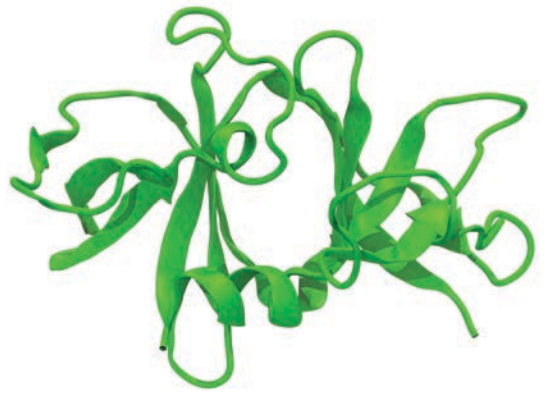
Zhang, P. et. al. *Direct visualization of Escherichia coli chemotaxis receptor arrays using cryo-electron microscopy*. Proc. Natl. Acad. of Sci. USA (2007). 104(10), 3777–81.

Computational Modeling of Chemoreceptor Array.

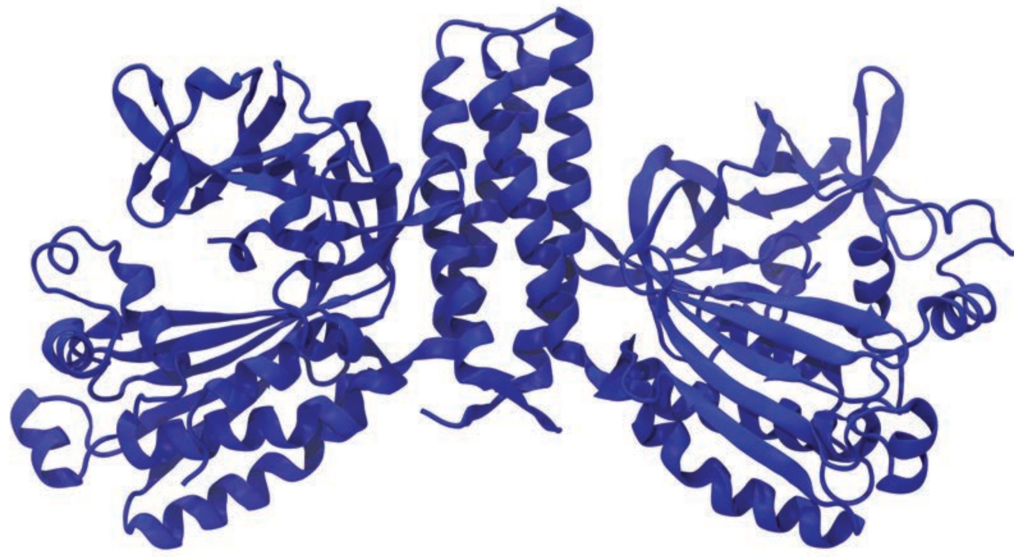
1 Begin with high-resolution crystal structures.
(Non-native environment)







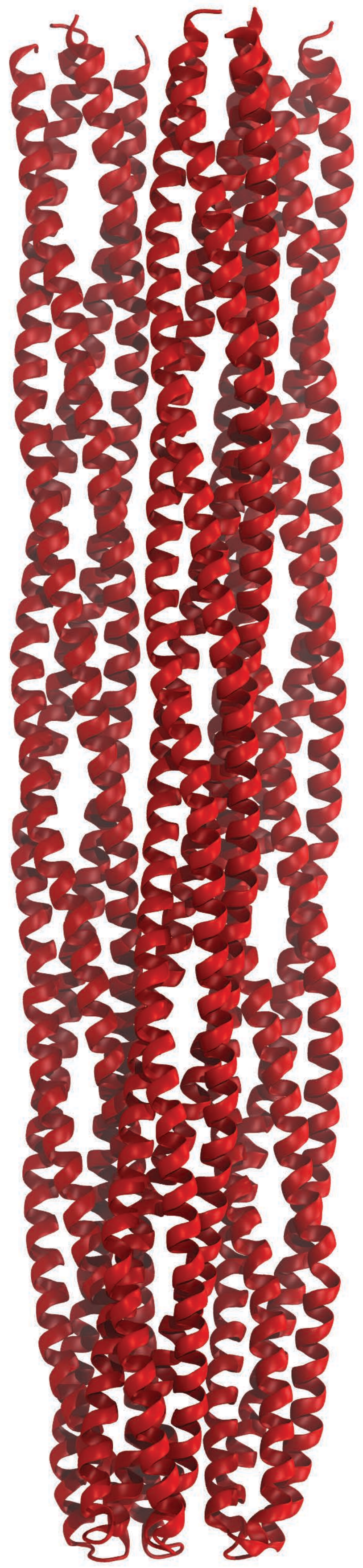
CheW
Adaptor protein
PDB: 3UR1



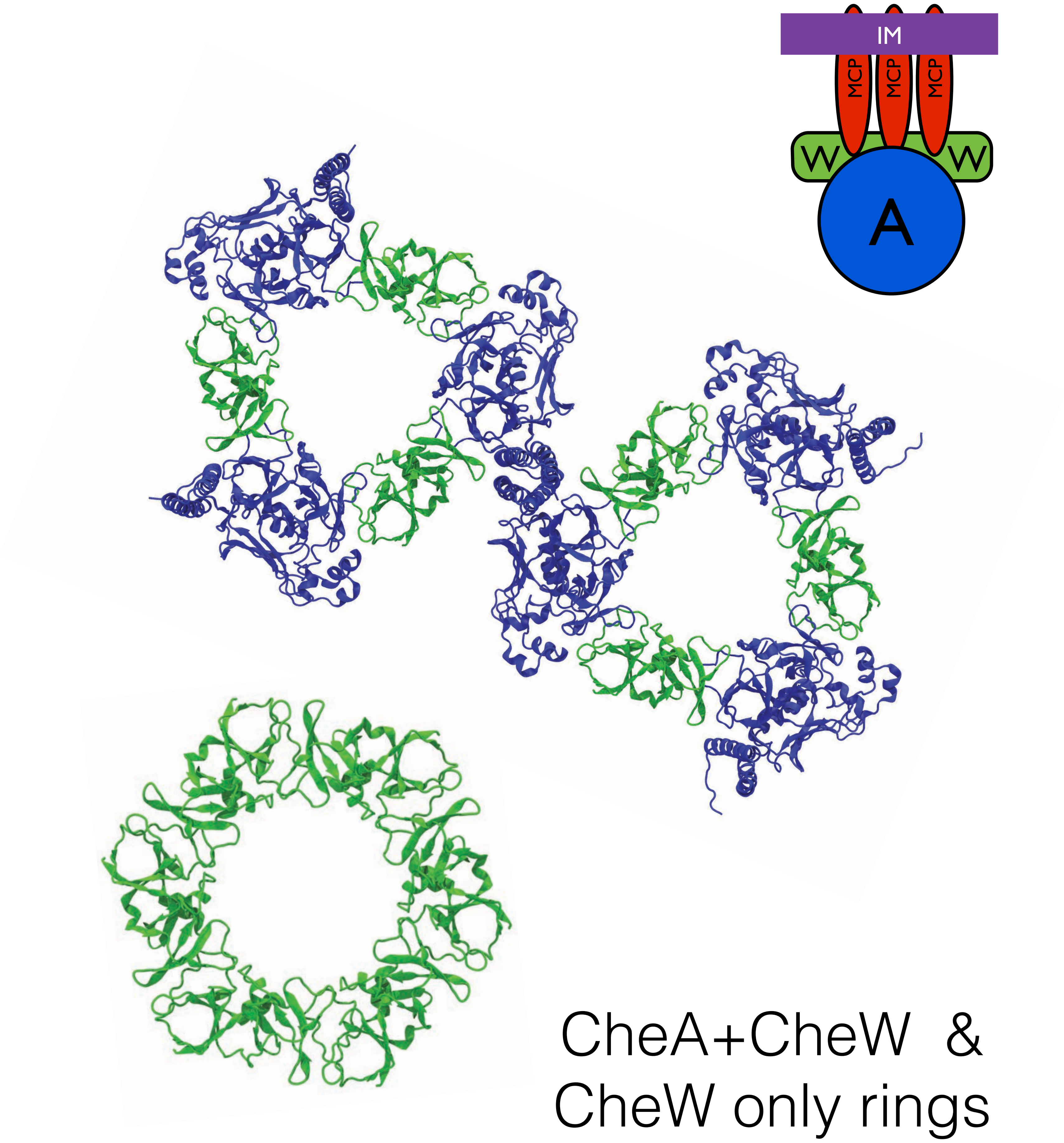
CheA
Histidine Kinase
PDB: 1B3Q

Chemoreceptor dimer
MCP
PDB: 2CH7

2 Computational modeling of array oligomers.



Trimers of chemoreceptor dimers (TODs)
PDB: 1QU7



CheA+CheW &
CheW only rings
PDB: 3UR1

Trimer of chemoreceptor dimers (TOD)

