Announcements

Office hours this week:
4-5 pm (right after section) on Tuesday the 1st

Wednesday from 7-8:30 pm on Zoom

or by appointment.

Section next week:
In-person on November 8th at 3 pm in M217.

Paper of the week
10.1126/science.abo7651
A new integrated semiconductor platform for single-molecule sequencing of short peptides with that can discriminate between amino acids using the lifetimes of different protein-based N-terminal binders. Very cool work!

Problem 1: Activation and repression curves for genetic circuits

In this problem, we construct the binding curves for activators and repressors of transcription, which can be applied to a variety of problems involving biological circuits.

a) Let's first consider a repressor protein X and its target DNA sequence D. The association rate of the two is $k_{on}[X][D]$, while the dissociation rate is $k_{off}[XD]$. In addition, we apply the constraint that the total number of DNA binding sites must remain constant: $[XD]+[D]=D_T$. Using this information, derive an expression for the fraction of DNA binding sites that are free at steady state.

b) Letting β denote the rate constant for the promoter of gene D, compute the promoter activity (i.e. the concentration times the rate constant). Sketch your result as a function of $[X]$.

c) Let's now instead consider a cooperative model of transcriptional activation. In this model, n molecules of protein S_D associate and then bind to D. Letting k_{on} and k_{off} denote the on and off rate of this system, respectively, write down the rate equations for nS_DD and conservation law for D.

d) Using the same procedure as previously, derive the fraction of DNA binding sites that are bound by the inducer. You should find that the solution is:

$$\frac{[nS_DD]}{D_{tot}} = \frac{[S_D]^n}{K_D^n + [S_D]^n}. \tag{1}$$

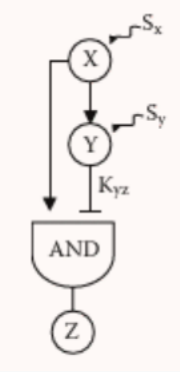
Make sure to specify the value of K_D^n.

e) Sketch your result from part (d) as a function of $[S_D]$ for different values of n. You might try $n = 1, 2, 4$. Can you describe what's happening to the curve.

This is a variant of the Hill equation, which is an extremely useful model of cooperative binding. Note that in practice, the exponent in the binding curve will always be less than n.

Problem 2: The incoherent type-1 feed-forward loop

Here we consider a common motif in a variety of gene regulatory networks. In this motif, the product of gene X has two effects when it is active: it activates gene Z and, in parallel, activates gene Y, whose product represses gene Z. The proteins produced by X and Y are denoted X^* and Y^*, respectively, when activated by proteins S_x or S_y.



Activating gene X therefore triggers a pulse of Z transcription. In this problem, we will examine these dynamics in more detail.

a) Initially, all protein products have concentration 0. When we suddenly increase the level of S_x, the product of gene X is activated and transitions to X^*. We can approximate this transition as a step function at $t = 0$. X^* binds to the promoter of Y, whose product can then be activated by S_y. The dynamics of the activated product are governed by the rate equation:

$$\frac{dY^*}{dt} = \beta_y - \alpha_y Y^*. \tag{2}$$

Find the solution to this differential equation and plot Y^* as a function of t.

b) When X is active, Z is also produced. Its dynamics are initially governed by the rate equation:

$$\frac{dZ}{dt} = \beta_z - \alpha_z Z. \tag{3}$$

To simplify calculations, we will make the so-called **logic approximation**, in which we replace the sigmoidal response curves you derived in the previous part with step functions at K. Using this approximation, once the concentration of Y^* reaches K_{yz}, the rate of Z production instantaneously drops to β_z'. Using your result from (a), find the time at which this occurs T_{rep} as a function of the rate constants for Y^* and K_{yz}.

c)Find an equation for the concentration of Z for $t > T_{rep}$. Combine this with your equation for $0 < t < T_{rep}$ to describe the dynamics of Z for all time. *Hint:* you shouldn't need to separately solve for the equation when $0 < t < T_{rep}$ and can instead figure this out from your result in (a).

d) We can define the repression factor F as the ratio of the steady state of Z prior to T_{rep} to the steady state after T_{rep}. Sketch the curve of Z for all $t > 0$ for $F = 2, 4,$ and 8.