Chemotaxis Signaling Pathway

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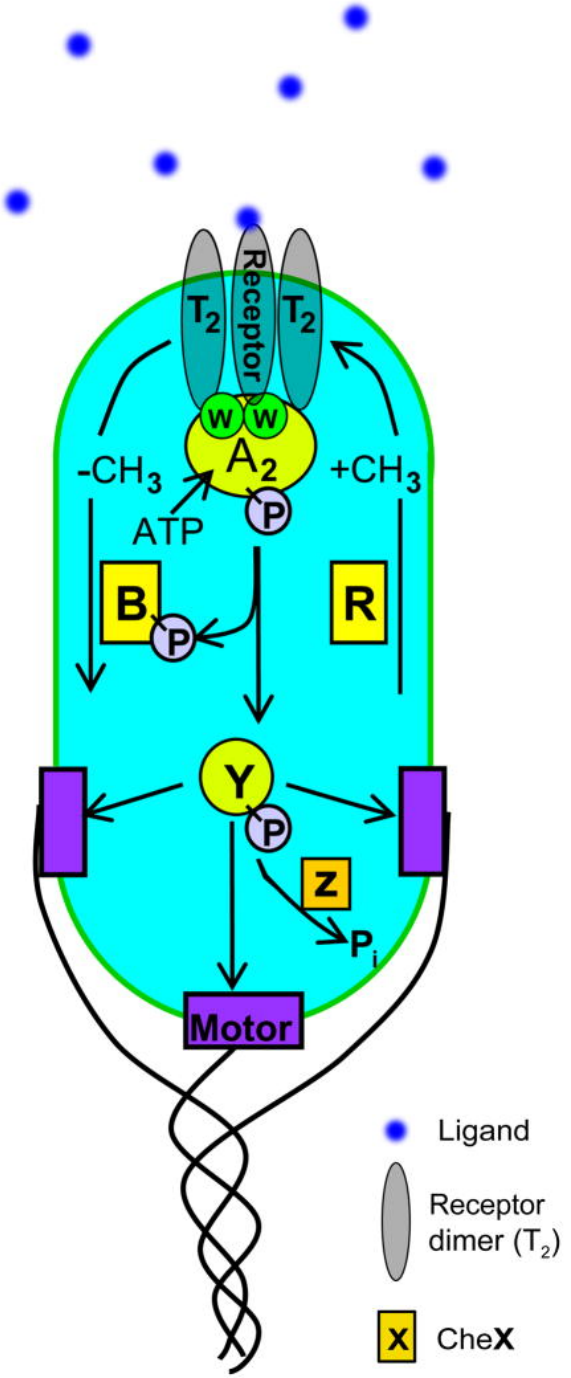
- Best-studied sensory signal transduction systems in biology
- Also serves as a model system for studying two-component signaling pathways, which are ubiquitous in the bacterial and plant kingdoms
- Despite its simplicity, bacterial chemotaxis exhibits rich biological behaviors such as robust adaptation, signal amplification, and ultrasensitivity.
- Major players are transmembrane receptors and six cytoplasmic proteins: CheA, CheB, CheR, CheW, CheY and CheZ
- The receptors form a complex with the histidine kinase CheA through the adaptor protein CheW
- The autophosphorylation activity of CheA is suppressed (enhanced) when chemoattractant (repellent) binds to the receptor



- The activated CheA acquires a phosphate group through autophosphorylation
- Transfers it to the response regulator CheY or the demethylation enzyme CheB
- The phosphorylated CheY can bind with the flagellar motor and can increase the motor's clockwise (CW) bias and the cell's tumble probability
- Adaptation is facilitated by the methylation and demethylation of the chemoreceptors, catalyzed by CheR and CheB-P, respectively.