Minimal signaling unit

2 x TOD

1 x CheA dimer

2 x CheW

Array hexagon

(~5 million atoms)

12 x TOD

9 x CheA dimer

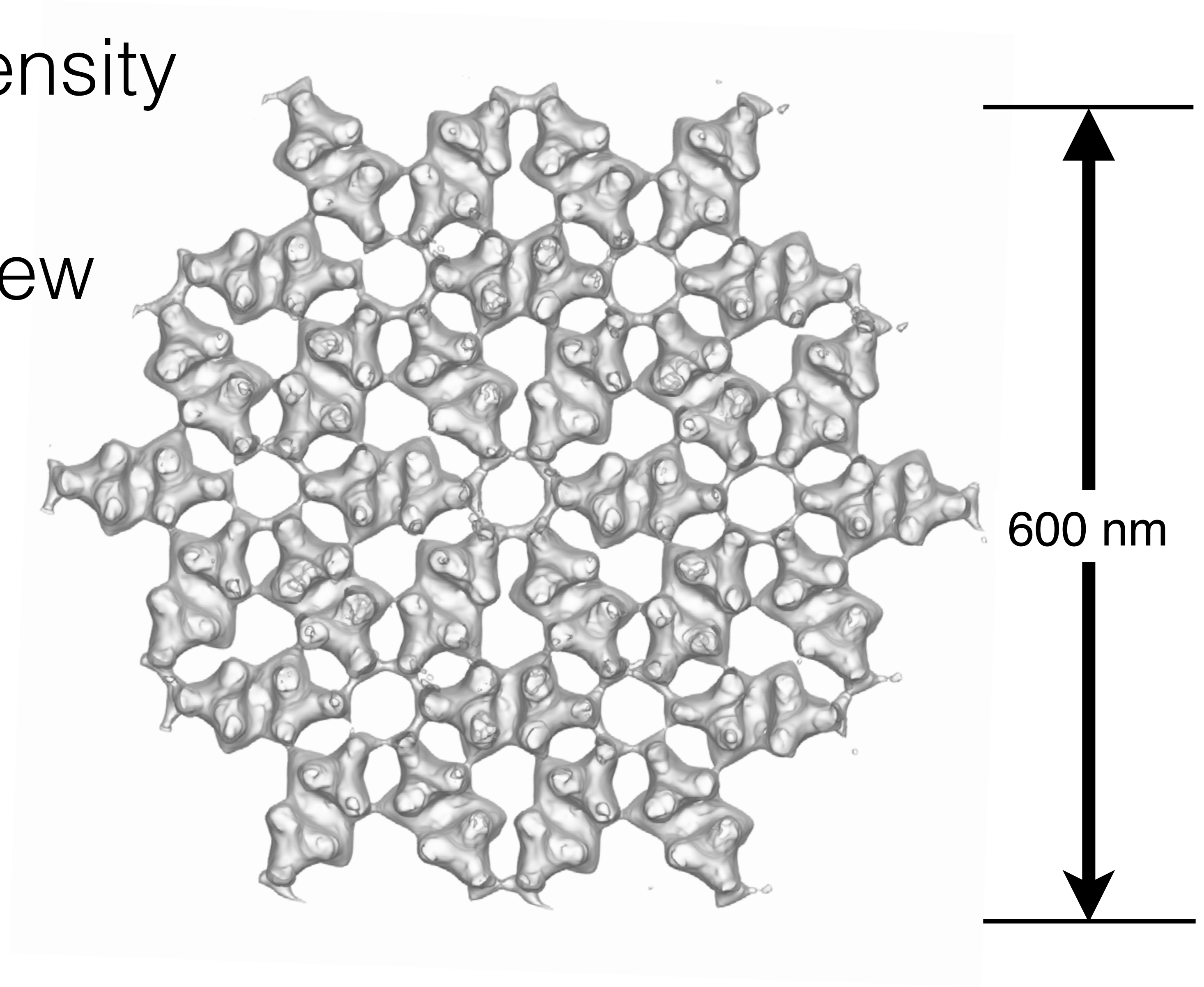
24 x CheW

Computational Modeling of Chemoreceptor Array.

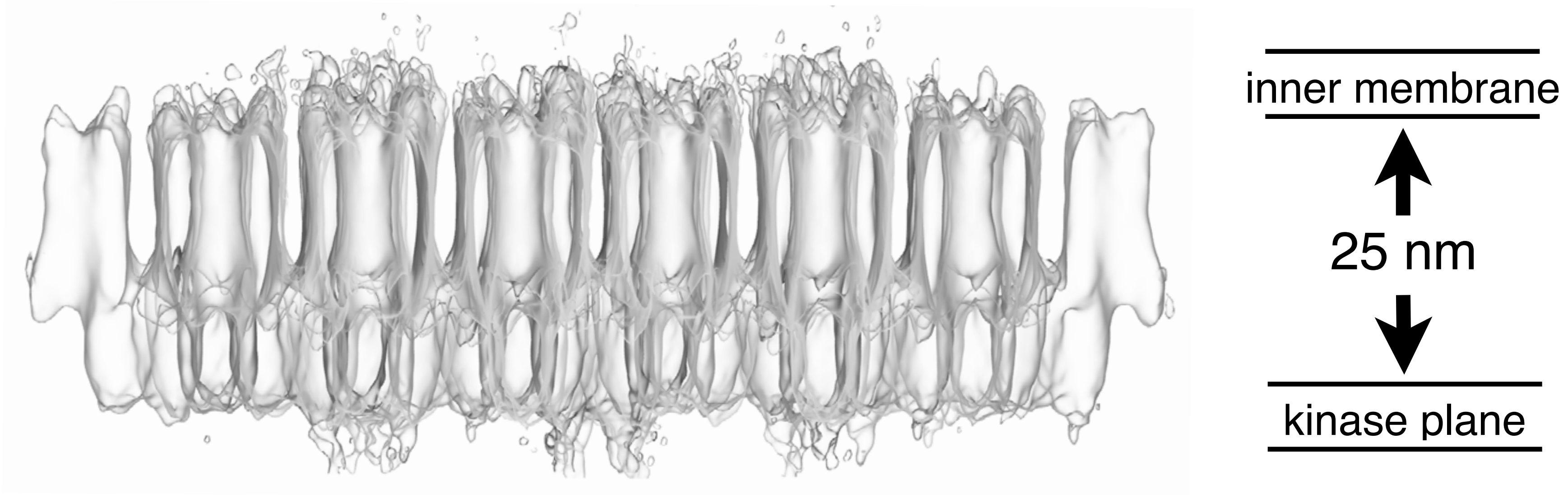
3 Electron Microscopy-guided modeling and refinement.
(Native environment)

