Environmental Sensing and Extracellular Communication

Bacterial signal-transduction systems.

- Origin and diversification.
- Coevolutionary integration of components.
- Emergence of new pathways.

Interconvertible proteins and ultrasensitivity.

- The cost of signal transduction.
- Similarities and differences in eukaryotic systems.

Chemotaxis.

- Accuracy of environmental assessment.

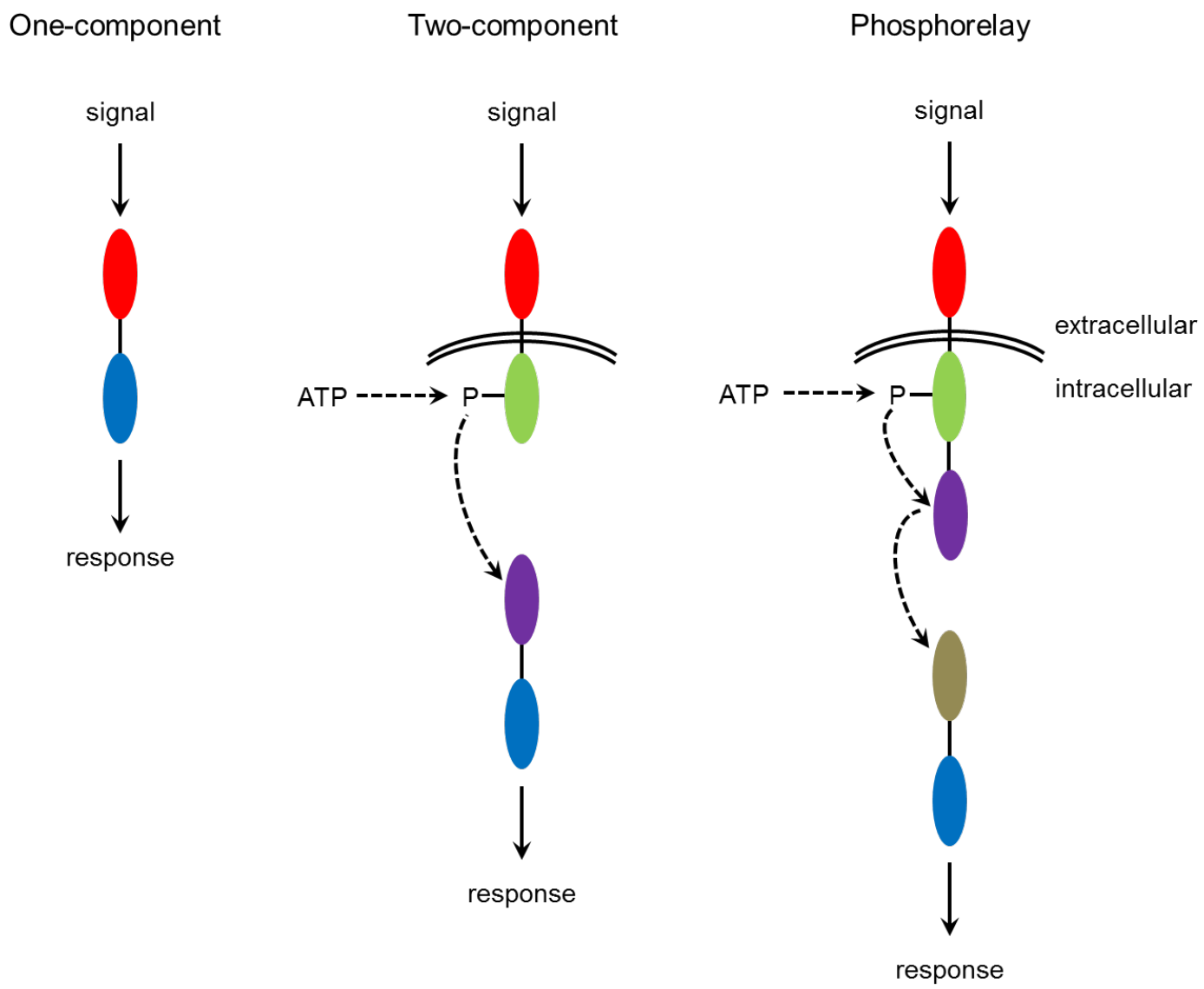
Phenotypic bimodality and bet-hedging.

- Adaptive fine-tuning vs. inadvertent by-products of pathway structure.

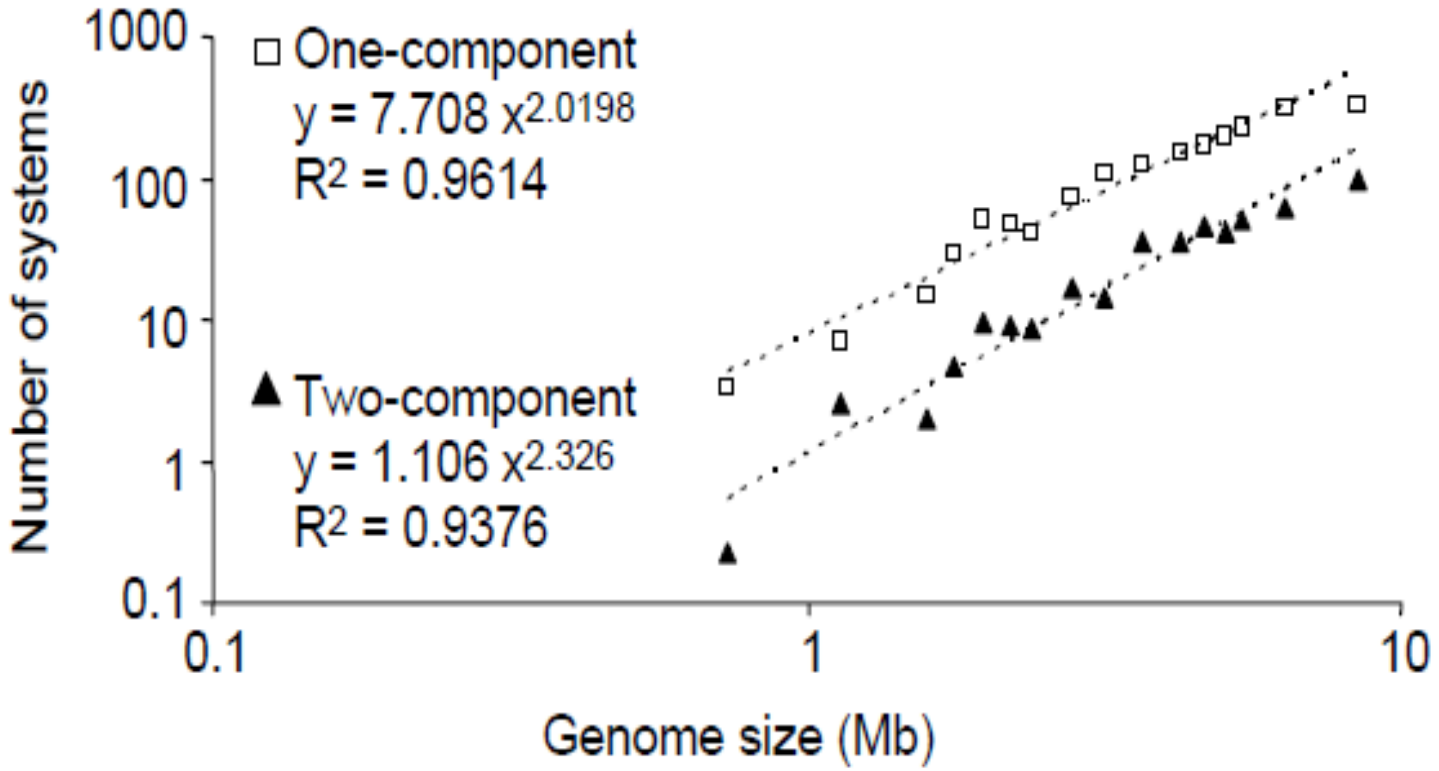
Relatively Simple, Well-Defined Systems Relying on High Molecular-Interaction Specificity:

- Transcription factors and their binding sites.
- Vesicle trafficking machinery – RABs and SNAREs.
- Interfaces of multimeric proteins.
- Bacterial toxin-antitoxin systems.
- Signal-transduction systems for environmental sensing.

