CS-E5885 Modeling biological networks
Ordinary differential equation models for biological networks

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January 25, 2022

Outline

- Classical continuous deterministic chemical kinetics
  - Mass-action kinetics
- Euler and Runge-Kutta methods for numerical simulation
- Equilibrium analysis
- Reversibility and conservation in deterministic model
- Enzyme kinetics: Michaelis-Menten kinetics
- Regulation of enzyme activity: competitive and allosteric
- Cooperativity: Hill kinetics
- Reading (see references at the end):
  - This lecture follows closely Section 6 from (Wilkinson, 2011) and Section 3 from (Ingalls, 2013)

Deterministic mass-action kinetics

- Motivation:
  - The expectation of stochastic kinetic model (coupled chemical reactions) corresponds to deterministic differential equation system
  - Ignoring the diffusion term in the chemical Langevian equation, we retrieve the same/similar deterministic differential equation model

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  - The expectation of stochastic kinetic model (coupled chemical reactions) corresponds to deterministic differential equation system
  - Ignoring the diffusion term in the chemical Langevian equation, we retrieve the same/similar deterministic differential equation model
- Lets now see how deterministic dynamics models can be formulated for chemical systems in the first place
- Consider e.g. the Lotka-Volterra model

Lotka–Volterra (reaction equations)

$$
\begin{aligned}
Y_1 &\to 2Y_1 \\
Y_1 + Y_2 &\to 2Y_2 \\
Y_2 &\to \emptyset
\end{aligned}
$$

Deterministic mass-action kinetics (2)

- Generally, the rate of a reaction is directly proportional to the concentration (denoted as $[\cdot]$) of reactants raised to the power of its stoichiometry
  - Recall stochastic rate laws for biochemical kinetics
- This kinetic law is known as mass-action kinetics
- Deterministic models are defined in terms of (deterministic) rate constants k_i

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Lotka–Volterra (reaction equations)

$$
\begin{aligned}
Y_1 &\to 2Y_1 && \text{(with rate } c_1\text{)} \\
Y_1 + Y_2 &\to 2Y_2 && \text{(with rate } c_2\text{)} \\
Y_2 &\to \emptyset && \text{(with rate } c_3\text{)}
\end{aligned}
$$

Lotka–Volterra (system of ODEs)

$$
\begin{cases}
\frac{d[Y_1]}{dt} = k_1[Y_1] - k_2[Y_1][Y_2] \\
\frac{d[Y_2]}{dt} = k_2[Y_1][Y_2] - k_3[Y_2]
\end{cases}
$$

Deterministic mass-action kinetics (3)

- The Lotka-Volterra ODE system in matrix form:

$$\frac{d}{dt}\begin{pmatrix}[Y_1]\\ [Y_2]\end{pmatrix} = \begin{pmatrix}1 & -1 & 0\\ 0 & 1 & -1\end{pmatrix}\begin{pmatrix}k_1[Y_1]\\ k_2[Y_1][Y_2]\\ k_3[Y_2]\end{pmatrix}$$

where the 2×3 matrix is the stoichiometric matrix S

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- In the following ODE models are written in a general form e.g. as

$$\frac{d}{dt}Y(t) = f(Y(t)|\theta),$$

  where in this case $Y(t) = ([Y_1], [Y_2])^T$, $f(\cdot)$ is defined by the right hand size, and $\theta = [k_1, k_2, k_3]^T$

Initial value problem for ODEs

- An initial value problem consists of
  - An ODE model $\frac{d}{dt}X(t) = f(X(t)|\theta)$, and
  - An initial value $X(t_0) = X_0$
- A solution to an initial value problem is a function $X(t)$ defined for all $t \in \mathbb{R}$ that
  - Is a solution of the ODE model
  - That also satisfies the initial value/condition $X(t_0) = X_0$

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  - Is a solution of the ODE model
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- In general, the solution to the initial value problem is (with some regularity conditions of f)

$$X(t) = X(t_0) + \int_{t_0}^{t} f(X(t)|\theta)dt$$

- For most ODE systems considered in this course, the solution to the initial value problem cannot be written in closed-form
- The solution to the initial value problem can be found using numerical techniques