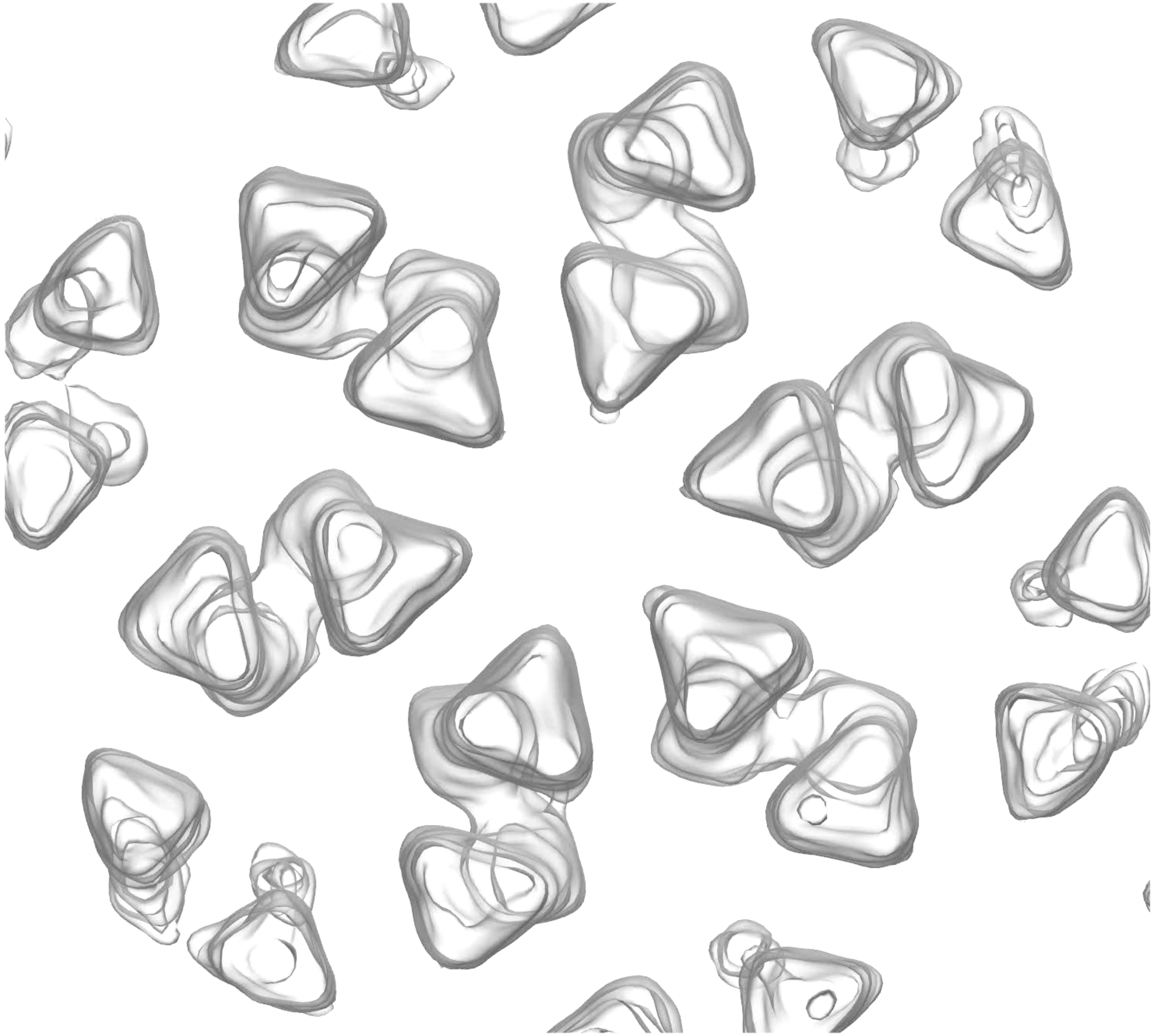
EM Density

Top View



Side View



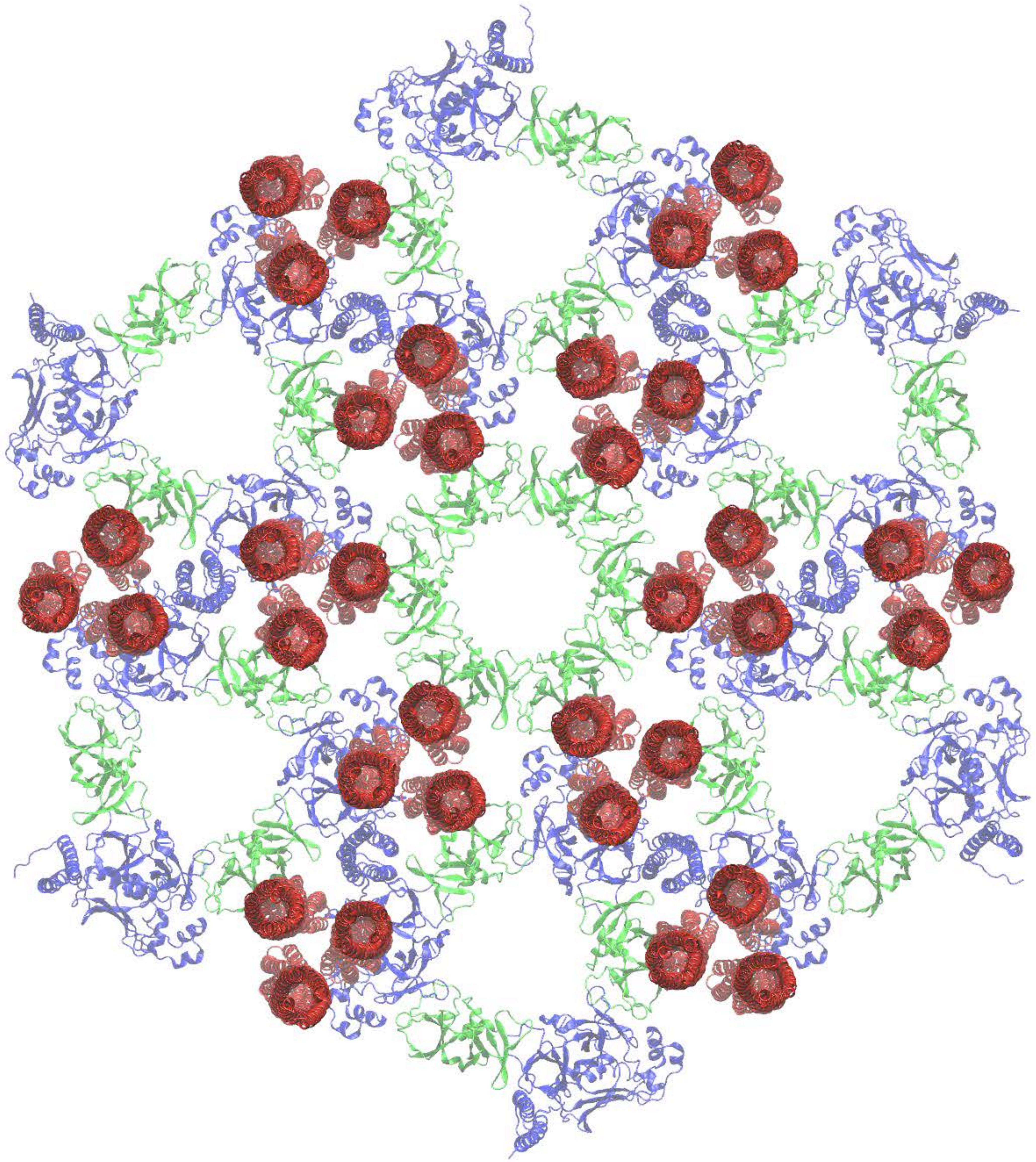


Lattice Model,
~50 million atoms

An experimentally-guided, all-atom model of chemoreceptor array is now available.

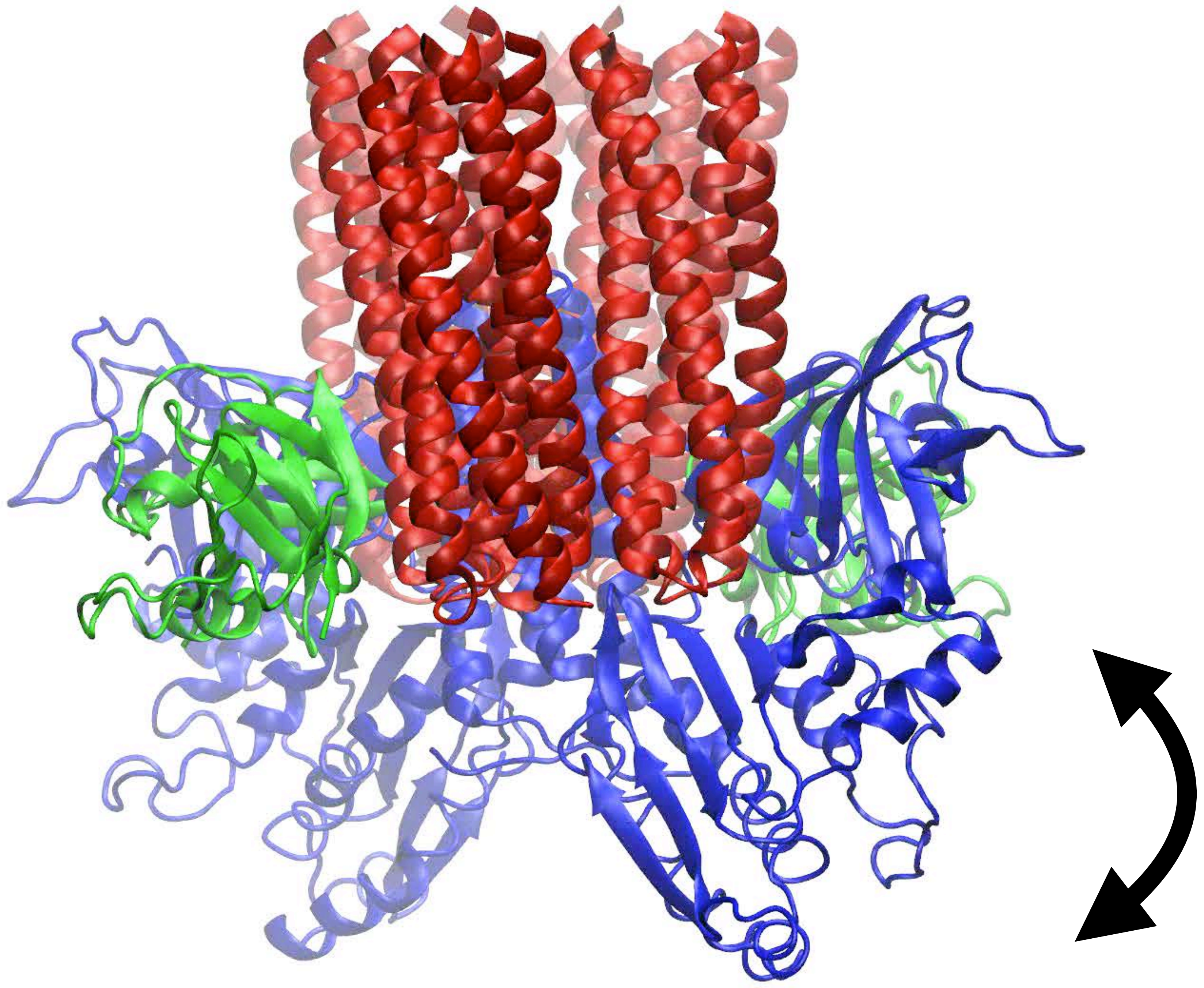
With this model, Blue Waters is allowing us to explore the chemoreceptor array as never before.

