# Solutions to Problem Set #1: Enzyme Kinetics

**1.** Let

$$B = \begin{pmatrix} b_{11} & b_{12} \\ b_{21} & b_{22} \end{pmatrix}, \; \vec{v} = \begin{pmatrix} v_a \\ v_b \end{pmatrix}.$$

(a) Show that if $B\vec{v} = \vec{0}$, and $\vec{v} \neq \vec{0}$, then $\det(B) = 0$.

(b) Show that if $\det(B) = 0$, then

  i. the rows of $B$ are linearly dependent (one row is a constant multiple of the other row) and
  ii. we can determine $\vec{v}$ only up to multiplicity (that is, $\vec{v}$ can be multiplied by any constant and still satisfy $B\vec{v} = \vec{0}$).

**Solution**

(a) $B\vec{v} = \vec{0} \Rightarrow b_{11}v_1 + b_{12}v_2 = 0$ and $b_{21}v_1 + b_{22}v_2 = 0$. Thus,

$$\begin{aligned} v_1 &= -\frac{b_{12}}{b_{11}}v_2 && (1) \\ \Rightarrow b_{21}\left(-\frac{b_{12}}{b_{11}}v_2\right) + b_{22}v_2 &= 0 && (2) \\ \Rightarrow (b_{11}b_{22} - b_{12}b_{21})v_2 &= 0 && (3) \end{aligned}$$

Since $v_1 \neq 0 \Rightarrow v_2 \neq 0$, then we must have $b_{11}b_{22} - b_{12}b_{21} = 0$ which is, by definition, $\det(B) = 0$.

(b) $\det(B) = 0 \Rightarrow b_{11}b_{22} - b_{12}b_{21} = 0$

  i. If the rows are linearly dependent, one row is a linear multiple of the other. Since $\det(B) = 0 \Rightarrow b_{11} = \frac{b_{12}b_{21}}{b_{22}}$ then

$$\begin{aligned} B &= \begin{pmatrix} b_{11} & b_{12} \\ b_{21} & b_{22} \end{pmatrix} && (4) \\ &= \begin{pmatrix} \frac{b_{12}b_{21}}{b_{22}} & b_{12} \\ b_{21} & b_{22} \end{pmatrix} && (5) \\ &= \begin{pmatrix} \frac{b_{12}}{b_{22}}b_{21} & \frac{b_{12}}{b_{22}}b_{22} \\ b_{21} & b_{22} \end{pmatrix} && (6) \end{aligned}$$

The first row is a linear multiple of the second row and the rows are linearly dependent.

ii. From above,

$$B\vec{v} = \begin{pmatrix} \frac{b_{12}}{b_{22}}b_{21} & \frac{b_{12}}{b_{22}}b_{22} \\ b_{21} & b_{22} \end{pmatrix}.$$

Thus,

$$\frac{b_{12}}{b_{22}}b_{21}v_1 + \frac{b_{12}}{b_{22}}b_{22}v_2 = 0 \,\text{and}\, b_{21}v_1 + b_{22}v_2 = 0.$$

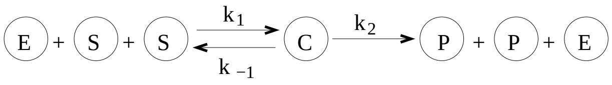
Assuming $b_{12} \neq 0, b_{22} \neq 0$, then we have one equation ($b_{21}v_1 + b_{22}v_2 = 0$) for two unknowns ($v_1$ and $v_2$). We must arbitrarily choose a value for, say, $v_1$, to determine $v_2$. Let $v_1 = a$, then $v_2 = -\frac{ab_{21}}{b_{22}}$ and

$$\vec{v} = a \begin{pmatrix} 1 \\ -\frac{b_{21}}{b_{22}} \end{pmatrix}.$$

Thus we can determine $\vec{v}$ only up to multiplicity.

**2.** Using a method similar to the one used in class, derive the degree-2 Hill function for a cooperative reaction where the enzyme must bind two substrate molecules to create the complex. Under what conditions is it valid? Assume the enzyme molecule binds the two substrate molecules simultaneously.

The reaction diagram is:



**Solution**
The ODEs are

$$\frac{dS}{dt} = -k_1ES^2 + k_{-1}C \tag{7}$$

$$\frac{dE}{dt} = -k_1ES^2 + k_{-1}C + k_2C \tag{8}$$

$$\frac{dC}{dt} = k_1ES^2 - k_{-1}C - k_2C \tag{9}$$

$$\frac{dP}{dt} = k_2C \tag{10}$$

Since $\frac{dE}{dt} + \frac{dC}{dt} = 0 \Rightarrow E + C = E_T$, where $E_T$ =constant.