Euler method for ODEs

- Euler method is the simplest possible numerical simulation method for ODEs
- State vector $X = (X_1, \ldots, X_n)^T \in \mathbb{R}^n$ and an arbitrary function $f : \mathbb{R}^n \to \mathbb{R}^n$ of $X(t)$ with parameters θ

$$\begin{aligned} \frac{dX(t)}{dt} &= f(X(t)|\theta) \\ \lim_{\Delta t \to 0} \frac{X(t+\Delta t) - X(t)}{\Delta t} &= f(X(t)|\theta) \end{aligned}$$

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- For small values of Δt this can be approximated with the finite diffidence as

$$\frac{X(t+\Delta t) - X(t)}{\Delta t} \approx f(X(t)|\theta)$$

and by solving for $X(t+\Delta t)$ one gets

$$X(t+\Delta t) \approx X(t) + \Delta t \cdot f(X(t)|\theta)$$

Euler method for ODEs (2)

- Given an initial value $X(t_0)$, the above equation can be applied recursively to compute $X(t_0)$, $X(t_0 + \Delta t)$, $X(t_0 + 2\Delta t)$, $X(t_0 + 3\Delta t)$, ...

$$\begin{aligned} X(t_0 + \Delta t) &= X(t_0) + \Delta t \cdot f(X(t_0)|\theta) \\ X(t_0 + 2\Delta t) &= X(t_0 + \Delta t) + \Delta t \cdot f(X(t_0 + \Delta t)|\theta) \\ X(t_0 + 3\Delta t) &= X(t_0 + 2\Delta t) + \Delta t \cdot f(X(t_0 + 2\Delta t)|\theta) \\ &\vdots \end{aligned}$$

  which can be used to approximate the exact solution

$$X(t) = X(t_0) + \int_{t_0}^{t} f(X(t)|\theta)dt$$

  at any time point $t \geq t_0$

Mass-action kinetics (2)

Lotka–Volterra (system of ODEs))

$$\begin{cases} \frac{d[Y_1]}{dt} = k_1[Y_1] - k_2[Y_1][Y_2] \\ \frac{d[Y_2]}{dt} = k_2[Y_1][Y_2] - k_3[Y_2] \end{cases}$$

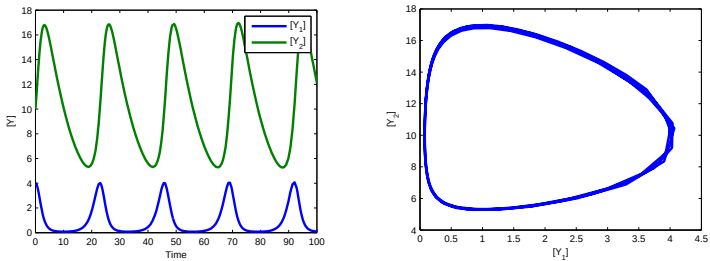

Figure: Lotka–Volterra dynamics in time-space and phase-space for initial values $[Y_1](0) = 4, [Y_2](0) = 10$ and kinetic rates $k_1 = 1, k_2 = 0.1, k_3 = 0.1$

Runge-Kutta methods

- A number of advanced methods exist for approximate solutions of ODEs
  - Linear multistep methods, Runge-Kutta methods, adaptive
  - Explicit vs. implicit methods
- Explicit Runge-Kutta methods are commonly used
- For example, in the classical fourth-order Runge-Kutta method one step of the numerical integration is defined as

$$X(t + \Delta t) = X(t) + \frac{1}{6}(k_1 + 2k_2 + 2k_3 + k_4),$$

where

$$\begin{aligned} k_1 &= \Delta t \cdot f(X(t)) \\ k_2 &= \Delta t \cdot f(X(t) + k_1/2) \\ k_3 &= \Delta t \cdot f(X(t) + k_2/2) \\ k_4 &= \Delta t \cdot f(X(t) + k_3) \end{aligned}$$

Runge-Kutta methods

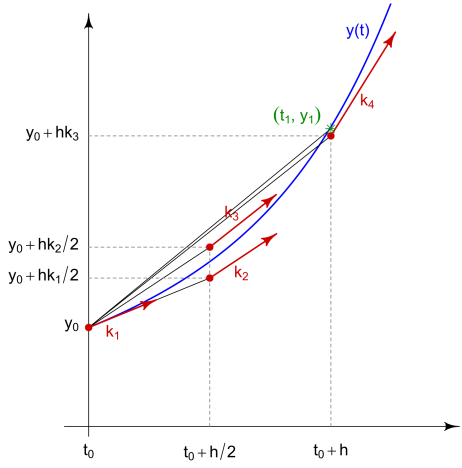

Figure: Illustration of the fourth-order Runge-Kutta method (Wikipedia)