Simulations have already offered great insight!



Simulation of Chemoreceptor Array.

1. How is receptor activity physically transducer to CheA?

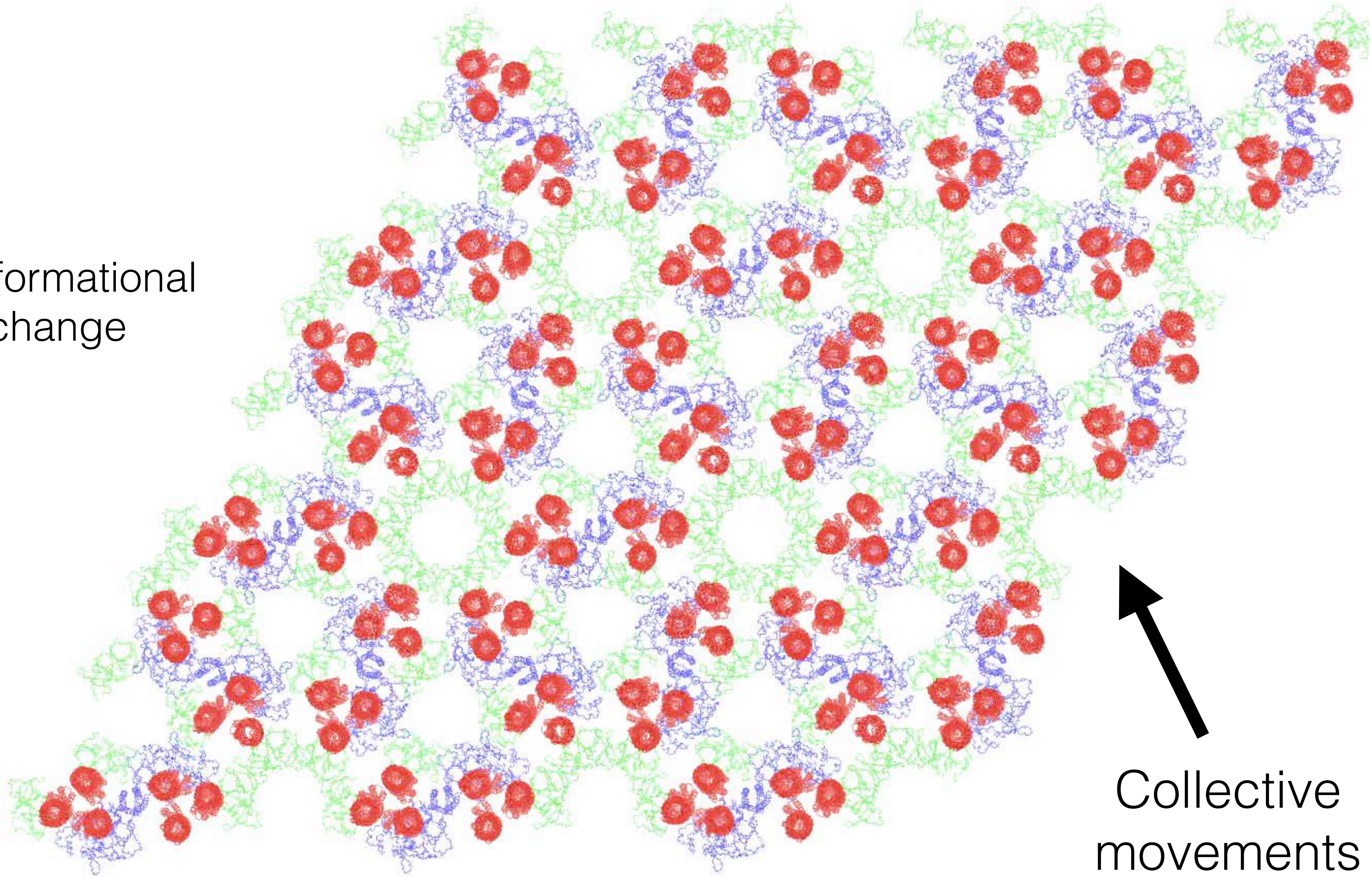


Conformational change

"Dip" in CheA domain possibly modulates kinase activity and is regulated by interactions with chemoreceptors.

2. How are the activities of different receptors coupled?

Modes extracted from simulation trajectories using PCA establish routes of communication between proteins.



Collective movements

Scientific Significance.

Centerpiece of the most thoroughly studied sensory signal transduction system in all of biology.

Represents the next frontier towards a complete understanding of a basic biological computer and sheds light on the fundamental mechanisms by which biological systems process information in general.

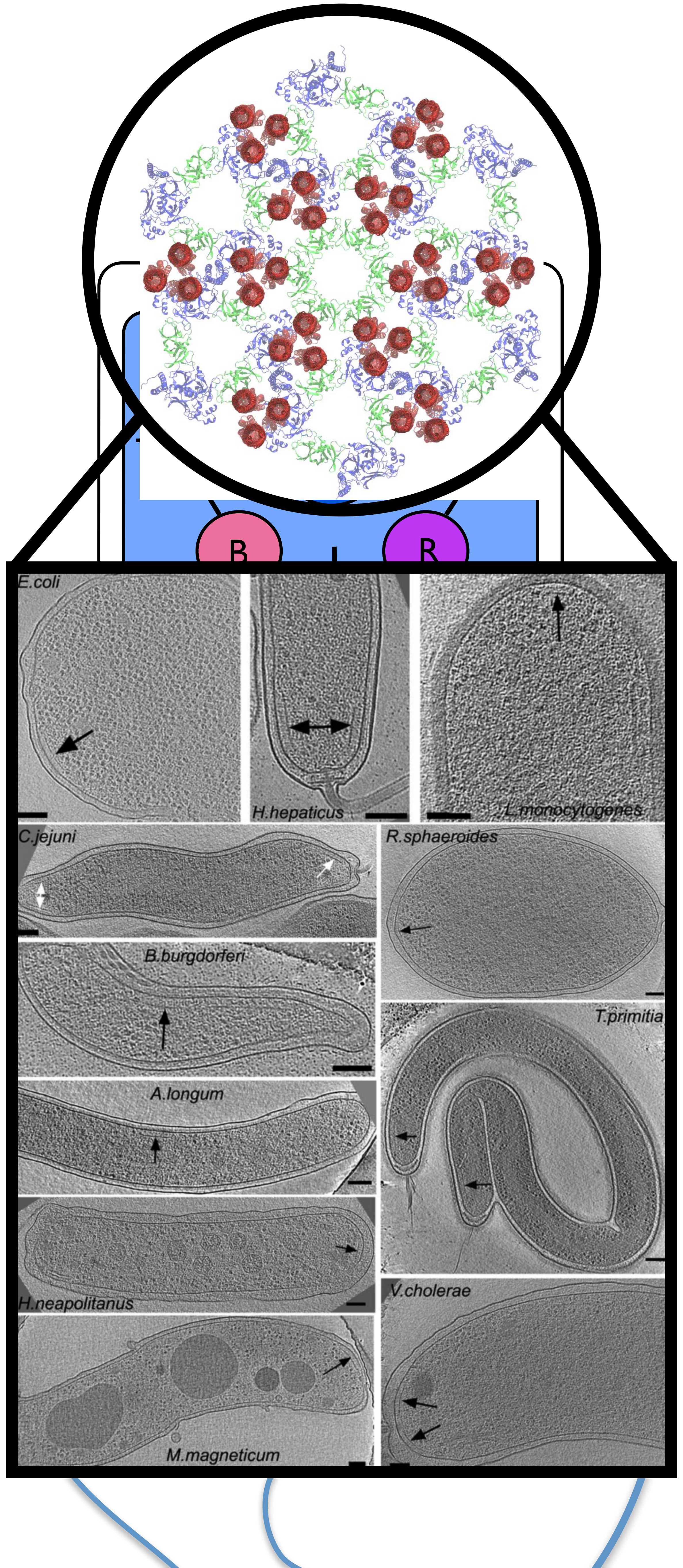
Provides transferability of functional mechanisms between the arrays of distantly related species.