Receptor clustering and Bray hypothesis

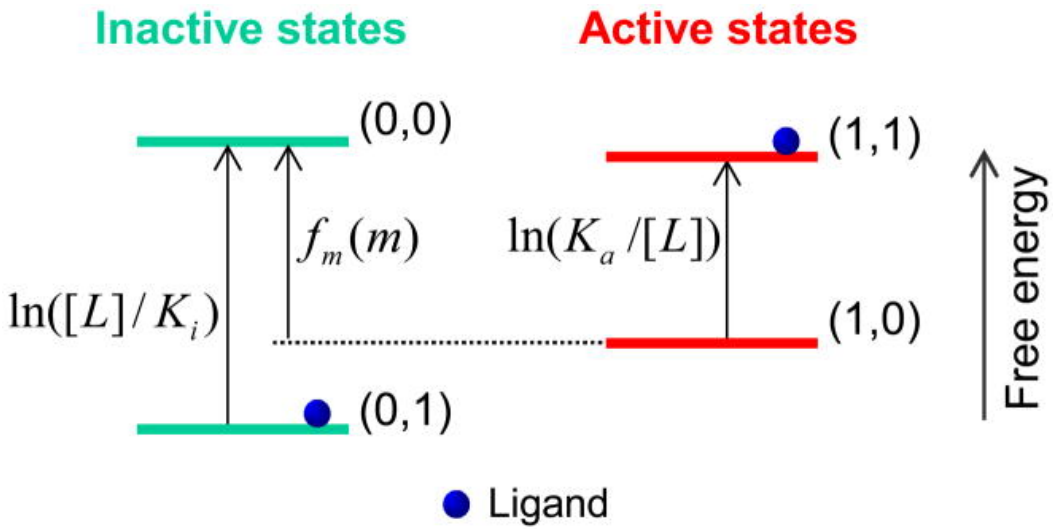
- A few tens of thousands of chemoreceptors in a single E. coli cell
- Chemoreceptors are seen to form large clusters near the cell pole [Maddock and Shapiro, *Science* 1993]
- What may be the function of such a large cluster of receptors?
- Cooperativity due to receptor clustering can lead to signal amplification in bacterial chemotaxis [Bray et al. *Nature* 1998]
- Conformational change of a chemoreceptor can be modulated by conformational changes of the neighboring receptors in the cluster.
- This 'infection model' can give rise to increased sensitivity as the binding of a ligand molecule to one receptor in the cluster can induce responses in many other receptors.

Two-state Ising model for chemoreceptor cluster

- Chemoreceptors form homo-dimer, each homo-dimer can bind with one ligand molecule
- The simplest model for describing the kinase activity of a chemoreceptor dimer assumes that it has two discrete conformations: one active and the other inactive $\Rightarrow$ Two-state Ising model
- Active receptor $\equiv$ upspin, inactive receptor $\equiv$ downspin
- The cooperative receptor-receptor interactions between nearest neighbors in the receptor cluster can then be modeled as the Ising ferromagnetic spin-spin interaction

$$\mathcal{H} = -\sum_{<ij>} J\, s_i s_j - \sum_i h\, s_i$$

- How to determine the effective 'magnetic' field for the receptor cluster?
- 4-state model of the chemoreceptor by considering the receptor's ligand binding status explicitly.
- State of a receptor can be characterized by two binary variables (a, l)
- $a = 0, 1$ for inactive and active forms of the receptor
- $l = 0, 1$ for vacant and ligand-bound receptors



- Ligand dissociation constants for active ($a = 1$) and inactive ($a = 0$) receptor are K_a and K_i
- In absence of ligand $l = 0$ and free energy difference between active and inactive states $f_m(m)$ only depends on its methylation level m
- In steady state

$$\frac{P(0,1)}{P(0,0)} = \frac{[L]}{K_i}, \qquad \frac{P(1,1)}{P(1,0)} = \frac{[L]}{K_a}, \qquad \frac{P(0,0)}{P(0,1)} = e^{-f_m(m)}$$

- Normalization condition $\sum_{a,l} P(a,l) = 1$

$$\langle a \rangle = P(1,0) + P(1,1) = \frac{e^{f_m(m)}(1 + [L]/K_a)}{1 + [L]/K_i + e^{f_m(m)}(1 + [L]/K_a)}$$

- Free energy difference between the active state $(P(1,0) + P(1,1))$ and inactive state $(P(0,0) + P(0,1))$

$$\Delta f(m, [L]) = f_m(m) + f_L([L]) = f_m(m) + \ln \frac{1 + [L]/K_a}{1 + [L]/K_i}$$

- Average activity $\langle a \rangle = (1 + e^{-\Delta f})^{-1}$
- Δf has one 'internal' contribution that depends on the internal state of the receptor, *viz* its methylation level and another 'external' contribution that depends on the external environment, *i.e.* the ligand concentration
- Δf plays the role of effective 'magnetic field'
- K_a and K_i set the range of sensitivity