Equilibrium

- An equilibrium solution is a set of concentrations that will not change over time
- Equilibrium can be found analytically or numerically by
  - Setting the right hand side to zero, or
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- Equilibrium can be found analytically or numerically by
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  - Via simulations
- For the LV system one has

$$\begin{aligned} k_1[Y_1] - k_2[Y_1][Y_2] &= 0 \\ k_2[Y_1][Y_2] - k_3[Y_2] &= 0 \end{aligned}$$

- In addition to the trivial solution ($[Y_1] = [Y_2] = 0$) one finds

$$[Y_1] = \frac{k_3}{k_2} \quad \text{and} \quad [Y_2] = \frac{k_1}{k_2}$$

Reversibility

- A dimerization reaction: $2P \longleftrightarrow P_2$ (or $2P \to P_2$ and $P_2 \to 2P$)

$$\begin{cases} \frac{d[P]}{dt} = 2k_2[P_2] - 2k_1[P]^2 \\ \frac{d[P_2]}{dt} = k_1[P]^2 - k_2[P_2] \end{cases} \qquad (*)$$

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- Equilibrium whenever

$$k_2[P_2] = k_1[P]^2 \Leftrightarrow \frac{[P_2]}{[P]^2} = \frac{k_1}{k_2} \equiv K_{\text{eq}},$$

where K_{eq} is the equilibrium constant of the system

Conservation

- Add twice the second equation of $(*)$ to the first equation

$$\frac{d[P]}{dt} + 2\frac{d[P_2]}{dt} = 0$$

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$$\begin{aligned} \frac{d[P]}{dt} + 2\frac{d[P_2]}{dt} &= 0 \\ \Rightarrow \frac{d}{dt}([P] + 2[P_2]) &= 0 \\ \Rightarrow [P] + 2[P_2] &= c \end{aligned}$$

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- This is a conservation equation
- Alternatively, the reaction matrix A can be obtained from the coupled chemical reactions model of $(*)$

$$A = \begin{pmatrix} -2 & 1 \\ 2 & -1 \end{pmatrix}$$

- A vector $y = (1, 2)'$ is seen to be a P-invariant, i.e., to satisfy $Ay = \mathbf{0}$

Conservation (2)

- Conservation ($2[P_2] = c - [P]$) can be used to reduce the dimension of the system

$$\frac{d[P]}{dt} = k_2(c - [P]) - 2k_1[P]^2$$

- Equilibrium: after substituting and rearranging we get

$$2K_{\text{eq}}[P]^2 + [P] - c = 0,$$

where a positive real root is $[P] = \frac{\sqrt{8cK_{\text{eq}}+1}-1}{4K_{\text{eq}}}$

Conservation (3)

- Reducing the dimension of an deterministic system helps in
  - Reducing the system will improve speed, accuracy and numerical stability of numerical ODE solvers
  - Mathematical analysis often requires a system of full-rank
- Conservation laws can be used to reduce the dimension of stochastic models as above, but is less important
  - Speed improvement is typically not significant/important
  - Exact algorithms, such as Gillespie, are exact

Introduction to biochemical reactions

- Individual chemical reaction events (binding, unbinding, and conversion) are called elementary reactions
- We used mass action to describe the rates of elementary reactions
- Contrary to previous models, individual biochemical reactions involve small networks of elementary reactions
- To develop rate laws for biochemical reactions, these networks are collapsed into single reaction events
- The rate laws that describe these 'lumped' reaction events are often referred to as biochemical kinetics

Enzyme kinetics

- Majority of reactions that occur within a cell are catalysed by enzymes
- Enzymes catalyse reactions by binding the reactants (called the enzyme substrates) and facilitating their conversion to the reaction products

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- Enzymes catalyse reactions by binding the reactants (called the enzyme substrates) and facilitating their conversion to the reaction products
- Enzyme catalysis reduces the energy barrier associated with the reaction event

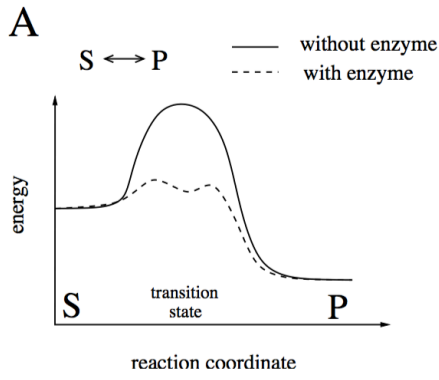

Figure: Figure 3.1A from (Ingalls, 2013)