Let $E = E_T - C$ and ignoring the equations for $P$, the system of equations is:

$$\begin{aligned} \frac{dS}{dt} &= -k_1(E_T - C)S^2 + k_{-1}C && (11) \\ \frac{dC}{dt} &= k_1(E_T - C)S^2 - k_{-1}C - k_2C && (12) \end{aligned}$$

Non-dimensionalize the equations by letting $S = \bar{S}S^*$, $E = \bar{E}E^*$, $t = \tau t^*$, we have

$$\begin{aligned} \frac{dS^*}{dt^*} &= -\tau k_1 \bar{S} E_T S^{*2} + \left(\tau k_1 \bar{C} \bar{S} S^{*2} + \frac{\tau k_{-1}\bar{C}}{\bar{S}}\right) C^* && (13) \\ \frac{dC}{dt} &= \frac{\tau k_1 \bar{S}^2 E_T}{\bar{C}} S^{*2} - (\tau k_1 \bar{S}^2 S^{*2} + \tau(k_{-1} + k_2))C^* && (14) \end{aligned}$$

Choose $\tau = \frac{1}{k_1 \bar{S} E_T}$, $\bar{S} = S_0$, $\bar{C} = E_T$ (as discussed in class), then (dropping the $*$s):

$$\begin{aligned} \frac{dS}{dt} &= -S^2 + (S^2 + K_n - K_{max})C && (15) \\ \epsilon\frac{dC}{dt} &= S^2 - (S^2 + K_n)C && (16) \end{aligned}$$

where $\epsilon = \frac{E_T}{S_0}$, $K_n = \frac{k_{-1}+k_2}{k_1 S_0^2}$, $K_{max} = \frac{k_2}{k_1 S_0^2}$. So if $\epsilon$ is very small (ie. $E_T \ll S_0$) then we can place the ODE for $C$ at quasi-steady state:

$$\epsilon\frac{dC}{dt} \approx 0.$$

Note: this is not the same as saying $\frac{dC}{dt} \approx 0$. For instance, suppose $E_T = 1\text{nM} = 0.001\mu\text{M}$ and $S_0 = 1\mu\text{M}$, then $\epsilon = 0.001$. Let's further suppose the kinetic rate constants are such that initially, $\frac{dC}{dt} \approx 10$. Then, $\epsilon\frac{dC}{dt} = 0.01 \ll 1$ and so $\epsilon\frac{dC}{dt} \approx 0$ but $\frac{dC}{dt} \not\approx 0$.

Under the condition $\epsilon \ll 1$ on the time scale $\tau = \frac{1}{k_1 S_0 E_T}$, product formation is governed by

$$\begin{aligned} \frac{dS}{dt} &= -S^2 + (S^2 + K_n - K_{max})C && (17) \\ 0 &= S^2 - (S^2 + K_n)C && (18) \\ \Rightarrow \frac{dS}{dt} &= -\frac{K_{max}S^2}{S^2 + K_n} && (19) \end{aligned}$$

This is shown graphically in Figure 1. Note the slope is initially 0 at the origin, in contrast to the the Michaelis-Menten curve.

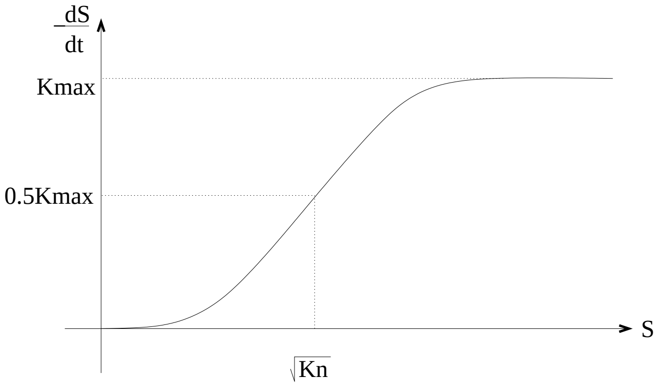


Figure 1: Plot of Hill function of degree 2.