- Using an Ising-type model, describe the kinase activity of a chemoreceptor cluster with total number of receptors N_t
- The activity of each receptor $i \in [1, N_t]$ in the cluster $a_i = 0, 1$
- Each neighboring receptor pair in the cluster interact with each other with an interaction strength J which favors the neighboring pair to have the same activity (0 *or* 1)
- The activity of an individual receptor is also affected by an effective magnetic field Δf
- The energy for a given activity pattern $\vec{a} = \{a_1, a_2, ..., a_{N_t}\}$

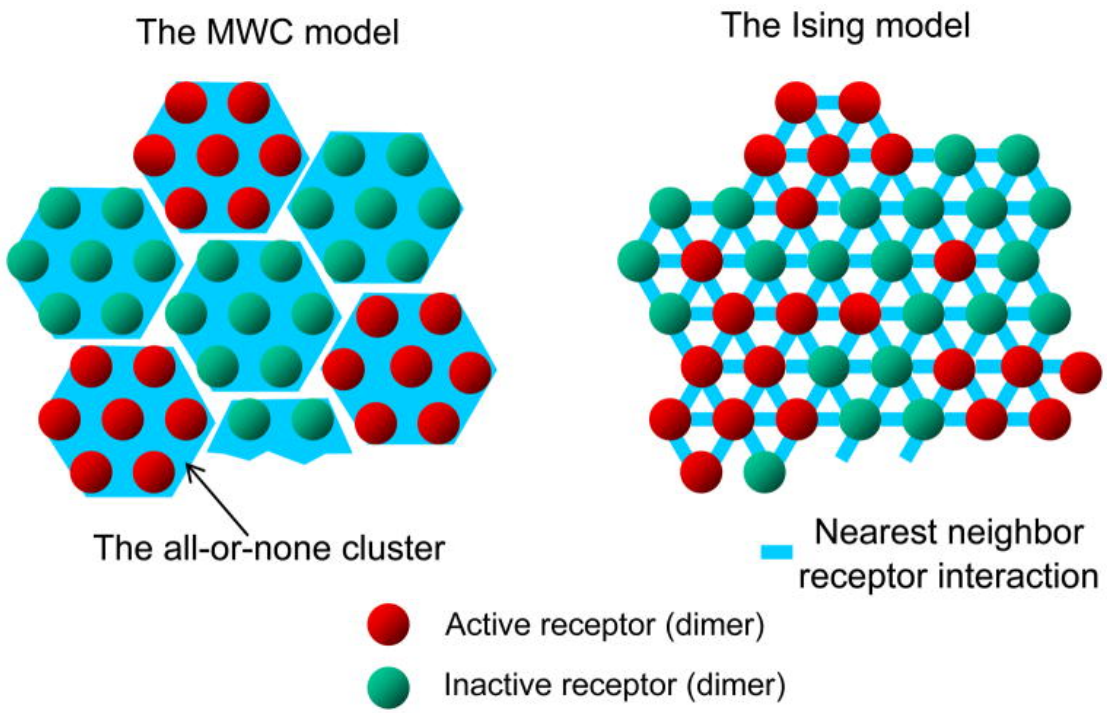
$$\mathcal{H}(\vec{a}) = -J \sum_{<i,j>} (2a_i - 1)(2a_j - 1) - \sum_i \Delta f(m_i, [L])a_i$$

Possible generalizations

- Mixed receptor cluster: 5 different types of MCP receptors in E. coli cell
- Serine sensing receptor Tsr and the aspartate sensing receptor Tar most abundant, $\sim 90\%$ of the total MCP population
- For a given receptor, $[L]$ stands for the concentration of the ligand to which the receptor binds
- Interaction strength J can also be different for different types of receotors
- J, K_a, K_i may depend on m: may be needed for quantitative agreement with experimental data

The all-or-none Monod-Wyman-Changeux (MWC) model

- An alternative approach for describing receptor cooperativity in the cluster is to divide it into smaller sub-clusters
- Within each sub-cluster, all the receptors are tightly coupled and always in the same state (either active or inactive)
- receptors from different sub-clusters do not correlate with each other at all



- In the MWC model, the degree of cooperativity is given explicitly by N, the size of the all-or-none sub-cluster
- In comparison, the degree of cooperativity in the Ising model can be described by a correlation length which increases with the receptor interaction strength J.
- Free energy difference between all-active and all-inactive states is simply $N\Delta f(m,[L])$
- Average activity $\langle a \rangle = (1+e^{-N\Delta f})^{-1}$

$$\langle a \rangle = \frac{L(1+[L]/K_i)^N}{L(1+[L]/K_i)^N + (1+[L]/K_a)^N} \qquad \textit{where} \;\; L = e^{-Nf_m(m)}$$