Enzyme kinetics

- On the other hand, enzyme catalysis has no effect on the equilibrium itself
- Enzyme catalyzed reaction is simply faster

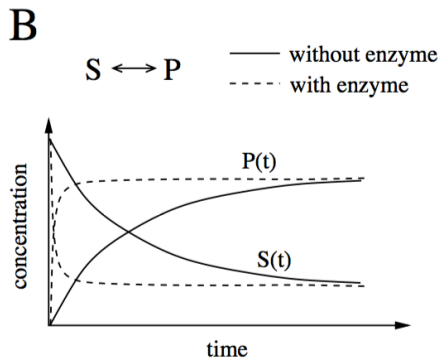

Figure: Figure 3.1B from (Ingalls, 2013)

Enzyme kinetics

- The standard 'lock-and-key' model of enzyme activity is illustrated below

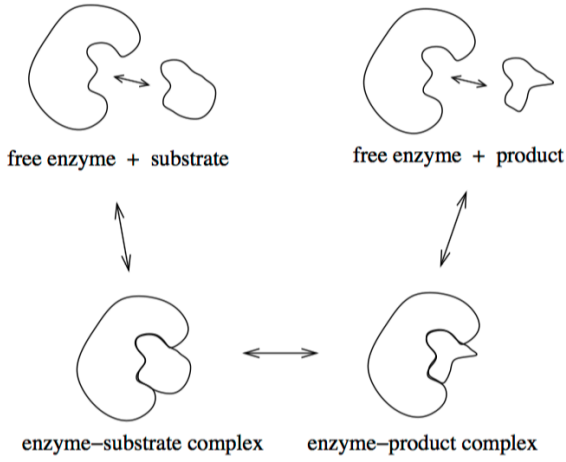

Figure: Figure 3.2A from (Ingalls, 2013)

Enzyme kinetics

- For reversible reactions, enzyme can catalyse the reaction in both directions
- Enzyme is unaltered by the reaction event
- Enzymes typically bind only a single substrate species
- Most enzymes catalyse only a specific reaction
- This allows each enzyme to function with remarkable efficiency and specificity

Enzyme kinetics

- Experimental observations of enzyme-catalysed reactions show that they do not obey mass action rate laws
- Saturating behaviour is caused by the limited amount of enzyme present

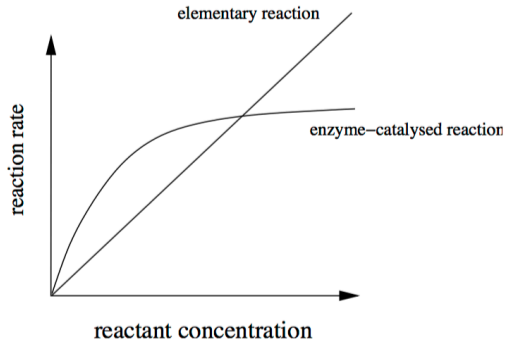

Figure: Figure 3.2B from (Ingalls, 2013)

Michaelis-Menten kinetics

- We will derive the rate law that describes enzyme-catalyzed reactions
  - Michaelis-Menten kinetics
  - Note that this is a rate law for a collection of reactions (i.e., a small network of elementary reactions)
- The chemical reactions involved in a single substrate enzyme-catalyzed reaction

$$\underbrace{S}_{\text{substrate}} + \underbrace{E}_{\text{enzyme}} \rightleftharpoons \underbrace{C_1}_{\text{enzyme-substrate complex}} \rightleftharpoons \underbrace{C_2}_{\text{enzyme-product complex}} \rightleftharpoons \underbrace{P}_{\text{product}} + \underbrace{E}_{\text{enzyme}}$$

Michaelis-Menten kinetics

- The derivation starts with two assumptions
  - Complexes C_1 and C_2 are combined by assuming that time scales of reactions $C_1 \rightleftharpoons C_2$ are fast compared to other reactions
    - So-called rapid equilibrium assumption
  - We assume that the product P never binds free enzyme

$$S + E \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} C \xrightarrow{k_2} P + E$$

- With these two assumptions the mode with a set of 6 reactions is down to 3 reactions