Bacterial Signal-Transduction Systems: signal receptors and response regulators

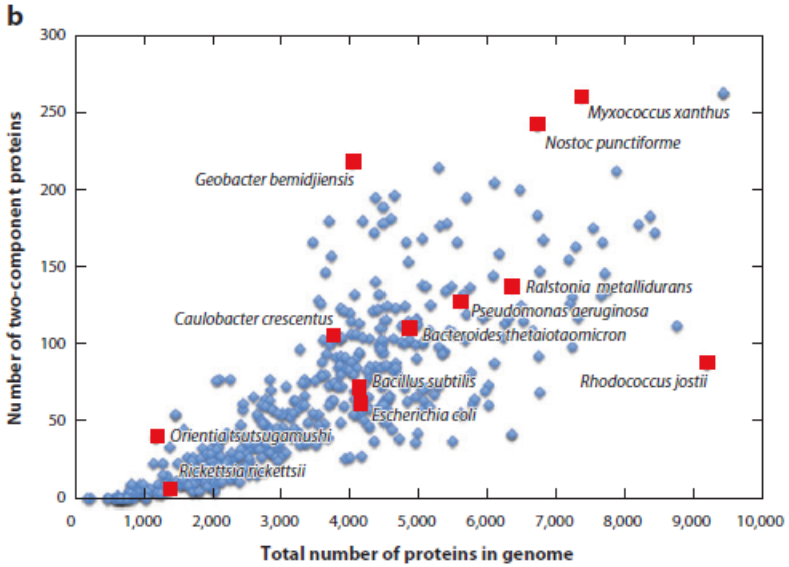


Scaling of Numbers of One- and Two-Component Signal-Transduction Systems in Bacteria



From: Ulrich et al. 2005

Scaling of the Numbers of Two-Component Signal Transduction Systems in Bacterial Genomes



- ~1 to 2% of the genes in most bacterial genomes are associated with signal transduction.

Capra and Laub 2012

Orphan vs. Linked Cognate Pairs of Components

Almost as many orphan components as operon-contained cognate pairs.

Approximately equal numbers of orphan sensors and responders.

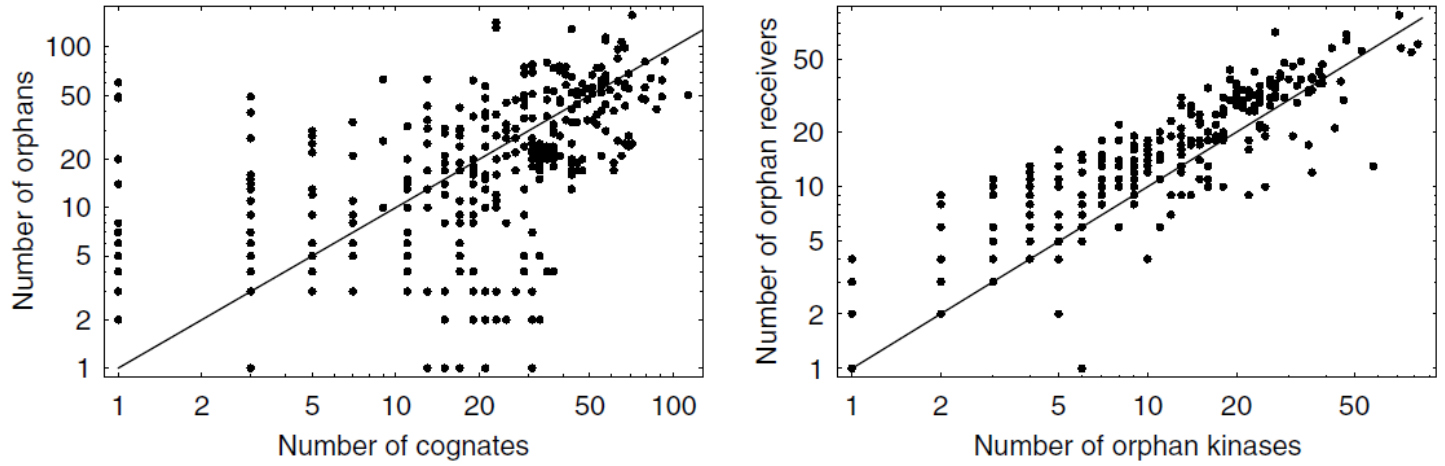


Figure 5 Total numbers of cognates, orphan kinases, and orphan regulators across 399 sequenced bacterial genomes. Left panel: The total number of cognates (horizontal axis) versus the total number of orphans (vertical axis). Right panel: The number of orphan kinases (horizontal axis) versus the number of orphan regulators (vertical axis). Each dot in each panel corresponds to a genome. All axes are shown on logarithmic scale. To be able to show genomes with zero genes in one or more of the categories, 1 was added to each count, that is, one on the axis corresponds to a count of zero.

Burger and van Nimwegen 2008

Simple Recognition-Sequence Motifs

CusS	TNTQEI
YedV	NAGQQV
QseC	TAVGEV
CpxA	TRLGAL
BasS	AGLHEL
BaeS	AVGEEA
PhoQ	AVSTRS
RstB	VRYREM
EnvZ	TRLAEM
PhoR	TVGYEM
KdpD	TVGQEI
CreC	AAGAEI
YfhK	ASEGEL
ZraS	SSGLKY
NtrB	GGGAQL
AtoS	TAGYQI
DpiB	STGLQM
NarX	LSKMVS
NarQ	LSRILT
UhpB	ITRTAG
YehU	NTAVRR
YpdA	NASSRL

- Six primary specificity residues for 22 *E. coli* histidine kinases used in signal transduction.
- Many pairs exist that are identical at three residues.

From: Capra et al. (2012, Cell)

Elevated Rates of AA Substitution in HK-RR Interfaces in Bacterial Two-Component Systems

