BE 25 Winter 2025
Homework #3
Due at 9 AM PST, January 28, 2025

Problem 3.1 (The dynamics of the Brusselator, 35 pts)**.**
The Brusselator is a chemical reaction system that displays interesting dynamics in that the concentrations of intermediate species oscillate in time. It was proposed by Ilya Prigogine and his group in Brussels (J. Chem. Phys., 1968), hence the name. They were interested in oscillating chemical systems in general, but comment on how their theoretical approach is useful for understanding biochemical mechanisms important in biochemical reactions, citing in particular enzymatic transformation of glucose via the Chance-Higgins mechanism. The Brusselator was foundational for further work on oscillating biochemical systems.

In this problem, you will numerically solve for the dynamics of the Brusselator and plot the results to observe these beautiful oscillatory dynamics.

A reaction scheme for the Brusselator is as follows.

$$\mathrm{A} \xrightarrow{k_1} \mathrm{X} \tag{3.1}$$

$$2\,\mathrm{X} + \mathrm{Y} \xrightarrow{k_2} 3\,\mathrm{X} \tag{3.2}$$

$$\mathrm{B} + \mathrm{X} \xrightarrow{k_3} \mathrm{Y} + \mathrm{D} \tag{3.3}$$

$$\mathrm{X} \xrightarrow{k_4} \mathrm{E} \tag{3.4}$$

$$\mathrm{D} \xrightarrow{k_5} \emptyset \tag{3.5}$$

$$\mathrm{E} \xrightarrow{k_6} \emptyset\,. \tag{3.6}$$

Note that I have included the autodegradation of D and E; these are not usually included in the Brusselator dynamics, but would be present if D and E are not stable. The Brusselator operates with a feed of A and B such that their concentrations are held constant.

a) Define by a lower case letter the concentration of a given species. For example, x is the concentration of X. Assume a and b are constant. Write differential equations for $\mathrm{d}x/\mathrm{d}t$, $\mathrm{d}y/\mathrm{d}t$, $\mathrm{d}d/\mathrm{d}t$, and $\mathrm{d}e/\mathrm{d}t$. *Hint*: It might be a little confusing that X appears on both sides of the second reaction. When writing your ODE for the production of X, think about the *net* production of X due to that reaction.

b) Numerically solve for the dynamics with parameters $k_1 = k_2 = k_3 = k_4 = 1$, $k_5 = 0.4$, and $k_6 = 0.1$, with $a = 1$ and $b = 3$. (Assume the units of concentration and time are all consistent. We will not specifically deal with real time or concentration units, since this is mostly a thought experiment.) You should take as your initial condition $x(0) = y(0) = 0$. Integrate from time $t = 0$ to $t = 100$. Plot the result. Comment on anything striking you see.

c) *Not graded.* For fun, mess around with parameters and see how the oscillations change, or if you even get oscillations for all parameter values. It is also fun to plot the trajectory of x and y in the x-y plane, or d and e in the d-e plane.

d) Is it possible to have sustained oscillations without feed?

Problem 3.2 (Evaluating the QSSA in Michaelis-Menten mechanisms, 50 pts). In this problem, you will explore the regimes in which the quasi-steady state approximation (QSSA) is a good approximation for Michaelis-Menten reaction scheme,

$$\mathrm{E} + \mathrm{S} \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} \mathrm{ES} \xrightarrow{k_2} \mathrm{E} + \mathrm{P}. \tag{3.7}$$

As we have learned, the Michaelis-Menten equation,

$$\frac{\mathrm{d}c_\mathrm{P}}{\mathrm{d}t} \equiv v_0 = k_\mathrm{cat}\, c_\mathrm{E}^0\, \frac{c_\mathrm{S}/K_\mathrm{M}}{1 + c_\mathrm{S}/K_\mathrm{M}}, \tag{3.8}$$

can be derived from the mass action ODEs for the reaction scheme using the QSSA, which gives $k_\mathrm{cat} = k_2$ and $K_\mathrm{M} = (k_{-1} + k_2)/k_1$. Here, you will directly solve the Michaelis-Menten ODEs numerically without approximation and compare your result to the numerical results to assess the validity of the QSSA.