Structural organization and overall mechanism is a universally conserved feature among all bacterial chemotactic systems.

Proteins key to *array's* function do not exist in mammalian organisms.

Combined with the *universality*, understanding the elemental underpinnings of array function could provide *mass reprogramming* and the development of novel antibiotics and fungicides.

Main point: Blue Waters is providing the unique, atomistic perspective needed to tackle a fundamental and previously inaccessible problem in biological information processing.



Briegel, A. et. al. *Universal architecture of bacterial chemoreceptor arrays.* Proc. Natl. Acad. Sci. USA (2009), 106:17181-17186

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Beckman Institute
UIUC

Blue Waters sustained-petascale computing project

National Center for Supercomputing Applications

Center for the Physics of Living Cells
NSF Physics Frontier Center, UIUC

UIUC

Thanks for listening!

Questions?

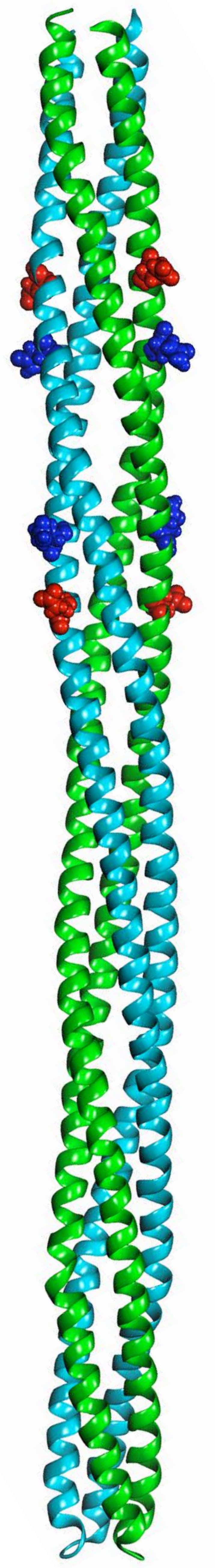
- How is receptor activity physically coupled to kinase activity?
- How are the activities of different receptors physically coupled?

Mathematical modeling and stochastic simulations suggest this characteristic adaptation response originates from clusters of interacting receptors.

These studies have revealed a characteristic "all or none" single-cell swimming behavior.

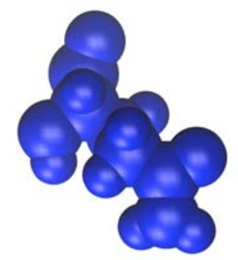
- How does overall architecture of the array lead to distinctive cell-scale regulation of CheA activity observed in Chemla experiments?

Experiements and quantitative modeling studies point to receptor clustering within the array to explain enhanced signaling features.



4-5 methylation sites per MCP
Wild-type (QEQE)

Glutamic Acid (E)



Glutamine (Q)

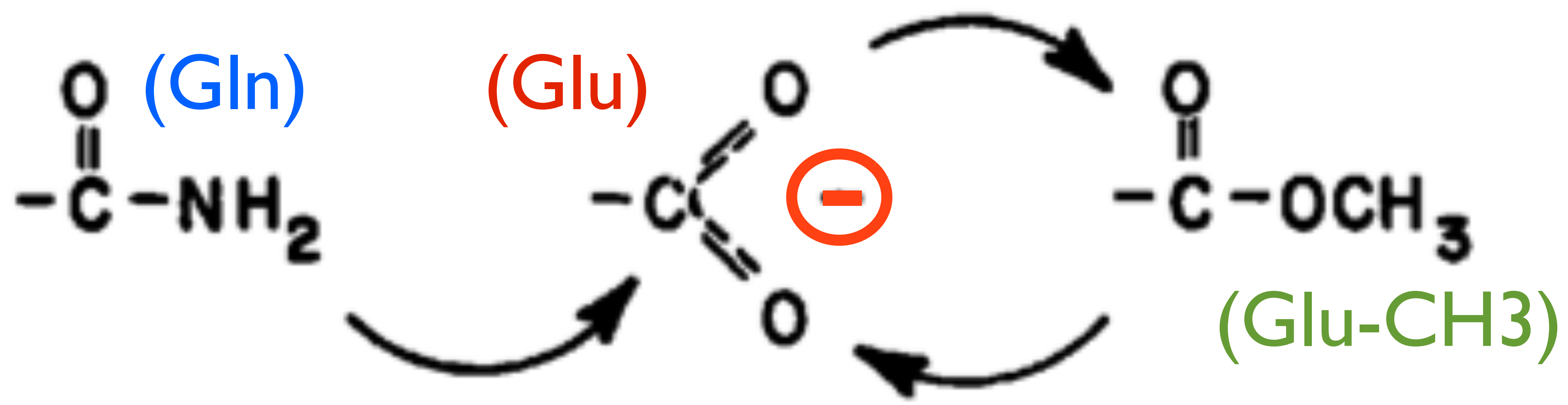
Gln (Q) mimics methylated Glu (E), encoding median level of activity

The cell remembers!

Bacterial cells temporally compare chemoeffector concentrations through receptor methylation levels

Adaptation methylation

CheR **methylates** Glu

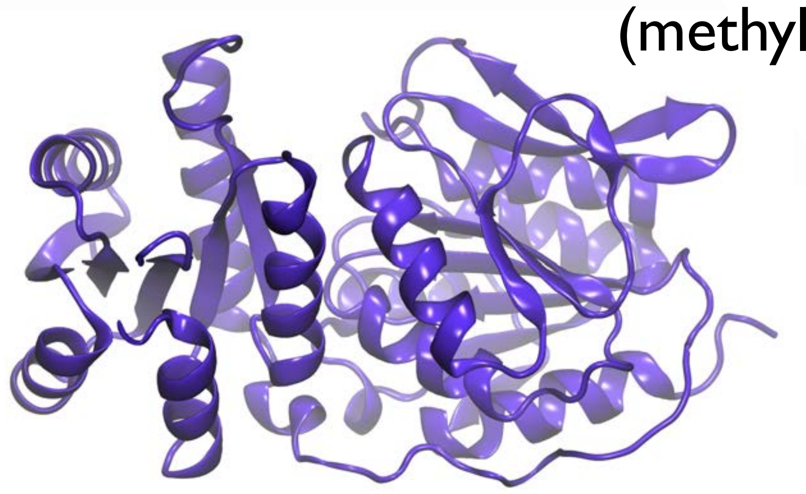


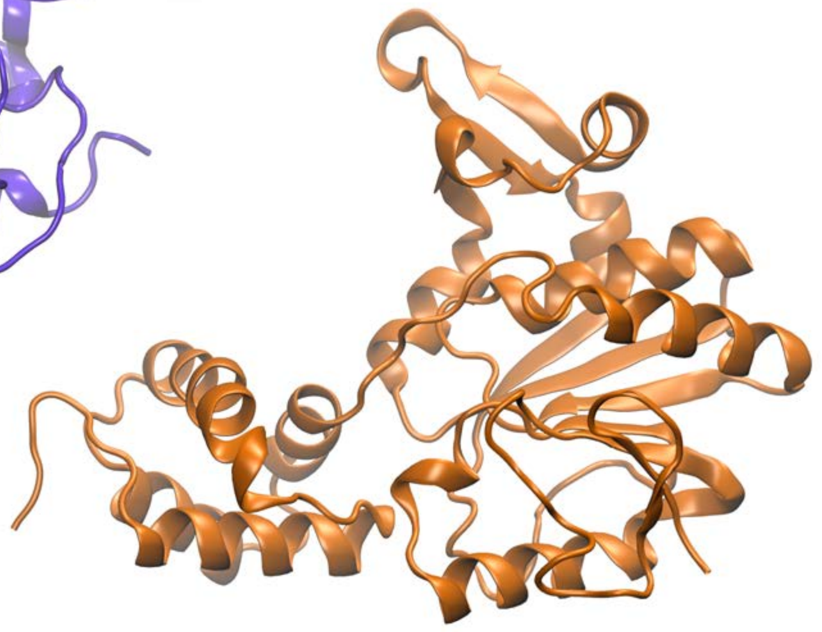
Gln is deamidated by CheB to participate in adaptation process

CheB **demethylates** Glu-CH3

CheR

(methyltransferase)



