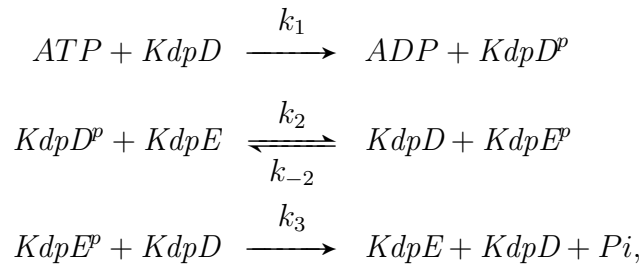


Systems Biology, Homework # 6

Signal Transduction Pathways

Due Wednesday April 13th 11:59 pm

1. **The two-component *KdpD/KdpE* signalling pathway.** When cells of the bacterium *E. coli* need to increase the rate at which they take up K^+ ions from the environment, they increase production of a high affinity K^+ uptake system. Production of this system is under the control of the protein *KdpE*, which is the response regulator in a two-component signalling pathway. *KdpE* is activated by a sensor protein called *KdpD*. Activation of *KdpE* is a two-step process: first, activated *KdpD* undergoes autophosphorylation; next, the phosphate group is transferred to *KdpE*. Inactivation is also mediated by *KdpD*; it acts as a phosphatase, removing the phosphate group from activated *KdpE*. In a 2004 paper, Andreas Kremling and his colleagues published a model of the *KdpD/KdpE* pathway (Kremling, A., Heerman, R., Centler, F., Jung, K., & Gilles, E. D. (2004). Analysis of two-component signal transduction by mathematical modeling using the KdpD/KdpE system of Escherichia coli. Biosystems, 78, 23–37). A simplified version of their model network is



where p indicates phosphorylation, and Pi is a free phosphate group. The parameter k_1 can be used as an input to the system.

- (a) Treating the concentration of ATP as constant, write a set of differential equations describing the system behaviour. (Do not include descriptions of the ATP , ADP or Pi dynamics). Take the total concentrations of the proteins (in the unphosphorylated and phosphorylated states) to be fixed at $KdpE_T$ and $KdpD_T$ respectively.
- (b) The parameter values reported by Kremling and colleagues are: (in μM) $[ATP]=1500$, $KdpE_T=1$, $KdpD_T=1$, (in $\mu M^{-1} h^{-1}$) $k_1 = 0.0029$, $k_2 = 108$, $k_{-2} = 1080$, $k_3 = 90$. This value of k_1 corresponds to an activated sensor. Run a simulation from initial condition of inactivity (no phosphorylation). How long does it take for the system to reach its steady-state response? Run another simulation to mimic inactivation of the activated system (i.e. by decreasing k_1 to zero from the active steady state). Does the system inactivate on the same time-scale?
- (c) Kremling and colleagues conducted *in vitro* experiments on this system at a low ATP level of $[ATP]=100 \mu M$. How does this change affect the system behaviour? Does it impact the activation and inactivation time-scales?

- (d) How would the system behaviour be different if, instead of $KdpD$, an independent phosphatase (with fixed activity level) inactivates $KdpE^p$? Does the fact that $KdpD$ act as as both kinase and phosphatase enhance or diminish the system's response? (Note, only the unphosphorylated form of $KdpD$ has phosphatase activity.) Confirm your conjecture by simulating a modified model in which an independent phosphatase inactivates $KdpE^p$.