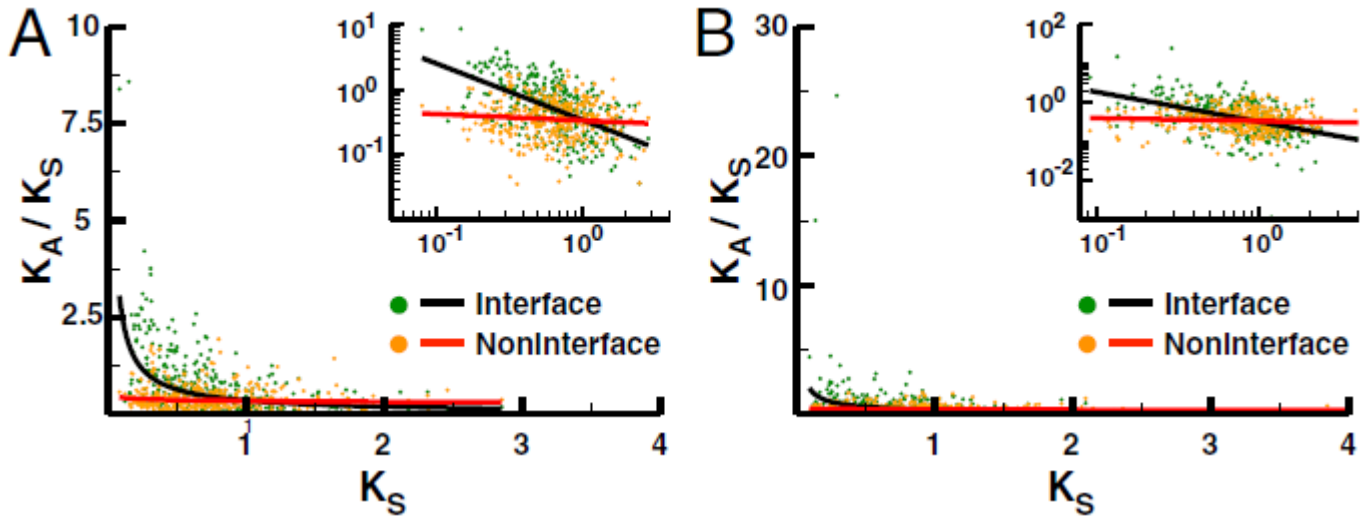
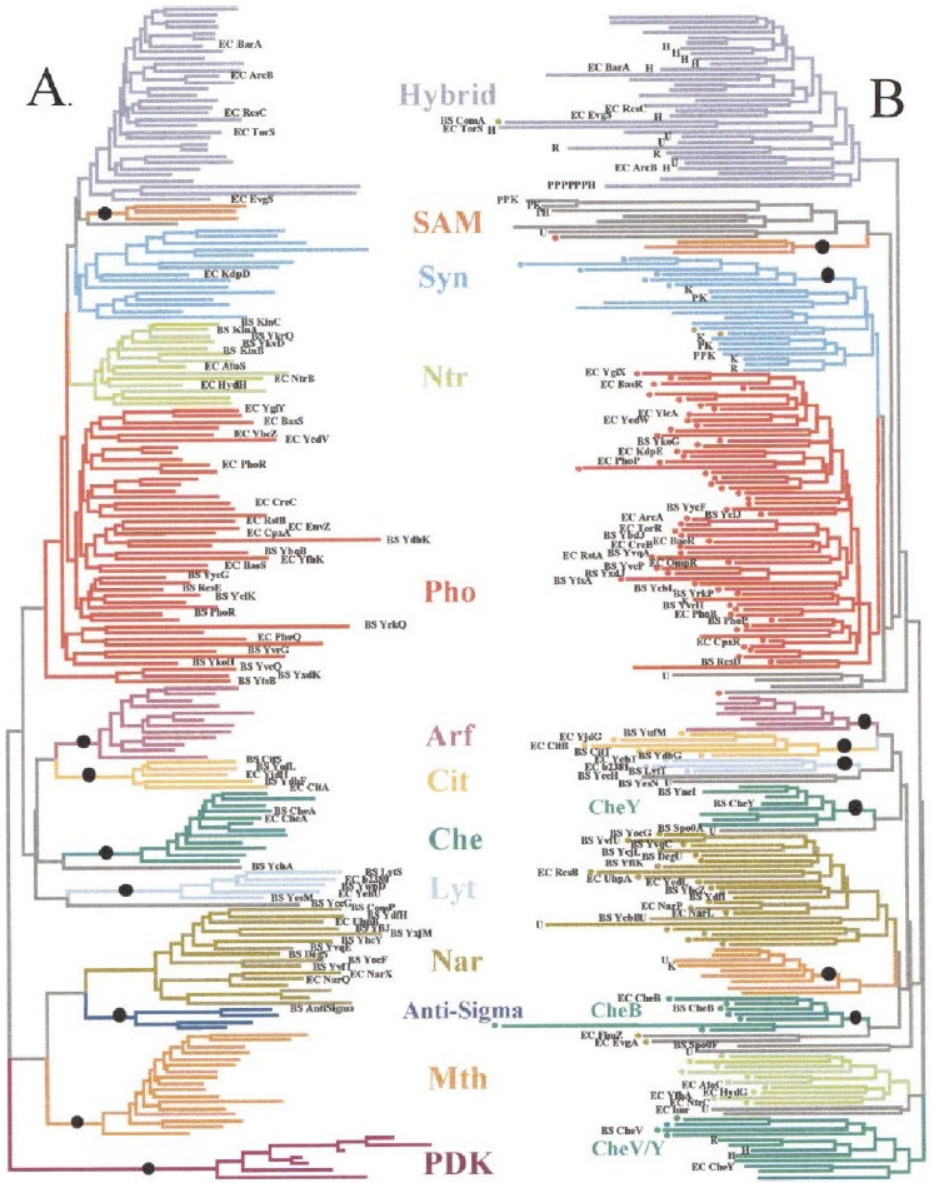


Fig. 4. Sequence analysis. (*A*) The K_A/K_S values as a function of K_S for non-HGT HK sequence pairs for both the K domain interface residues (green circles) and noninterface residues (orange circles). The black and red lines correspond to power-law regressions of the interface and noninterface data, respectively. (*Inset*) The same data and fits, plotted on a log-log scale. (*B*) A plot similar to that in *A*, but for the interface and noninterface residues of RR proteins. (*C*)

- A consequence of frequent evolutionary modifications for new cross-talk partners?

From: Rowland and Deeds 2014

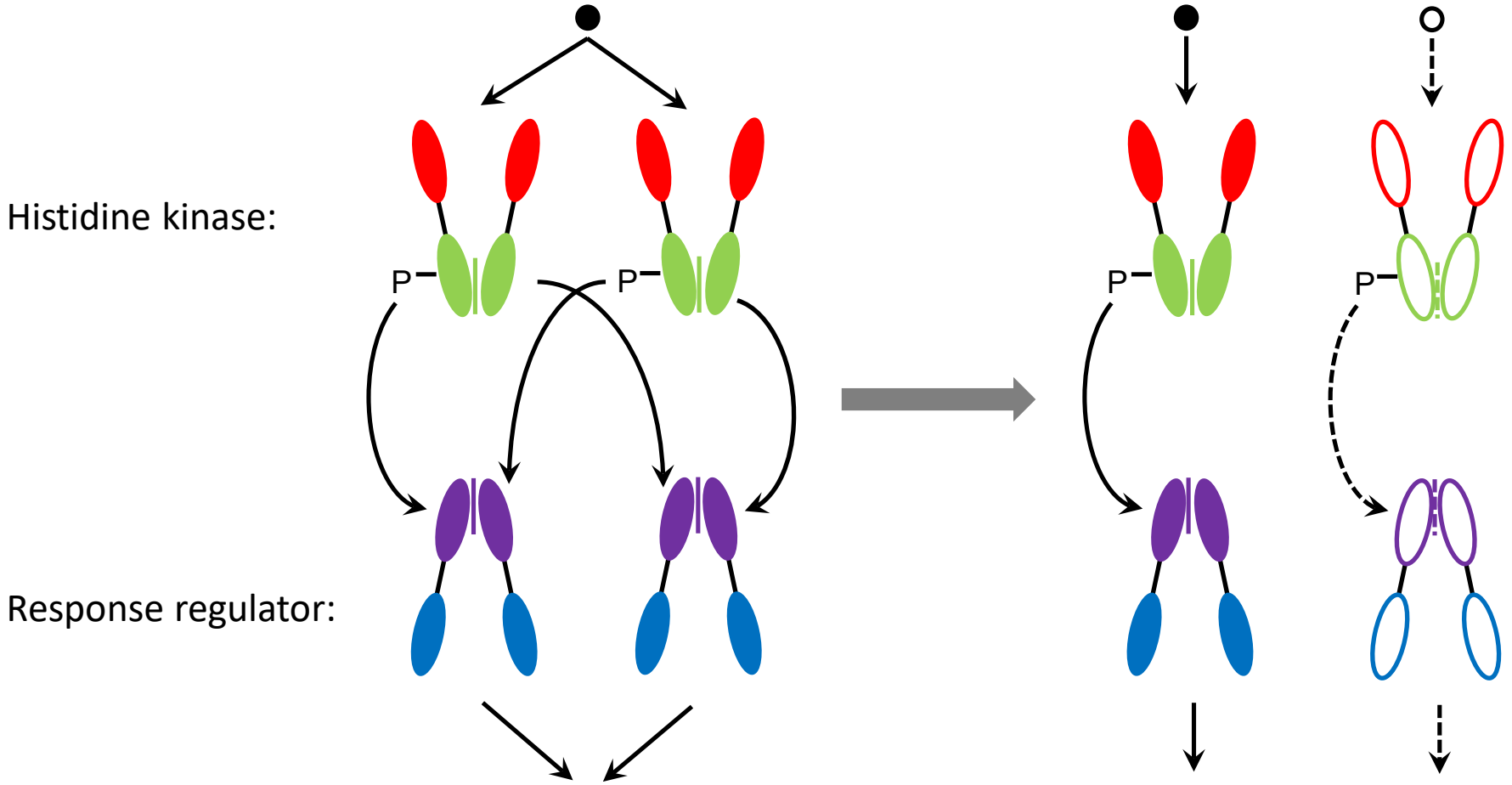
Largely congruent phylogenies of sensory histidine kinases and their cognate response regulators suggests the origin of new systems by operon duplication and divergence.