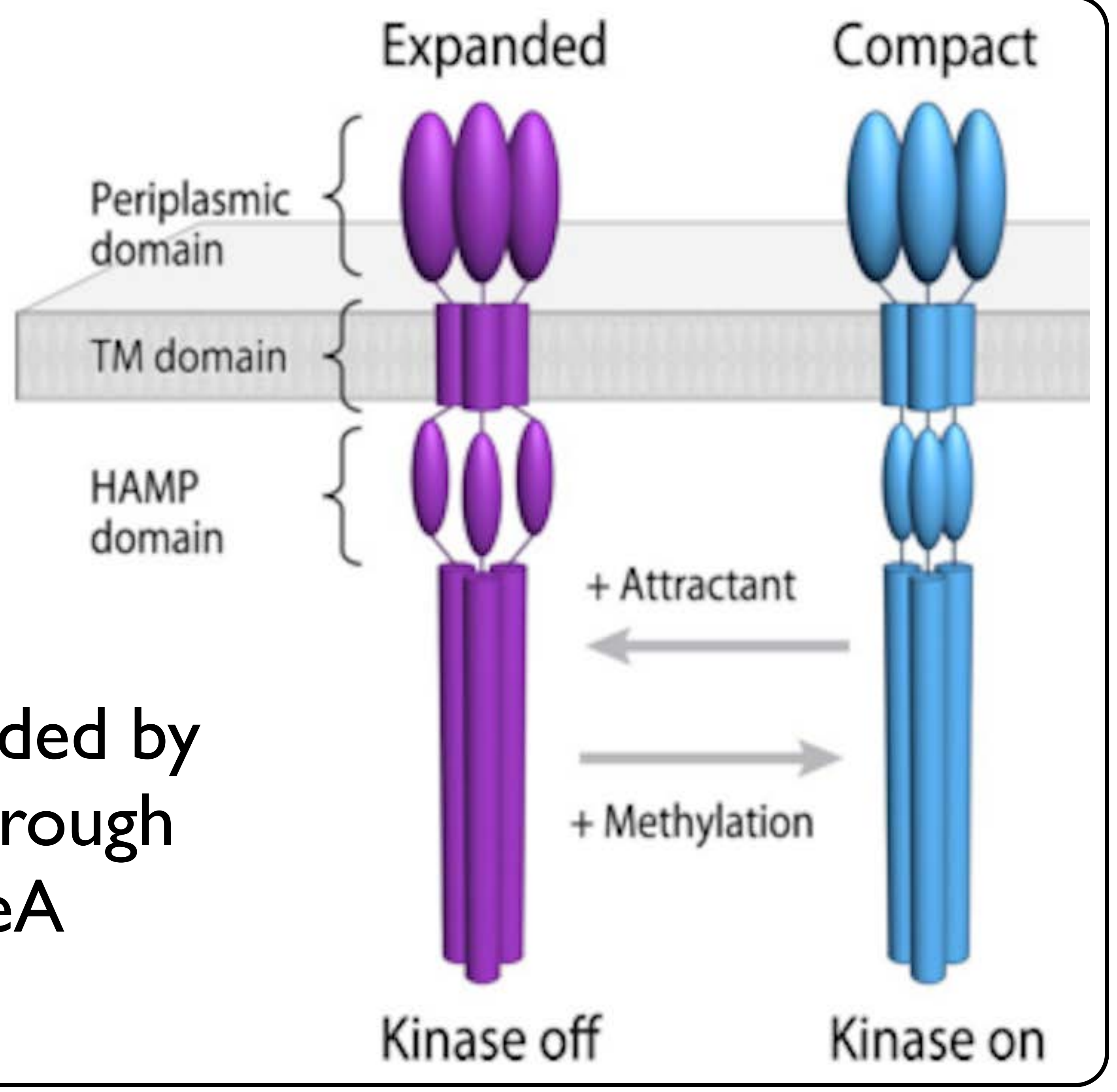


CheB

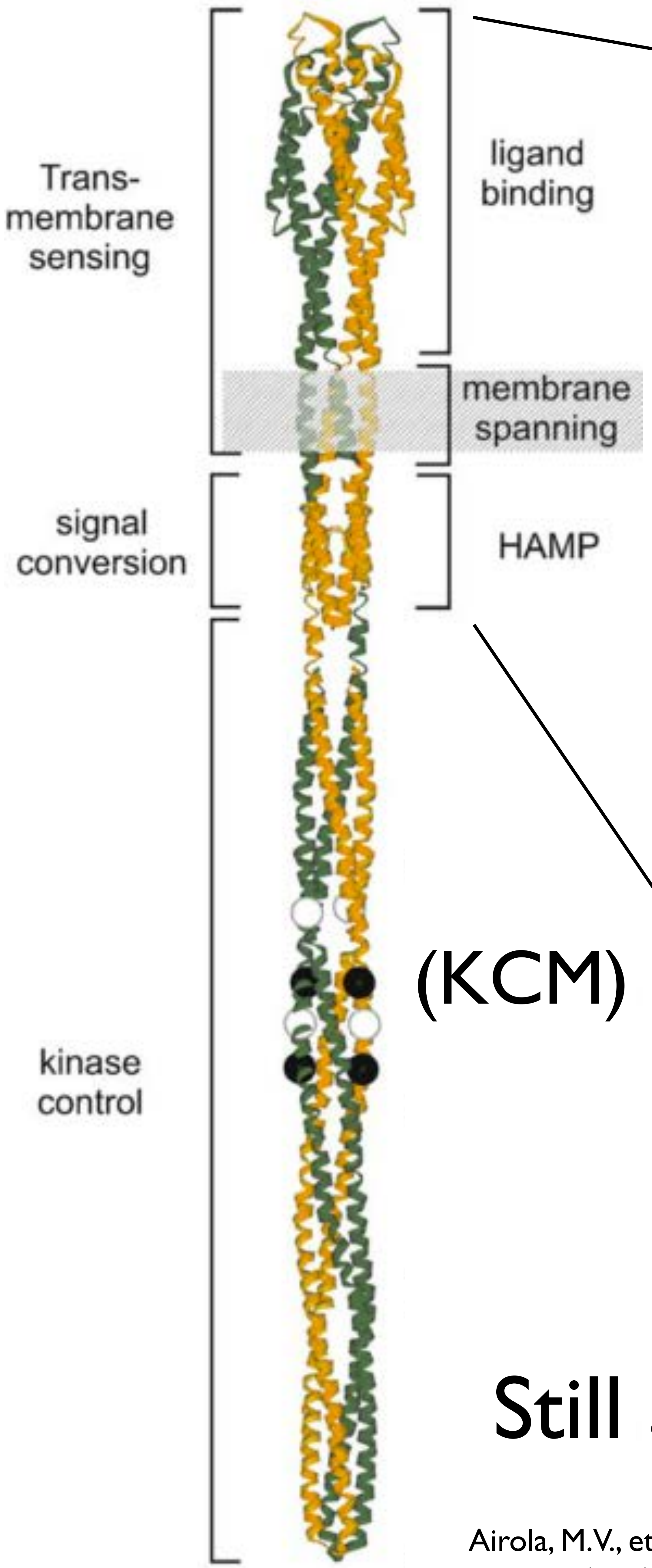
(methylesterase)

Receptor methylation alters distribution of receptor trimer-of-dimers conformations and CheA activity

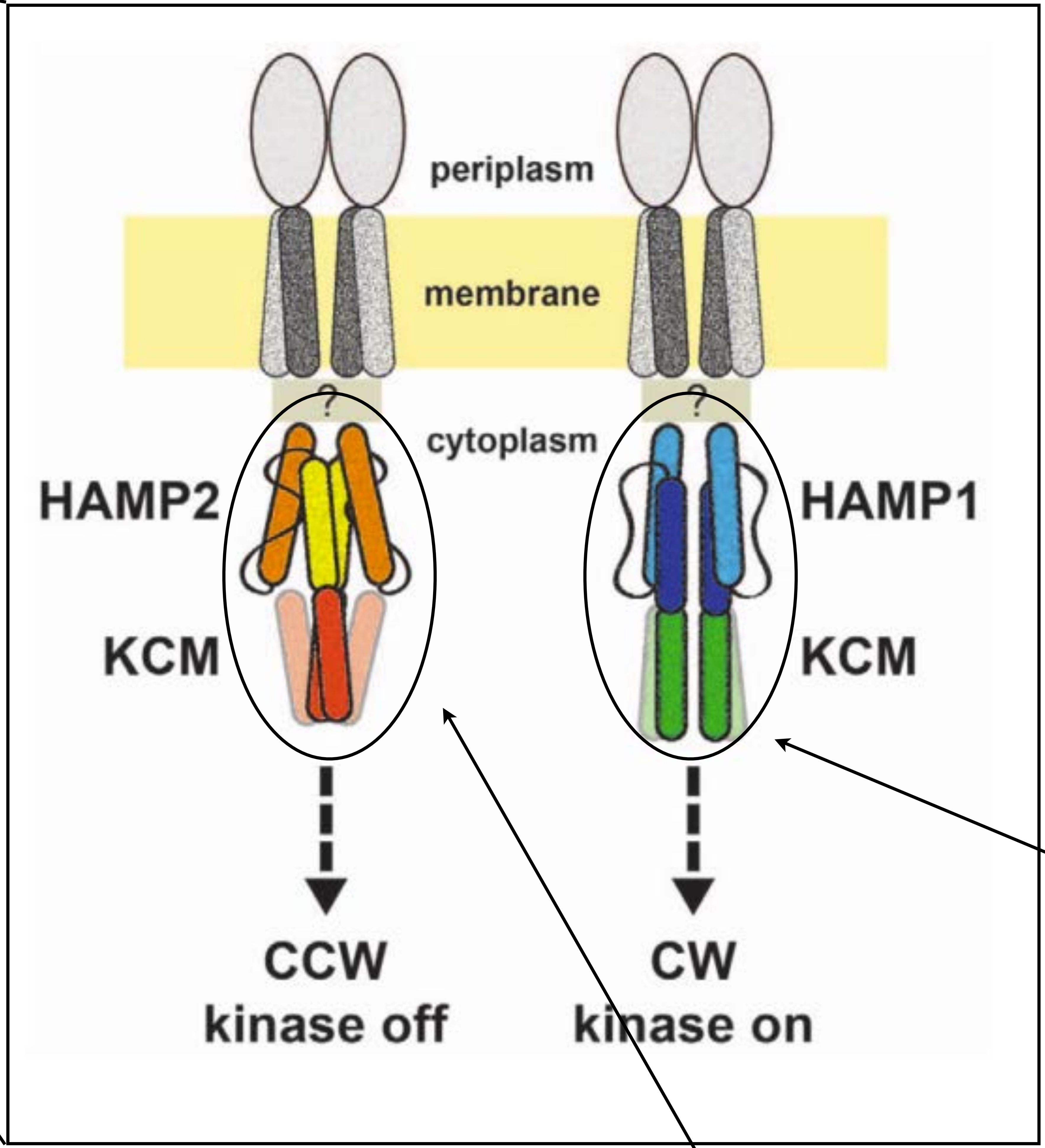
Feedback mechanism provided by increased CheB activity through phosphoylation by CheA