Michaelis-Menten kinetics

- Applying mass-action kinetics to the simplified enzyme-catalyzed reaction network from the previous page

$$S + E \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} C \xrightarrow{k_2} P + E$$

gives the following ODE system

$$\begin{aligned}
\frac{d}{dt}s(t) &= -k_1 s(t)e(t) + k_{-1}c(t) \\
\frac{d}{dt}e(t) &= -k_1 s(t)e(t) + k_{-1}c(t) + k_2 c(t) \\
\frac{d}{dt}c(t) &= -k_{-1}c(t) + k_1 s(t)e(t) - k_2 c(t) \\
\frac{d}{dt}p(t) &= k_2 c(t)
\end{aligned}$$

Michaelis-Menten kinetics

- Because the enzyme E is not consumed its total concentration e_T remains constant
- Let us denote the free enzyme concentration as

$$e(t) = e_T - c(t)$$

and use it to reduce the dimension of the system

$$\begin{aligned}
\frac{d}{dt}s(t) &= -k_1 s(t)(e_T - c(t)) + k_{-1}c(t) \\
\frac{d}{dt}c(t) &= -k_{-1}c(t) + k_1 s(t)(e_T - c(t)) - k_2 c(t) \\
\frac{d}{dt}p(t) &= k_2 c(t)
\end{aligned}$$

Michaelis-Menten kinetics

- Simulation of the reduced system is shown on right
- The figure reveals a separation of time-scales
  - Fast time-scale: $S + E \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} C$
  - Slow time-scale: $C \xrightarrow{k_2} P + E$

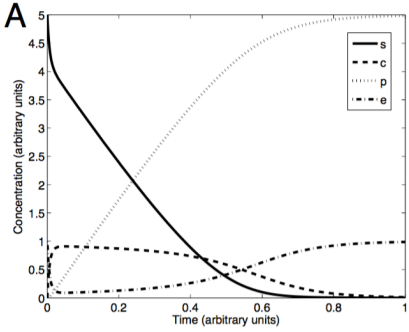

Figure: Figure 3.3A from (Ingalls, 2013)

Michaelis-Menten kinetics

- The different time-scale are due to
  - Difference in reaction time constants
  - Differences in concentrations: e.g. for intra-cellular metabolic reactions substrate tends to be more abundant than the enzyme and thus the enzyme-complex comes to quasi-steady state with respect to more abundant substrate

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  - Differences in concentrations: e.g. for intra-cellular metabolic reactions substrate tends to be more abundant than the enzyme and thus the enzyme-complex comes to quasi-steady state with respect to more abundant substrate
- Quasi-steady state means/implies that

$$\frac{d}{dt}c(t) = -k_{-1}c^{\text{qss}}(t) + k_1 s(t)(e_T - c^{\text{qss}}(t)) - k_2 c^{\text{qss}}(t) = 0$$

which gives us

$$c^{\text{qss}}(t) = \frac{k_1 e_T s(t)}{k_{-1} + k_2 + k_1 s(t)}$$

Michaelis-Menten kinetics

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which gives us

$$c^{\text{qss}}(t) = \frac{k_1 e_T s(t)}{k_{-1} + k_2 + k_1 s(t)}$$

and further

$$\begin{aligned} \frac{d}{dt}s(t) &= -\frac{k_1 k_2 e_T s(t)}{k_{-1} + k_2 + k_1 s(t)} \\ \frac{d}{dt}p(t) &= \frac{k_1 k_2 e_T s(t)}{k_{-1} + k_2 + k_1 s(t)} \end{aligned}$$

Michaelis-Menten kinetics

- The last two equations on the previous slide describe $S \rightarrow P$ as a single non-elementary reaction
  - Michaelis-Menten rate law

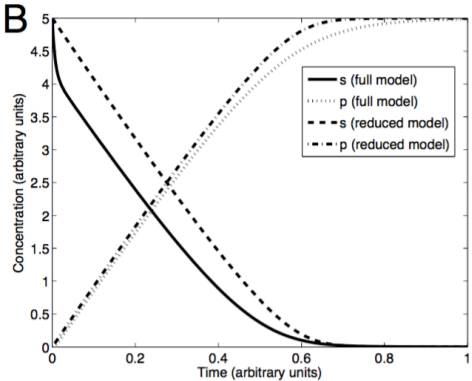

Figure: Figure 3.3B from (Ingalls, 2013)