From: Koretke et al. 2000

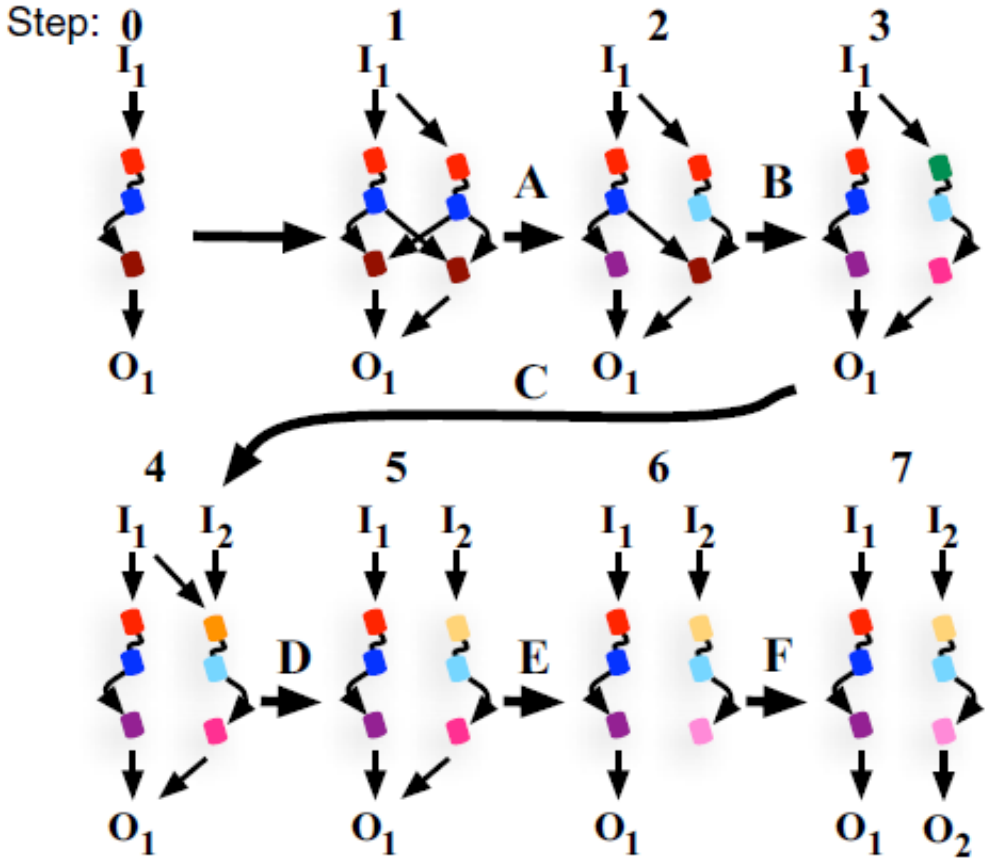
Evolution of a New Two-Component System by Gene Duplication

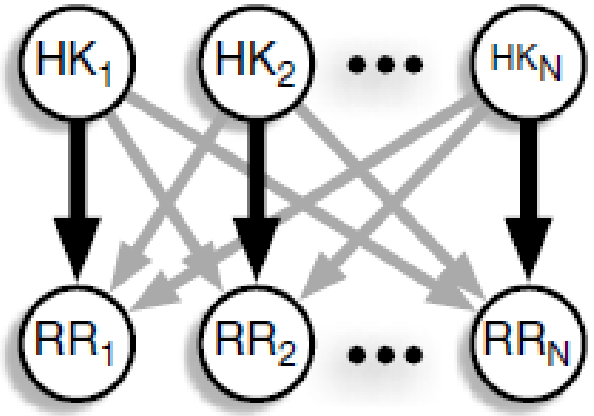
- Avoiding cross talk.
- Avoiding heterodimerization.
- Preservation by gain of a new favorable function?



Neutral Paths to the Origin of New Two-Component System by System Duplication

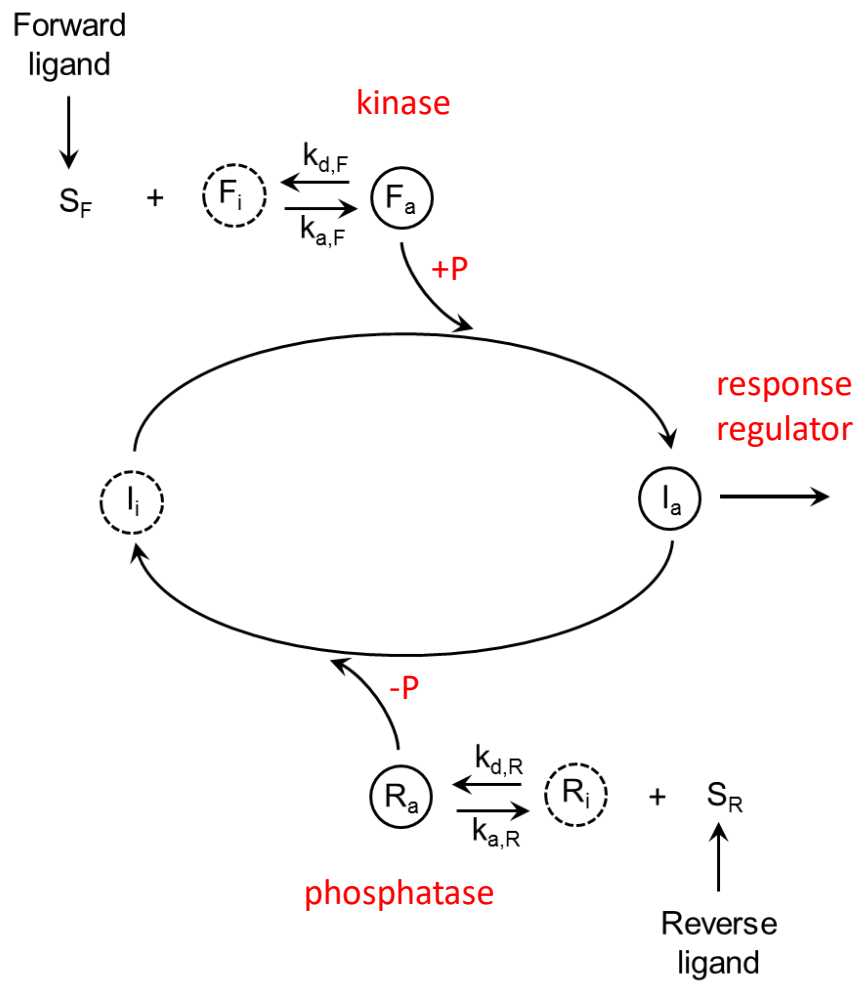
- All crosstalk must be eliminated prior to the origin of a new function?