HAMP signaling in chemoreceptors



(KCM)



Current Opinion

1. Stimulus-induced conformational changes are conveyed to HAMP domains through piston or rotary motions from adjoining transmembrane helices

2. Input stimuli modulate HAMP signaling by shifting the relative stabilities of two alternative HAMP structural states to regulate bacterial chemotaxis.

3. HAMP domains probably control the output activity by modulating the packing stabilities of the output helices through oppositional structural coupling.

A dynamic HAMP1 forms a continuous four-helix coiled coil across the junction to generate kinase activation and CW flagella rotation.

HAMP2 imparts a two-helix coiled coil across KCM junction, which results in CheA kinase inhibition and CCW flagella rotation.

Still a lot of work to be done!

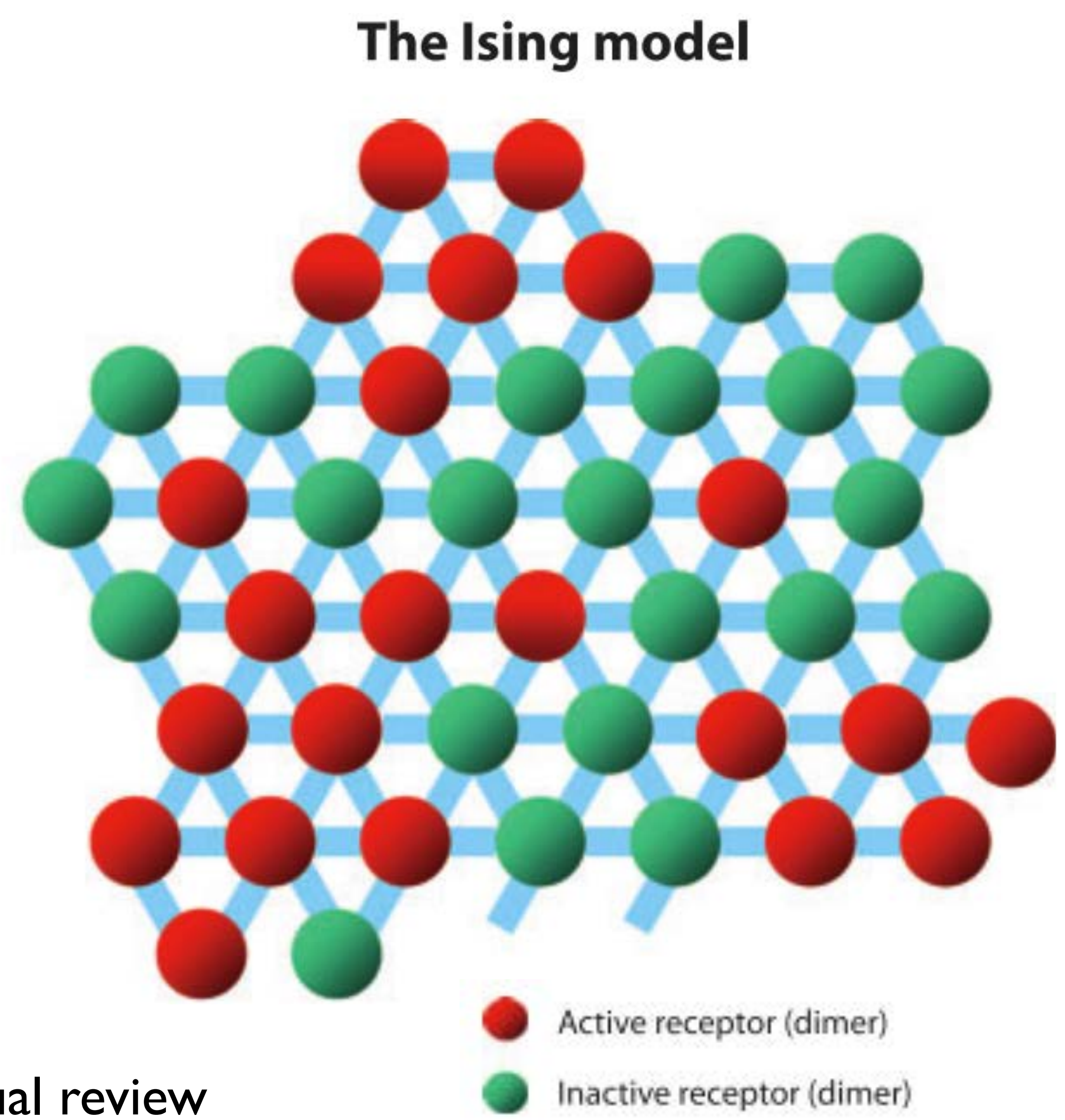
Airola, M.V., et al. 2013. HAMP domain conformers that propagate opposite signals in bacterial chemoreceptors. *PLoS Biol* 11(2)

Theoretical Insights

Gain & Sensitivity

The Ising model is often used to model two-state nature of chemoreceptor signaling

$$\langle a \rangle = \frac{e^{-f_m(m)}(1 + [L]/K_a)}{1 + [L]/K_i + e^{-f_m(m)}(1 + [L]/K_a)}$$



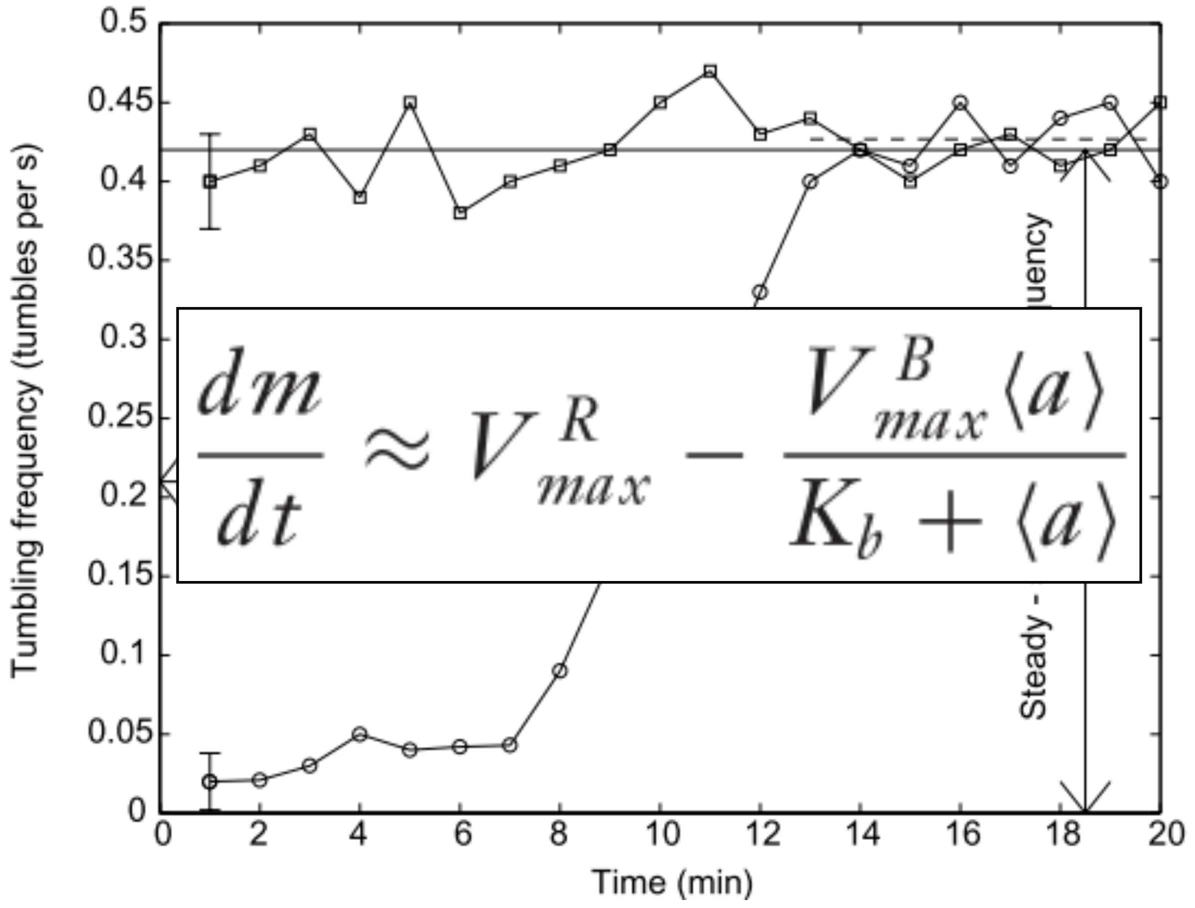
Tu, Y., 2013. Annual review of biophysics. 42, 337-359.