a) The ODEs corresponding to the Michaelis-Menten chemical reaction scheme can be written as

$$\frac{\mathrm{d}c_\mathrm{S}}{\mathrm{d}t} = -k_1(c_\mathrm{E}^0 - c_\mathrm{ES})c_\mathrm{S} + k_{-1}\, c_\mathrm{ES}, \tag{3.9}$$

$$\frac{\mathrm{d}c_\mathrm{ES}}{\mathrm{d}t} = k_1(c_\mathrm{E}^0 - c_\mathrm{ES})c_\mathrm{S} - (k_{-1} + k_2)c_\mathrm{ES}, \tag{3.10}$$

$$\frac{\mathrm{d}c_\mathrm{P}}{\mathrm{d}t} = k_2\, c_\mathrm{ES}, \tag{3.11}$$

where I have used conservation of enzyme to eliminate c_E from the dynamical equations. There are four parameters (k_1, k_{-1}, k_2, and c_E^0) in the above equations, which makes exploration of parameter space in our numerical calculations difficult. **Nondimensionalization** is a common procedure to reduce the number of parameters we need to consider. To nomdimensionalize a set of equations, we divide all variables (in this case, c_S, c_P, c_ES, and t) by a constant defined by some combination of the parameters. Show that by defining dimensionless variables

$$\tilde{t} = \frac{k_2\, c_\mathrm{E}^0}{K_\mathrm{M}}\, t \tag{3.12}$$

$$\tilde{c}_\mathrm{S} = c_\mathrm{S}/K_\mathrm{M}, \tag{3.13}$$

$$\tilde{c}_\mathrm{P} = c_\mathrm{P}/K_\mathrm{M}, \tag{3.14}$$

$$\tilde{c}_\mathrm{ES} = c_\mathrm{ES}/c_\mathrm{E}^0, \tag{3.15}$$

the dynamical equations may be written as

$$\kappa\, \frac{\mathrm{d}\tilde{c}_\mathrm{S}}{\mathrm{d}\tilde{t}} = -(1-\tilde{c}_\mathrm{ES})\tilde{c}_\mathrm{S} + (1-\kappa)\tilde{c}_\mathrm{ES}, \tag{3.16}$$

$$\kappa\,\zeta\, \frac{\mathrm{d}\tilde{c}_\mathrm{ES}}{\mathrm{d}\tilde{t}} = (1-\tilde{c}_\mathrm{ES})\tilde{c}_\mathrm{S} - \tilde{c}_\mathrm{ES}, \tag{3.17}$$

$$\frac{\mathrm{d}\tilde{c}_\mathrm{P}}{\mathrm{d}\tilde{t}} = \tilde{c}_\mathrm{ES}, \tag{3.18}$$

allowing us to reduce the number of parameters from four to two. Be sure to clearly write ζ and κ in terms of the respective parameters that comprise them. Henceforth for notational convenience, you can drop the tildes from the dimensionless variables, operating with the understanding that all variables are dimensionless.

b) Provide a physical interpretation of the two parameters ζ and κ.

c) Using a QSSA, write down the dimensionless Michaelis-Menten equation using the nondimensionalization scheme from part (a). In so doing, show that the dimensionless Michaelis-Menten equation is parameterless.

d) Write a code to numerically solve the dimensionless system of ODEs given by equations (3.16), (3.17), and (3.18). Use initial conditions $\tilde{c}_\mathrm{S} = 1$ and $\tilde{c}_\mathrm{P} = \tilde{c}_\mathrm{ES} = 0$. Plot the result. Do this for various values of the parameters κ and ζ. Overlay a plot of the solution to the appropriately nondimesionalized Michaelis-Menten equation (3.8). You can either use the analytical solution of the Michaelis-Menten equation or a numerical solution. If you choose to plot the analytical solution, the `scipy.special.lambertw()` function may be useful. Comment on what you see. Specifically, for what parameter regimes is the QSSA a good approximation?

Problem 3.3 (Firefly luciferase, 15 pts)**.**
This problem is based on Problem 4.5 of WTHS.

In many of the systems we have investigated, the dynamics of the concentrations of the respective species over time are of interest. Oftentimes, though, the *rate* of an enzyme catalyzed reaction is important. As an example, firefly luciferase is a commonly used reporter enzyme. It catalyzes oxidation of luciferin, emitting a photon in the process.

a) Why is it important to consider the *rate* of this reaction?

b) Under certain conditions, this enzymatic reaction can be described with Michaelis-Menten kinetics with Michaelis constant $K_\mathrm{M} = 100$ µM and

$$k_\mathrm{cat} = 0.04\,\frac{\text{mol oxyluciferin}}{\text{mol luciferase}\cdot\text{s}}. \tag{3.19}$$

(This is a rather slow luciferase, see, e.g., a “typical luciferase.”) In a typical assay, 100,000 cells are suspended in a 100 µL microplate well, with each cell having expressed about 100,000 molecules of luciferase. Luciferin is added suddenly such that its concentration in the well is 1 mM and the total well volume is still approximately 100 µL. What is the maximal rate of photon emission? Approximately how long does it take for the photon emission rate to be 90% of its maximal value? Given this result, do you need to worry about losing signal during the time scale of a typical biochemical experiment?