Read. *Mathematical Concepts in Modern Biology*, by Robeva/Hodge. Ch. 1: Boolean models of the *lac* operon in *E. coli*, and Ch. 2: Bistability in the *lac* operon: a comparision of ODE and Boolean models, pp. 1–74.

Exercises.

1. Consider the reactions where two substrates S and T compete for binding to an enzyme E to produce two different products P and Q:

$$E + S \underset{p_2}{\overset{p_1}{\rightleftharpoons}} ES \xrightarrow{p_3} P + E$$
$$E + T \underset{q_2}{\overset{q_1}{\rightleftharpoons}} ET \xrightarrow{q_3} Q + E$$

   Assume that each reaction follows the Michaelis-Menten kinetics. Also, assume that that the initial enzyme concentration is $E_0 = [E] + [ES] + [ET]$.

   (a) Derive rate equations for P and Q in this system in terms of $[ES]$ and $[ET]$. That is, determine $d[P]/dt$ and $d[Q]/dt$.

   (b) Derive rate equations for ES and ET.

   (c) Assume that the enzyme-substrate complexes reach equilibrium quickly: $d[ES]/dt \approx 0$ and $d[ET]/dt \approx 0$. Solve for $[E]$ in each of these equations.

   (d) Equate the two expressions for $[E]$ from Part (c) and solve for $[ET]$.

   (e) Solve for $[ES]$ by plugging your answers to Parts (c) and (d) into $E_0 = [E] + [ES] + [ET]$. You should not have $[E]$ or $[ET]$ in your final answer.

   (f) Plug this into the original ODE for $d[P]/dt$.

   (g) Derive an analogous ODE for $d[Q]/dt$.

   (h) Explain the effects of the competition occuring.

2. The Hill equation is an approximation for multi-molecule binding and it assumes simultaneous binding of n-moecules of a substrate S to the enzyme E. Suppose that two molecules of the substrate S are undergoing a reaction with an enzyme in an ordered manner as follows:

$$E + S \underset{k_2}{\overset{k_1}{\rightleftharpoons}} ES, \qquad ES + S \underset{k_4}{\overset{k_3}{\rightleftharpoons}} ES_2 \xrightarrow{k_5} P + E.$$

   Assume that the reaction follows the Michaelis-Menten kinetics and that the initial enzyme concentration is $E_0 = [E] + [ES] + [ES_2]$.

   (a) Derive rate equations for P, ES, and ES_2.

   (b) Assume that $d[ES_2]/dt \approx 0$ and solve for $[ES]$.

   (c) Assume that $d[ES]/dt \approx 0$. Plug your answer to Part (b) into this and solve for $[E]$.

(d) Plug your expressions for $[E]$ and $[ES]$ back into $E_0 = [E] + [ES] + [ES_2]$ and solve for $[ES_2]$.

(e) Derive an ODE for $[P]$ of the form $d[P]/dt = f([S])$.

(f) Compare your answer to Part (d) to the Hill equation with Hill coefficient $n = 2$:

$$\frac{d[P]}{dt} = \frac{V_{\max}[S]^2}{K_m + [S]^2}\,.$$

When do these two equations become roughly the same, and why?

3. Consider a Boolean model of the *lac* operon, based on five variables: mRNA (M), β-galactosidase (B), *lac* permease (P), allolactose (A), and intracellular lactose (L), and the following transition functions:

$$\begin{aligned} f_M &= A \\ f_B &= M \\ f_P &= M \\ f_A &= A \vee (L \wedge B) \\ f_L &= P \vee (L \wedge \overline{B}) \end{aligned}$$

(a) As we saw in class, the dynamics do not accurately reflect the behavior of the biological system it is meant to model. Therefore, something is wrong. Four of these functions are reasonable; justify each one in a single well-written sentence.

(b) Given the knowledge that β-galactosidase can also cleave allolactose into glucose and galactose, explain why one of these functions does not accurately reflect the underlying biology and/or the model assumptions. Propose a modification, aimed at eliminating the biologically infeasible fixed point, $(0, 0, 0, 1, 0)$. Give a rationale for your modification and specify the biological mechanism or model assumptions that justify the change.

(c) Use Cyclone to find the fixed point and plot the state space. Include a screenshot.

(d) Justify why the long-term behavior of the system (fixed points) agrees with what we should expect biologically.