Such coarse-grained approaches can be tuned to match experiment, but do not allow investigation of the underlying molecular details giving rise to such systems-level features

Adaptation

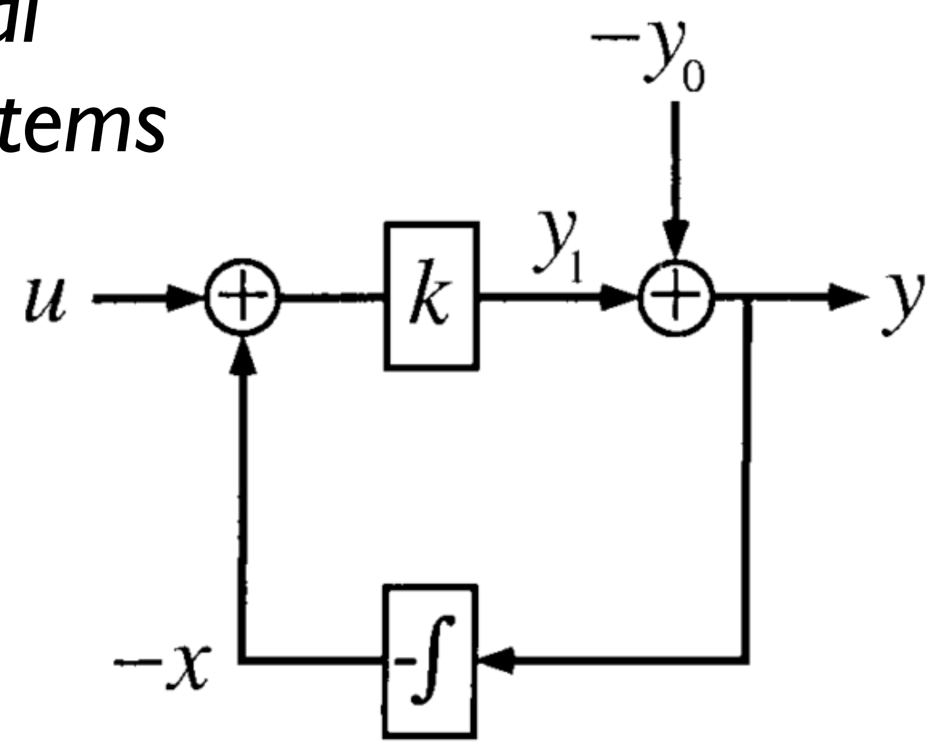
The bacterial chemotaxis network exhibits precise adaptation that is robust against variation in biochemical parameters

Alon, U., Surette, M. G., Barkai, N. & Leibler, S., 1998. Nature. 397, 168–171.



$$\frac{dm}{dt} \approx V^R_{max} - \frac{V^B_{max}\langle a \rangle}{K_b + \langle a \rangle}$$

Robustness is an innate feature of integral feedback control systems

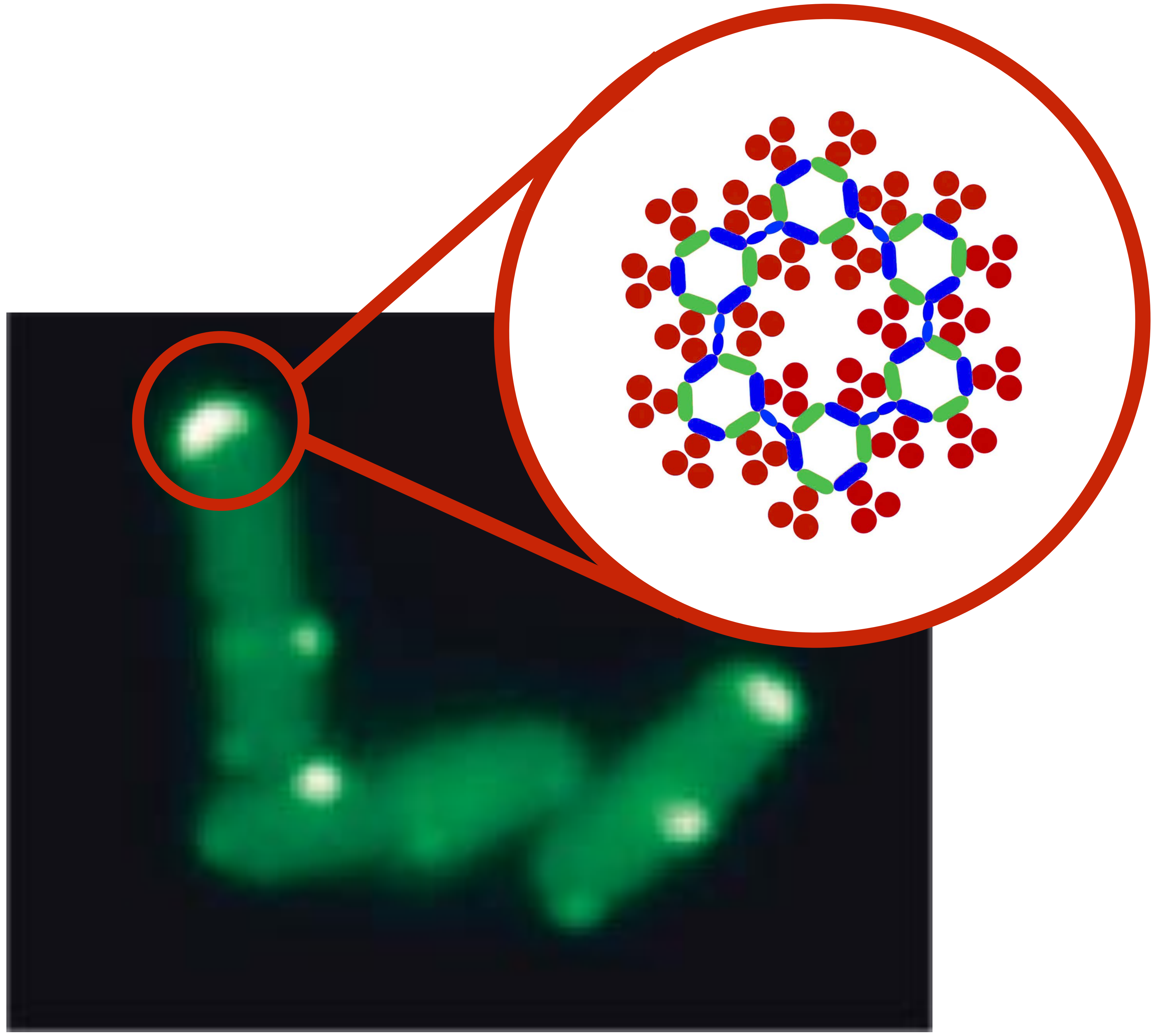


Yi, T., Huang, Y., Simon, M., Doyle, J., 2000. Proc. Natl. Acad. Sci. 97(9), 4649–4653.

Tindall, M. J., Maini, P. K., Porter, S. L., & Armitage, J. P., 2008. Bulletin of mathematical biology. *70*(6), 1570-1607.

The Bacterial Brain: A naturally-evolved, mechanical computer.

RECEPTOR CLUSTERING within the chemoreceptor array has been proposed to explain enhanced signaling features.



Fluorescence images of receptor clusters in whole *E. coli* cells.

Sourjik, V., Armitage, J.P., Spatial organziation in bacterial chemotaxis. EMBO J. 2010 August 18; 29(16): 2724–2733.