**3.** Use Mathematica (or other software of your choice) to explore the equations for enzyme kinetics discussed in class (and reproduced below). Vary the parameters (including making them negative if you want) to make the steady state stable or unstable and the solution oscillatory or smooth. Plot your results as a function of time.

The equations are:

$$\frac{dS}{dt} = -S + (S + K_n - K_{max})C \tag{20}$$

$$\epsilon\frac{dC}{dt} = S - (S + K_n)C \tag{21}$$

**Solution**

There are five possible behaviours for the steady state at (0,0). For the following, I have set $\epsilon = 0.1$, and the corresponding trajectories are plotted in Figure 2.

(a) Stable node: $K_{max} = 1$, $K_n = 1$, eigenvalues: $\lambda_1 = -1$, $\lambda_2 = -10$.

(b) Unstable node: $K_{max} = 1$, $K_n = -1$, eigenvalues: $\lambda_1 = 7.7$, $\lambda_2 = 1.3$.

(c) Saddle node: $K_{max} = -1$, $K_n = 1$, eigenvalues: $\lambda_1 = 0.8$, $\lambda_2 = -11.8$.

(d) Stable spiral: $K_{max} = 10$, $K_n = 1$, eigenvalues: $\lambda_1 = -5.5 + 8.3i$, $\lambda_2 = -5.5 - 8.3i$.

(e) Unstable spiral: $K_{max} = 10$, $K_n = -$, eigenvalues: $\lambda_1 = 4.5 + 8.9i$, $\lambda_2 = 4.5 - 8.9i$.

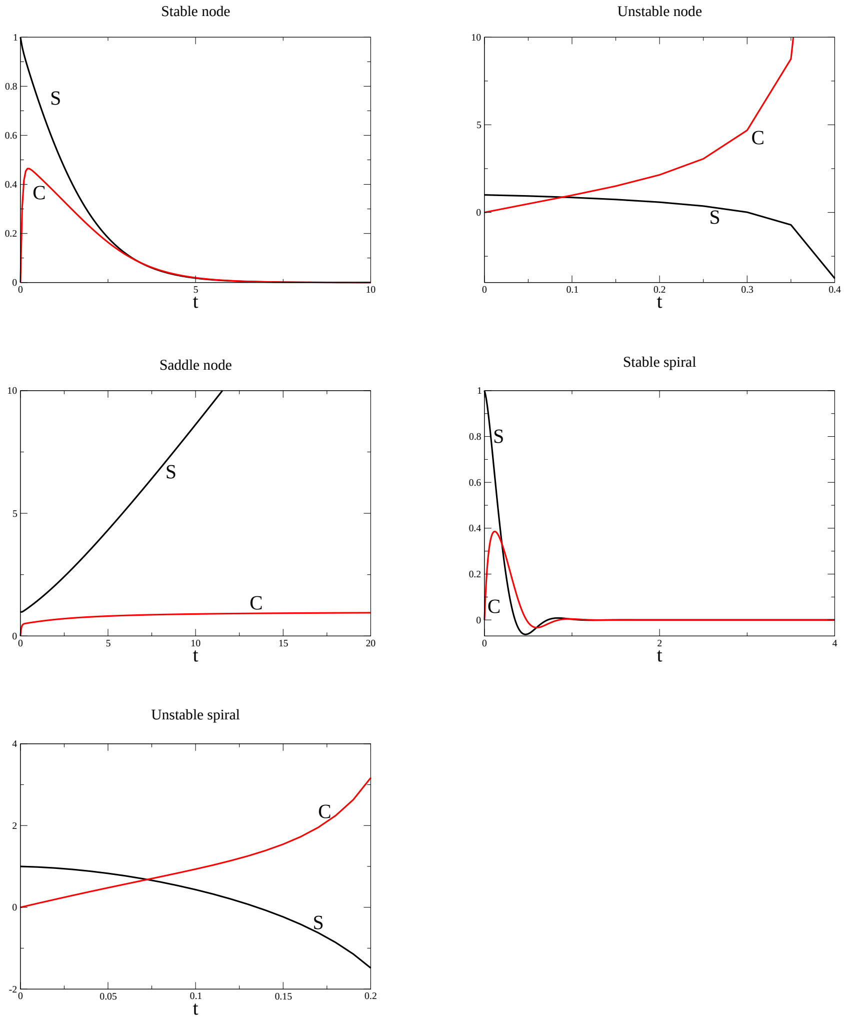


Figure 2: Trajectories for the five possible behaviours of the enzyme kinetics model.