Michaelis-Menten kinetics

- A commonly used form of the Michaelis-Menten rate law is the following

$$\text{rate of } S \to P = k_2 c^{\text{qss}} = \frac{V_{\max} s}{K_M + s},$$

where

  - $V_{\max} = k_2 e_T$ is the maximal (limiting) rate and
  - $K_M = \frac{k_{-1}+k_2}{k_1}$ is the Michaelis (half-saturating) constant

- Maximal rate $V_{\max}$ is obtained by letting $s \to \infty$
- Half-saturating constant: rate equals $\frac{1}{2} V_{\max}$ when $s = K_M$

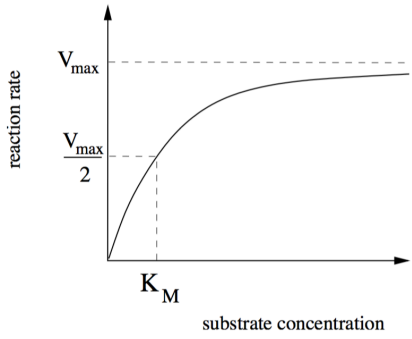

Figure: Figure 3.4 from (Ingalls, 2013)

Regulation of enzyme activity

- Enzyme activity can be regulated by several mechanisms
- Transcriptional regulation can affect the total enzyme concentration e_T
  - A slow-scale process
- Biochemical modification of enzyme molecules
  - A process in a faster time-scale
  - Competitive inhibition
  - Allosteric regulation

Competitive inhibition

- An inhibitor is a molecule that mimic's the substrate but does not lead to reaction of producing a product
- An example: the drug aspirin binds the active site of the enzyme cyclooxygenase and thus inhibits the production of prostoglandins

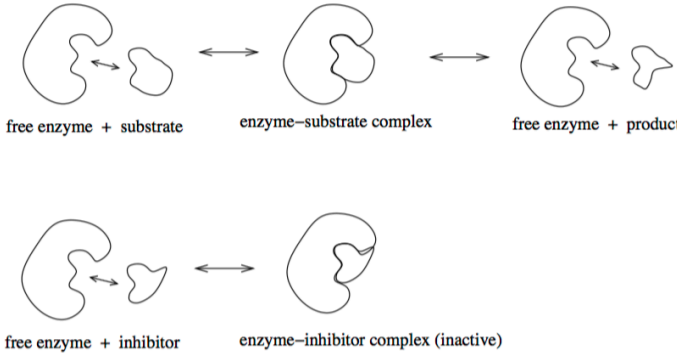

Figure: Figure 3.5 from (Ingalls, 2013)

Competitive inhibition

- Consider the following competitive inhibition system that includes both the enzyme-catalysed and inhibition reactions

$$S + E \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} C \xrightarrow{k_2} P + E$$

$$I + E \underset{k_{-3}}{\overset{k_3}{\rightleftharpoons}} C_I$$

where I and C_I denote the inhibitor and inhibitor-enzyme complex

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- Mass-action kinetics for C and C_I

$$\begin{aligned} \frac{d}{dt}c(t) &= -k_{-1}c(t) + k_1 s(t)e(t) - k_2 c(t) \\ \frac{d}{dt}c_I(t) &= k_3 e(t)i(t) - k_{-3}c_I(t) \end{aligned}$$

Competitive inhibition

- We can assume that inhibitor I is far more abundant than the enzyme E
  - Thus, $i(t) = i$
- Similarly as before, we make an assumption that complexes C and C_I are in quasi-steady state, i.e., $\frac{d}{dt}c(t) = 0$ and $\frac{d}{dt}c_I(t) = 0$

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- Using a conservation $e(t) = e_T - c(t) - c_I(t)$ gives (after some manipulation)

$$c^{\text{qss}} = \frac{e_T s}{\frac{iK_M}{K_i} + s + K_M}$$

where $K_M = \frac{k_{-1}+k_2}{k_1}$ and $K_i = \frac{k_{-3}}{k_3}$

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  - Thus, $i(t) = i$
- Similarly as before, we make an assumption that complexes C and C_I are in quasi-steady state, i.e., $\frac{d}{dt}c(t) = 0$ and $\frac{d}{dt}c_I(t) = 0$
- Using a conservation $e(t) = e_T - c(t) - c_I(t)$ gives (after some manipulation)

$$c^{\text{qss}} = \frac{e_T s}{\frac{iK_M}{K_i} + s + K_M}$$

  where $K_M = \frac{k_{-1}+k_2}{k_1}$ and $K_i = \frac{k_{-3}}{k_3}$
- Finally, the rate law can be written as

$$\text{rate of } S \to P = k_2 c^{\text{qss}} = \frac{V_{\max} s}{K_M(1 + i/K_i) + s},$$