From: Rowland and Deeds 2014

Interconvertible Proteins and Ultrasensitivity



$$I_i + ATP \xrightarrow{\text{kinase, } F_a} I_a + ADP$$

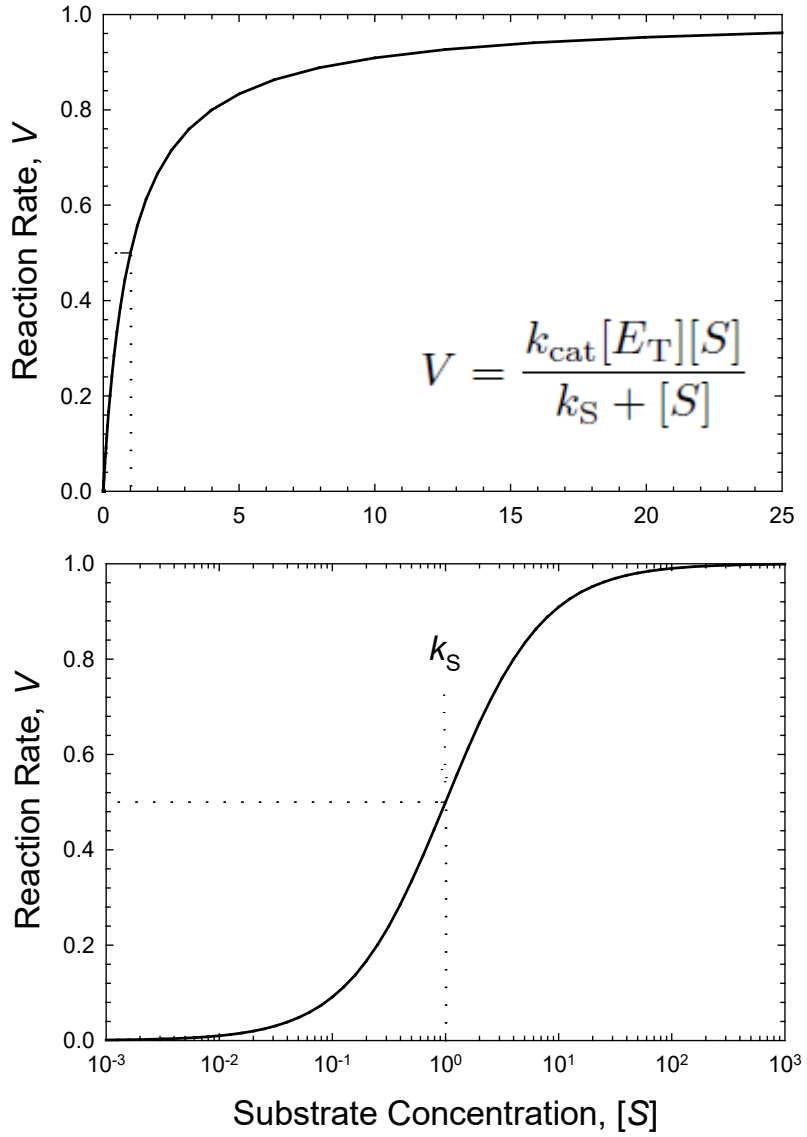
$$I_a + H_2O \xrightarrow{\text{phosphatase, } R_a} I_i + Pi$$

- Given fixed concentrations of ligands and converting enzymes, the fraction of active enzyme I_a reaches a steady state, with the rates of forward and reverse conversion being equal.
- Unlike the single M-M equation with two parameters, 10 parameters govern the behavior.
- Increased sensitivity to low external ligand concentrations.
- Altered amplitude of response.

Michaelis-Menten Enzyme Kinetics

$$\mathrm{E} + \mathrm{S} \rightleftharpoons \mathrm{ES} \rightarrow \mathrm{E} + \mathrm{P}$$

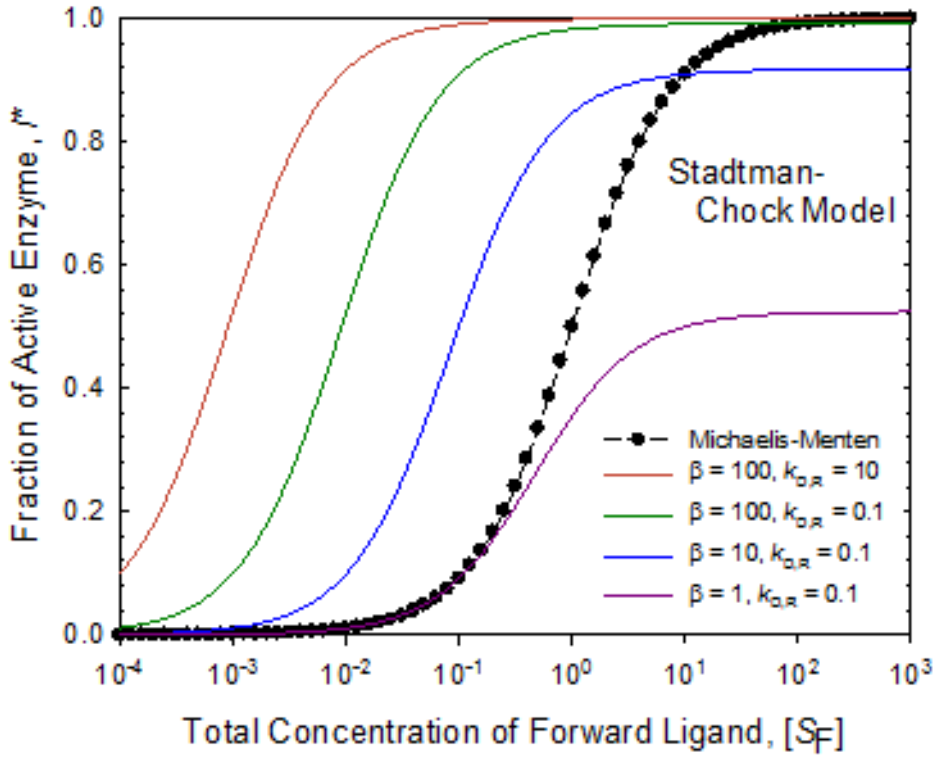
$$k_\mathrm{S} = (k_\mathrm{d} + k_\mathrm{cat})/k_\mathrm{a}$$



Altered Kinetics of an Interconvertible Enzyme

When concentration of interconvertible enzyme is low:

- The behavior of the system is still hyperbolic, with the "half-saturation" constant and the amplitude defined by the amounts and kinetic efficiencies of the two converter enzymes.