- Familiar expression for average activity of an all-or-none MWC complex

Comparison with experiments

- First in vivo measurements of the kinase activity of receptor cluster using FRET [Sourjik and Berg *PNAS* 2002]
- Different stimuli in wild type cells and different mutant strains
- Rich set of quantitative data to test and refine the idea of high cooperativity enabled by receptor clustering
- Experimental data can be explained if Ising type model with mixed receptor clusters are considered [Mello and Tu *PNAS* 2003]
- Effects of variation in the composition of clusters was experimentally studied and theoretically explained using MWC model with homogeneous clusters [Sourjik and Berg *Nature* 2004]
- Heterogeneous MWC model was rigorously developed and implemented to quantitatively explain experimental data [Mello and Tu, *PNAS* 2005]

Adaptation in bacterial chemotaxis

- E. coli chemotaxis is a model system for sensory adaptation
- Robustness vs fine-tuning for exact adaptation
- Fine tuning unrealistic in noisy biological systems
- Barkai-Leibler model: CheR works at saturation and CheB only acts on active receptors
- Assumptions sufficient but not necessary for exact adaptation
- CheR need not work at saturation, as long as it favors the inactive receptors as its substrate [Mello and Tu, *Biophys J* 2003]
- What is the benefit of exact adaptation?

Effect of accurate adaptation: maintaining high gain

- At a background ligand concentration $[L]_0$ and average methylation level m_i the receptors have average activity

$$a_0 = \frac{L_0(1+[L]_0/K_i)^N}{L_0(1+[L]_0/K_i)^N + (1+[L]_0/K_a)^N} \qquad with \quad L_0 = e^{Nf_m(m_i)}$$

- Change in ligand concentration $[L] = [L]_0 + \Delta[L]$ yields an immediate activity change

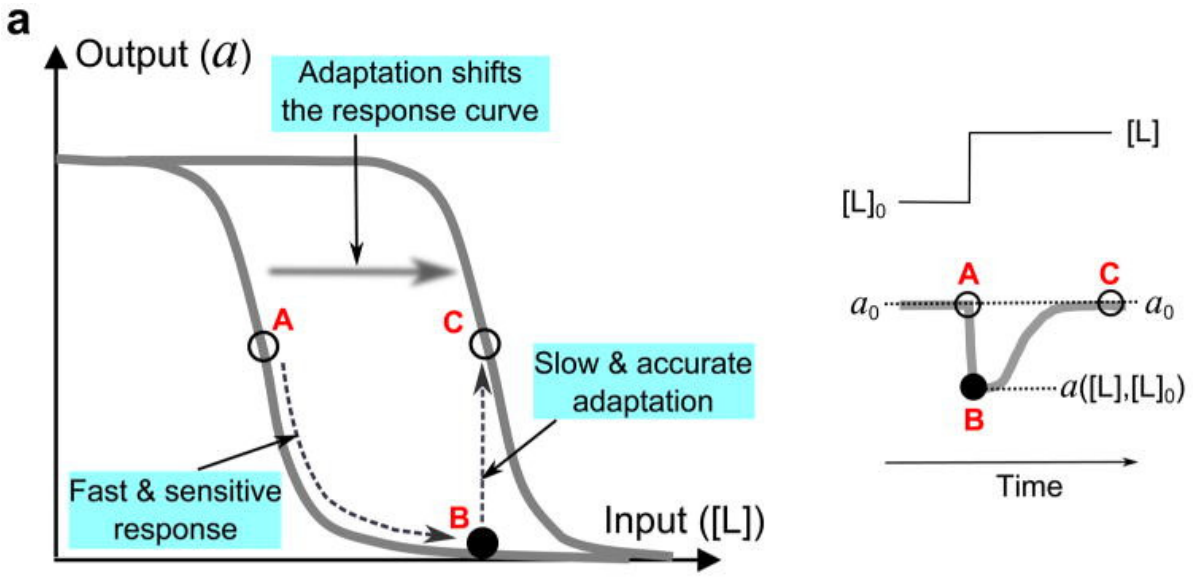
$$\Delta a = \frac{L_0(1+([L]_0+\Delta[L])/K_i)^N}{L_0(1+([L]_0+\Delta[L])/K_i)^N + (1+([L]_0+\Delta[L])/K_a)^N} - a_0$$