- Competitive inhibition changes half-saturating constant to $K_M(1 + i/K_i)$ but not change the limiting rate $V_{\max}$

Competitive inhibition

- An illustration of the rate law for competitive inhibition model
- Competitive inhibition changes half-saturating constant to $K_M(1 + i/K_i)$ but not change the limiting rate $V_{\max}$

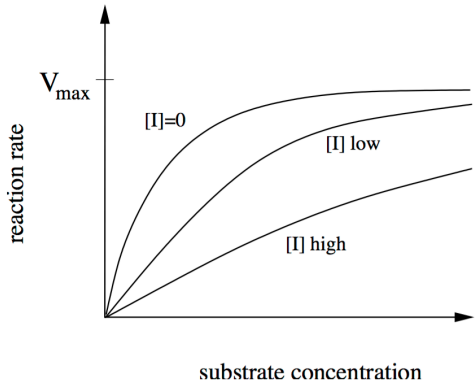

Figure: Figure 3.6 from (Ingalls, 2013)

Allosteric regulation

- Enzyme efficiency depends on the conformation of the active site
  - Tertiary structure of the enzyme
- Modifications to enzyme conformation can be made by proteins that bind the enzyme

Allosteric regulation

- Enzyme efficiency depends on the conformation of the active site
  - Tertiary structure of the enzyme
- Modifications to enzyme conformation can be made by proteins that bind the enzyme
- This is known as allosteric enzyme regulation
- The regulator can bind the enzyme at sites which are distinct from the active site
  - Consequently, the regulator does not need to resemble the substrate

Allosteric regulation

- Binding of the allosteric regulator invokes a transition between active and inactive enzyme states
- An illustration for the case where allosteric regulator inhibits enzyme catalysis, but does not affect the substrate binding

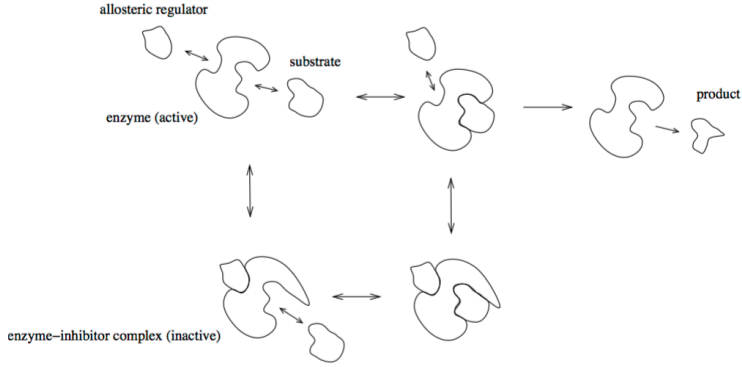

Figure: Figure 3.7 from (Ingalls, 2013)

Allosteric regulation

- The reaction scheme for the allosteric regulation model from the previous slide (assume independence for binding of E and I)

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

$$I + E \underset{k_{-3}}{\overset{k_3}{\rightleftharpoons}} EI$$

$$I + ES \underset{k_{-3}}{\overset{k_3}{\rightleftharpoons}} ESI$$

$$S + EI \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} ESI$$

- This model implements a non-competitive inhibition

Allosteric regulation

- For this allosteric regulation model a conservation is

$$e_T = [E] + [ES] + [EI] + [ESI]$$

Allosteric regulation

- For this allosteric regulation model a conservation is

$$e_T = [E] + [ES] + [EI] + [ESI]$$

- By applying the quasi-steady state assumption to all the complexes one gets

$$\text{rate of } S \to P = k_2[ES] = \frac{V_{\max}}{1 + i/K_i} \frac{s}{K_M + s}$$

  where constants $V_{\max}$, K_M and K_i are defined as previously
- Allosteric regulation changes the limiting rate but does not change the half-saturating constant