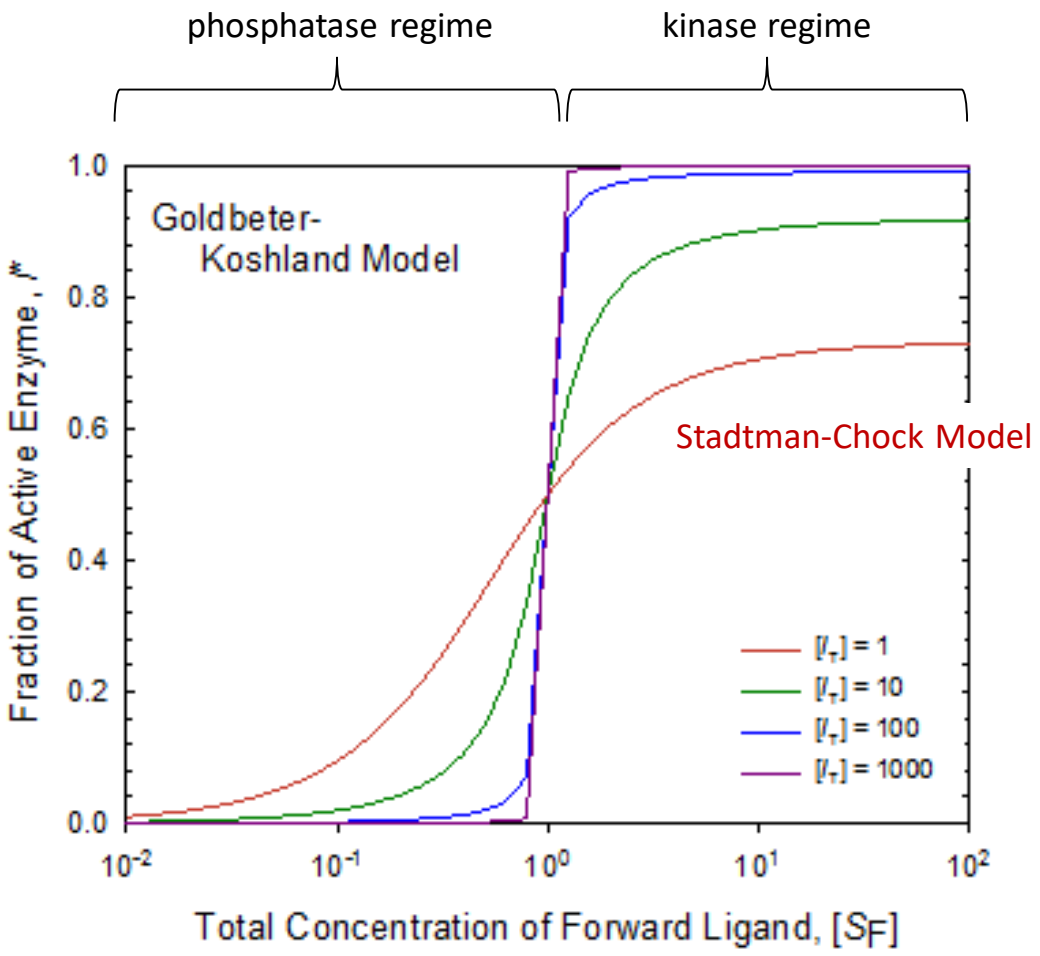
$$I^* = \frac{\beta C}{1 + \beta C},$$

$$\beta = \frac{\phi_{\mathrm{F}}[F_{\mathrm{T}}]}{\phi_{\mathrm{R}}[R_{\mathrm{T}}]}$$

$C = \dfrac{[S_{\mathrm{F}}](k_{\mathrm{D,R}} + [S_{\mathrm{R}}])}{[S_{\mathrm{R}}](k_{\mathrm{D,F}} + [S_{\mathrm{F}}])}$ is the ratio of degrees of saturation of the input reactions.

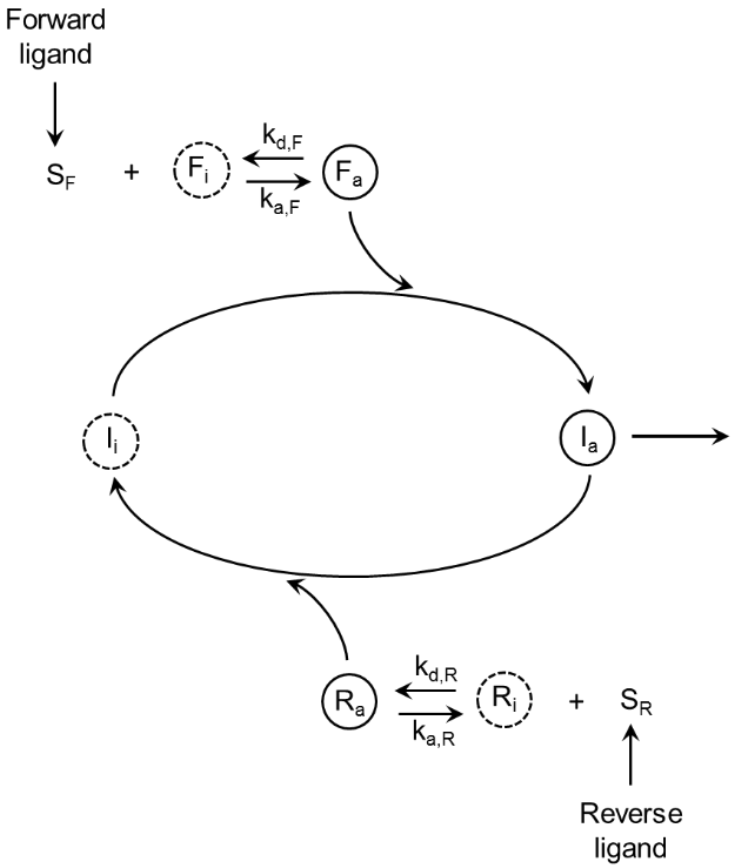
$\phi_x = k_{\mathrm{cat,x}}/k_{\mathrm{S,x}}$ being the specificity constant of enzyme x

Zero-order Ultrasensitivity / Switch-Like Behavior



- When the concentration of interconvertible enzyme is high, the converter enzymes become saturated, sharpening their responses near the threshold between the kinase and phosphatase domains on the scale of ligand concentrations.

Cost of Signal Transduction: Maintaining an Interconvertible Enzyme



- Steady-state rates of phosphorylation / dephosphorylation:

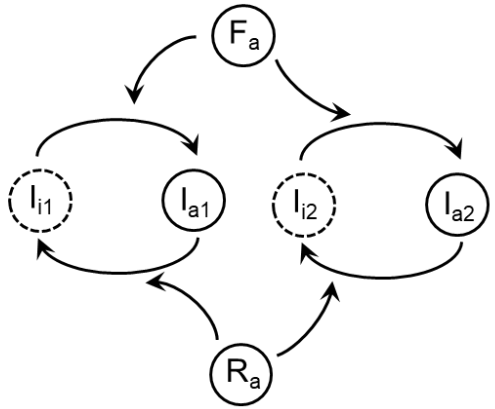
$$V_{\mathrm{ATP}} = \frac{k_{\mathrm{cat,R}}[R_{\mathrm{a}}][\mathrm{I}^*]}{k_{\mathrm{R}}^* + [\mathrm{I}^*]} = \frac{k_{\mathrm{cat,F}}[F_{\mathrm{a}}](1 - [\mathrm{I}^*])}{k_{\mathrm{F}}^* + 1 - [\mathrm{I}^*]}$$

- Pyruvate kinase in liver, V_{ATP} = 20 to 200 uM / minute.
- Accounting for the volume of a cell and cell-division time, total expenditure per cell cycle is 10^{10} to 10^{12} ATP hydrolyses.
- ~5% of cell's basal maintenance requirements.
- Cost of construction of the enzymes is ~10^{11} ATPs.

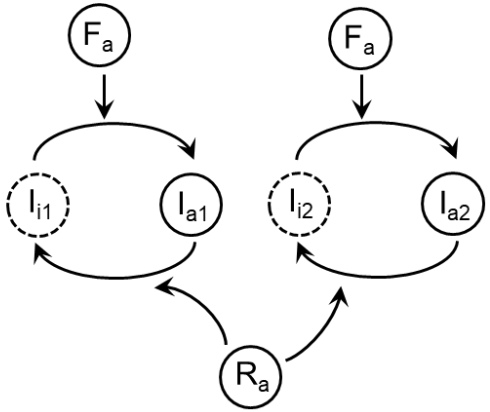
Common Variants in Eukaryotic Systems

- Typical reliance on phosphorylation of serine, threonine, and tyrosine rather than histidine.