- Since m_i changes slowly, L_0 remains unchanged

- For small change in ligand concentration $\Delta[L] \ll [L]_0$

$$\Delta a \approx -N a_0(1-a_0)\frac{(K_i - K_a)\Delta[L]}{(K_i + [L]_0)(K_a + [L]_0)}$$

- Size N of the receptor cluster directly amplifies the response
- Response also depends on the adapted pre-stimulus activity a_0
- Response vanishes at two extreme values $a_0 = 0, 1$ and is maximum around the middle
- However, a_0 depends on $[L]_0$ and remains close to $1/2$ only for very small range of $[L]_0$ if L_0 is fixed
- Therefore, exact adaptation is needed to restore the activity to a_0 in order to maintain high signal amplification or gain
- Essentially, adaptation drives a (slow) change in m and consequently a change in the L_0, which balances the change of the stimulus and restore the activity back to a_0



- Adaptation corresponds to a shift of the response curve so that the adapted activity at the new ligand concentration returns to a_0

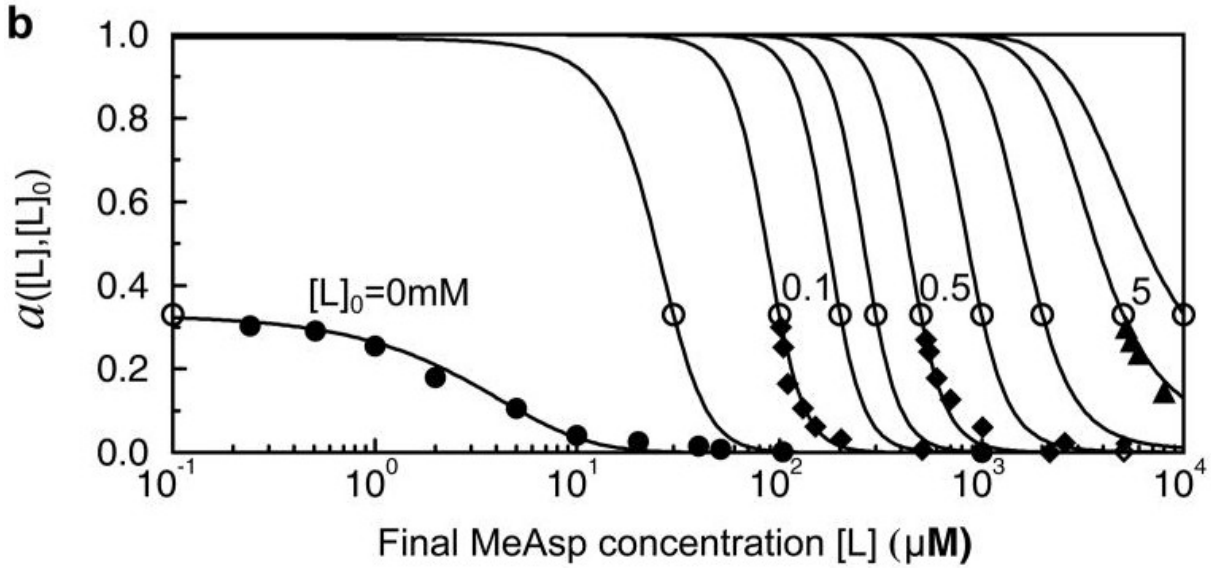
- When the system adapts to the ambient ligand concentration $[L]_0$

$$L_0 = \frac{a_0}{1-a_0} \times \frac{(1+[L]_0/K_i)^N}{(1+[L]_0/K_a)^N}$$

- The value of L_0 can then be used to predict the immediate response of the system upon a change of ligand concentration from $[L]_0$ to $[L]$
- Activity $a([L],[L]_0)$ right after the ligand concentration changes from $[L]_0$ to $[L]$

$$\frac{a_0(K_i+[L]_0)^N(K_a+[L])^N}{a_0(K_i+[L]_0)^N(K_a+[L])^N+(1-a_0)(K_a+[L]_0)^N(K_i+[L])^N}$$

- Only contains four essential parameters, N, a_0, K_a, K_i, each with clear biological definition
- Can be used to fit experimental data



- Response curves for cells that are pre-adapted to different backgrounds $[L]_0$
- Empty circles represent the adapted states
- $N \sim 6$, $K_a = 18\mu M$, $K_i = 3mM$, $a_0 \sim 1/3$