Physics of Behavior
Uri Alon
Exercise 6

**1. Repressilator**: Three repressors regulate each other in a cycle: X represses Y, Y represses Z and Z represses X. Z also represses a green-fluorescent protein (GFP) gene which is a readout of the circuit.
a. Sketch the circuit diagram.
b. Sketch the oscillatory dynamics of X,Y and Z as a function of time.
c. An experiment by Elowitz (2000) built this circuit in *E coli*. In a test-tube, the bacterial culture showed a constant green fluorescence when measured in a fluorimeter. But when looking at individual cells in the microscope, each cell was seen to have production of green fluorescence that oscillated with a period of about 8h. Explain.
d. 8h is longer than the cell generation time of 2h. How could the individual cells show oscillations? Sketch a cell "family tree" to illustrate.

***2.*** **Cell-cycle oscillator**. The frog (*Xenopus*) embryonic cell-cycle is driven by a protein circuit that acts like an autonomous oscillator. It can be described by the following two-ODE model.

$$\frac{dx}{dt} = \alpha_1 - \beta_1\, x \frac{y^{n_1}}{k_1^{n_1} + y^{n_1}} + \alpha_3\,(1-x)\frac{x^{n_2}}{k_3^{n_2} + x^{n_2}}$$

$$\frac{dy}{dt} = \alpha_2(1-y)\frac{x^{n_3}}{k_2^{n_3} + x^{n_3}} - \beta_2\, y$$

Here $x$ is the fraction of active $CDK1$ and $y$ is the fraction of active $APC^*$.

a. Explain the meaning of each of the parameters in the equations.
b. Simulate the dynamics using a computer. Search for oscillations as a function of the parameter $\alpha_1$ (throughout this exercise use: $\alpha_2 = \alpha_3 = \beta_1 = 3,\ \beta_2 = 1,\ k_1 = k_2 = k_3 = 0.5$, $n_1 = n_2 = n_3 = 8$). Plot x(t) for a value $\alpha_1$ which admits a stable steady state and for a value of $\alpha_1$ which admits stable oscillations.
c. Plot {x(t), y(t)} on the phase plane (plot y(t) versus x(t)) for both values of the parameter $\alpha_1$ starting at the initial condition x=y=0.5.
d. What effect does changing $\alpha_1$ has on the behavior of the oscillations (look at amplitude and frequency)? Plot the amplitude and frequency as a function of $\alpha_1$, using numerical simulations at several values of $\alpha_1$.

3. **Optimality with limiting substrate.** Protein $\mathrm{X}$ is an enzyme that acts on a substrate to provide fitness to the organism. The substrate concentration is $\mathrm{L}$.

a. Calculate the fitness function $\mathrm{f(X, L)}$ assuming linear cost, $\mathrm{c} \sim -\eta \mathrm{X}$, and a benefit that is a Michaelis–Menten term, $\mathrm{b(L, X)} = \mathrm{b_0\, L}\frac{\mathrm{X}}{\mathrm{X+K}}$, appropriate for cases where the substrate, rather than the enzyme $\mathrm{X}$, is limiting.
b. Plot the fitness function for various values of L.
c. Calculate the optimal enzyme level $\mathrm{X_{opt}}$ as a function of $\mathrm{L}$ and $\mathrm{K}$.
d. What is the minimal substrate level $\mathrm{L_c}$ required for maintenance of the gene for X by the organism? When is the gene lost? Explain.

4. **Optimal expression of a subunit.** Multiple units of protein X act together in a multi-unit complex. The benefit is a Hill function, $b(X) = b_0 \frac{X^n}{K^n + X^n}$, and the cost function is linear in $X$. What is the optimal protein level? Explain.

5. **Global warming.** An organism with a generation time of $T$ years has a fitness that depends on a trait $X$. The fitness function $F(X)$ is shaped like a mountain with a maximum, $F(X) = F_0 - F_1\left(X - X_{opt}\right)^2$. When temperature rises, the position of the fitness maximum shifts. The position of the maximum $X_{opt}$ rises by $q\%$ per degree of warming. Natural selection is able to change $X$, but not faster than a maximal rate of $s\%$ per generation.
a. Sketch the fitness landscape at two different times.
b. Discuss how the fitness of the species will change with time, assuming that temperature was constant for a long time, and then begins to rise at a rate of $d\%$ per year.
c. What effect does the generation time $T$ have? Are organisms with long generation times more or less vulnerable to the rise in temperature?