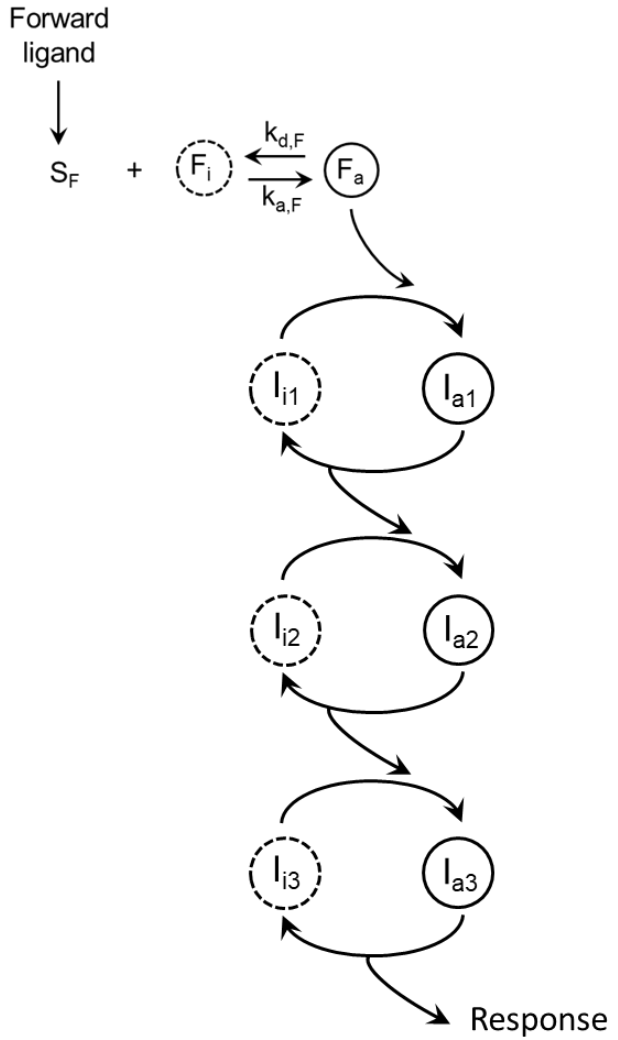
- Enzymes often engage with multiple substrate proteins, leading to more complex networks.



A kinase and a phosphatase sharing two intermediate substrates

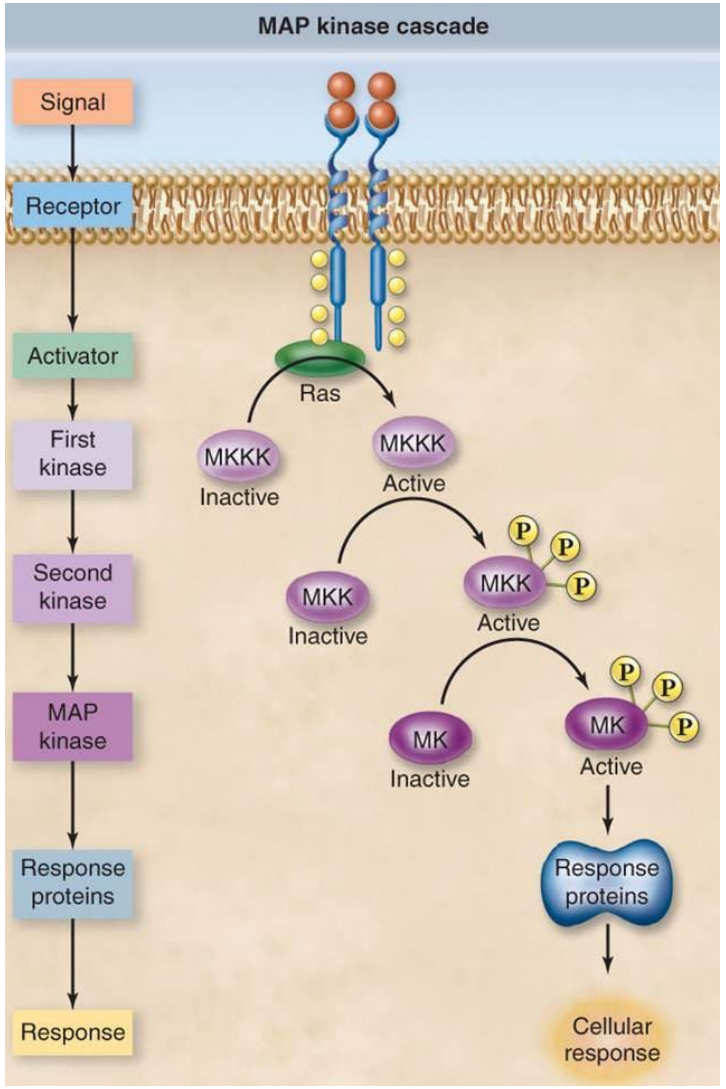


Two independently acting kinases, with one shared phosphatase

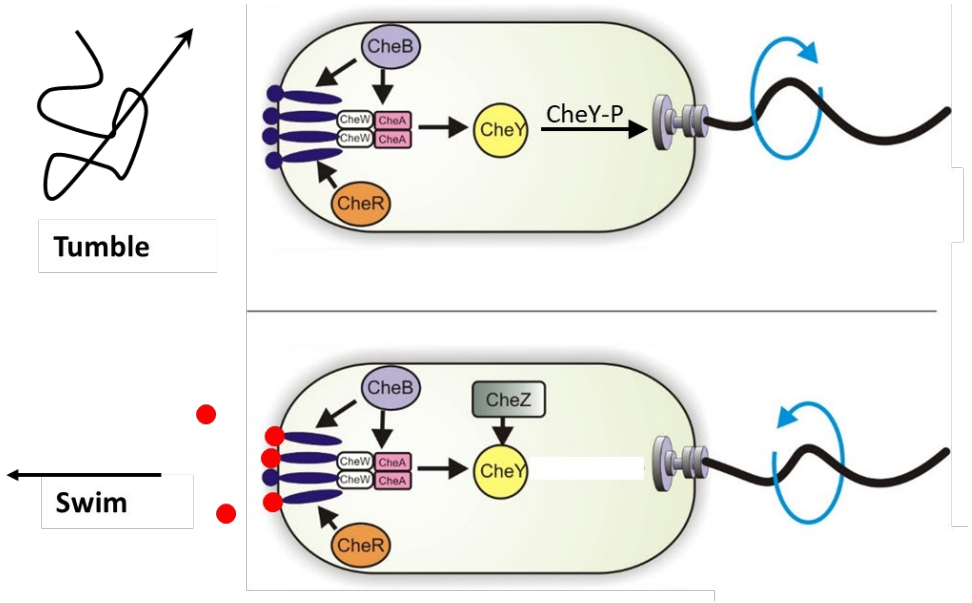


Triple cascade

Eukaryotic Kinase Cascades: Why should such long chains evolve, and why are they largely absent from prokaryotes?



Chemotaxis



- Directs motility towards particular chemo-attractants (or away from repellants).
- Although such systems have a kinase and a response regulator at their core, the output of the system is modulated by up to nine other participants

- Array of ~10,000 methyl-accepting chemotaxis proteins relay information to CheA (the hisitidine kinase).
- Response regulator (CheY) binds directly to base of flagellum, yielding a much more rapid response than transcription regulation.
- Methyltransferases and methylesterases modify the sensitivity in response to the environment, like an eye adjusting to the light.

Evolutionary Issues

- ~50% of bacteria have a chemoreceptor system.
- Arrays can contain 1 to ~30 types of receptors.
- Directional swimming instructions have flipped between *Escherichia* and *Bacillus*.

- Unlike bacteria, which monitor their environment in a temporal fashion, larger eukaryotic cells (e.g., the slime mold, *Dictyostelium*) often populate their entire cell surface with receptors, enabling sensing of spatial variation between the two ends of the cell.

Accuracy of Environmental Monitoring

- Berg and Purcell (1977): degree of occupancy of receptor as a counting mechanism.

$$p = c_0/(K_D + c_0).$$

c_0 = environmental concentration of the ligand
K_D = dissociation constant

Maximum sensitivity occurs with high K_D, as $p \approx c_0 / K_D$, a linear response.

- Error in inference of the environmental concentration (coefficient of variation of inferred concentration):

r_s = radius of receptor

$$\frac{\sigma_c}{c_0} = \sqrt{\frac{1}{2Dr_s c_0(1-p)T}}$$

rate of particle diffusion to a receptor

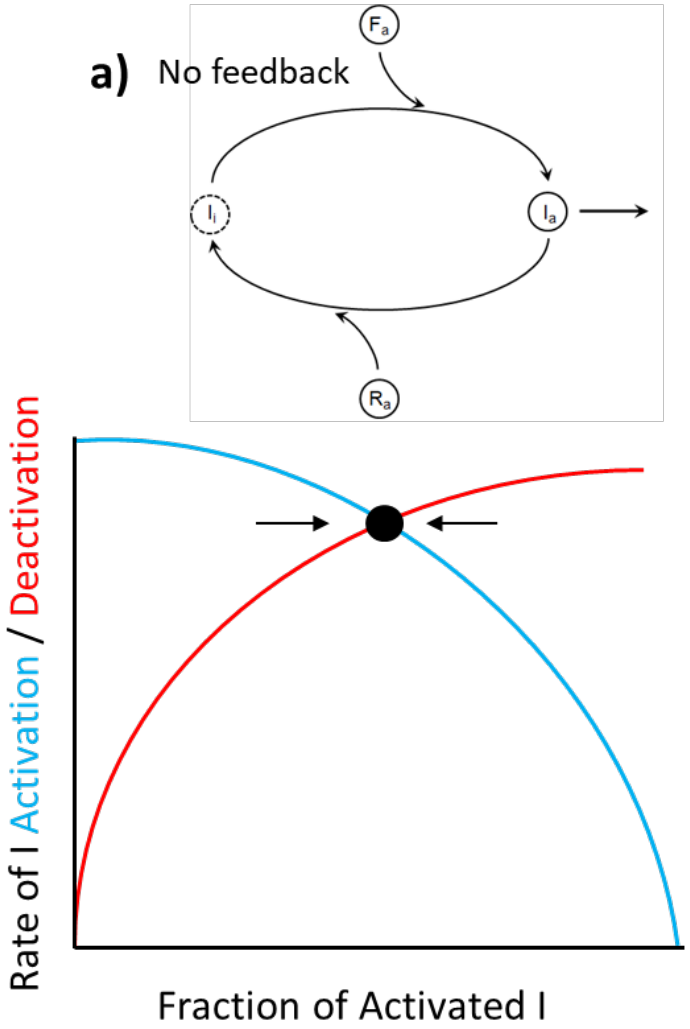
monitoring time

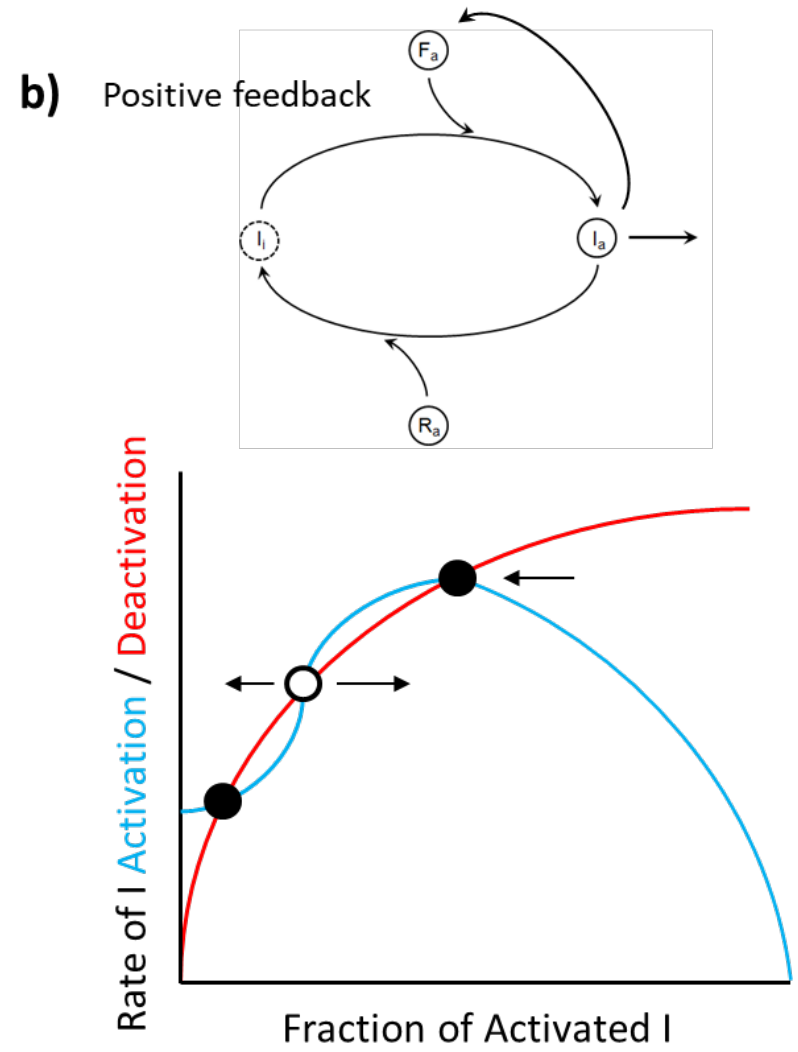
- For typical ligand concentrations, a few seconds of monitoring can reduce the CV (error rate) to ~0.01.

Unanswered Evolutionary Questions

- Relative to the situation with the nervous systems of metazoans, how much of the energy budgets of single-celled organisms is devoted to environmental monitoring and decision making?
  - What is the optimal allocation of resources to environmental sensors for balancing the costs of production of such molecules and the advantages accrued?
- To what extent does the investment in sensing increase with increasing variance in environmental states?
  - At what point does the energetic cost of the maintaining such systems offset the advantage of an ability to track environmental changes, such that sensory overload necessitates the evolution of a constitutive general-purpose genotype?

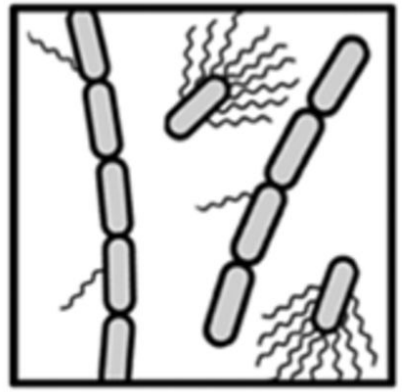
Phenotypic Bimodality and Bet-Hedging





- An unstable equilibrium can lead to sustained bimodality in the absence of genetic variation.

Adaptive Fine-Tuning vs. Inadvertent By-Products of Pathway Structure?



Bacillus subtilis: motile vs. chained forms (Norman et al. 2013).

- Motile state is “memory-less”: switches to chained state with constant probability at each cell division, giving a mean switching time of 81 cell divisions – due to stochastic molecular fluctuations.

- Switching from chains to motile form: pulse of matrix material followed by progressive dilution, giving a mean of 8 cell divisions.

- Multimodality is not always advantageous, and can be disadvantageous:

  - Genotypes with the highest long-term exponential growth rates are favored.

  - If the selection differentials in two environments are substantially different, stochastic switching can be disfavored, as the monomorphic type favored in a common environment with large effects can overwhelm the smaller, short-lived disadvantage in the opposite environment.