Allosteric regulation

- An illustration of allosteric regulation model

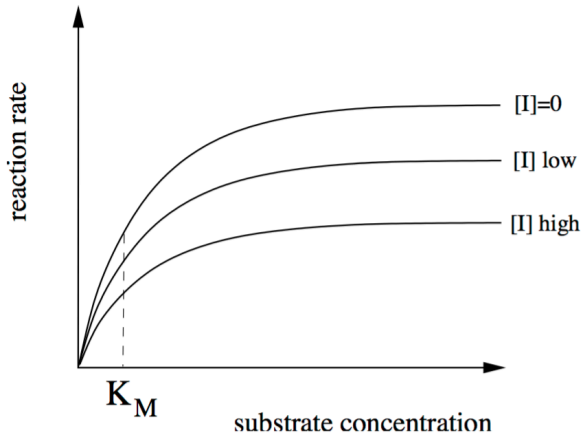

Figure: Figure 3.8 from (Ingalls, 2013)

Cooperativity

- Cooperativity denotes potentially independent binding events that have a significant influence on one another
  - Results in nonlinear behavious
- An example: Oxygen binding to hemoglobin protein
  - Hemoglobin is a tetrameric protein with each monomer binding one oxygen molecule.
- Hemoglobin's efficiency as an oxygen carrier can be quantified by the fraction of protein in the oxygen-bound form as a function of the abundance of oxygen
  - These curves were found to be sigmoidal instead of hyperbolic
  - Compare with the similar curve for a monomeric myoglobin

Cooperativity

- Sigmoidal binding curve for hemoglobin

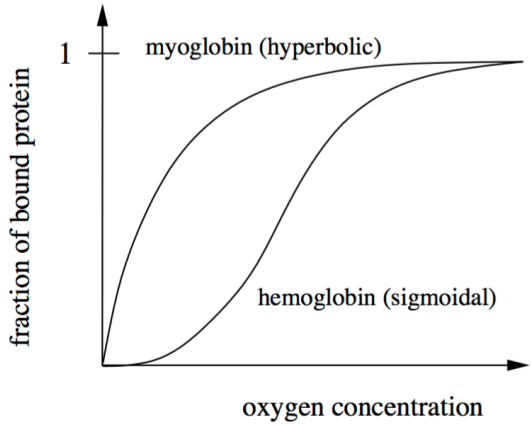

Figure: Figure 3.9 from (Ingalls, 2013)

Cooperativity

- Consider a molecule X (generally called a ligand) binding a protein P

$$P + X \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} PX$$

- The fractional saturation of a protein pool is defined as the fraction of binding sites that are occupied by ligand

$$Y = \frac{\text{number of occupied binding sites}}{\text{total number of binding sites}} = \frac{[PX]}{[P] + [PX]}$$

Cooperativity

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$$Y = \frac{\text{number of occupied binding sites}}{\text{total number of binding sites}} = \frac{[PX]}{[P] + [PX]}$$

- In steady-state $[PX] = [P][X]/K$, where $K = \frac{k_{-1}}{k_1}$ and thus

$$Y = \frac{[P][X]/K}{[P] + [P][X]/K} = \frac{[X]/K}{1 + [X]/K} = \frac{[X]}{K + [X]}$$

- This is hyperbolic

Cooperativity

- Consider a protein that has four ligand binding sites (with non-independent, i.e. cooperative, binding events)

$$
\begin{aligned}
P + X &\underset{k_{-1}}{\overset{4k_1}{\rightleftharpoons}} PX_1 \\
PX_1 + X &\underset{2k_{-2}}{\overset{3k_3}{\rightleftharpoons}} PX_2 \\
PX_2 + X &\underset{3k_{-3}}{\overset{2k_3}{\rightleftharpoons}} PX_3 \\
PX_3 + X &\underset{4k_{-4}}{\overset{k_4}{\rightleftharpoons}} PX_4,
\end{aligned}
$$

where complex PX_i has i ligands bound and rate constants depend on the number of bound ligands (correspond to stoichiometric coefficients)

Cooperativity

- The fractional saturation is given by

$$Y = \frac{[PX_1] + 2[PX_2] + 3[PX_3] + 4[PX_4]}{4([P] + [PX_1] + [PX_2] + [PX_3] + [PX_4])}$$

Cooperativity

- The fractional saturation is given by

$$Y = \frac{[PX_1] + 2[PX_2] + 3[PX_3] + 4[PX_4]}{4([P] + [PX_1] + [PX_2] + [PX_3] + [PX_4])}$$

- When the binding events are in equilibrium, then

$$Y = \frac{[X]/K_1 + 3[X]^2/(K_1K_2) + 3[X]^3/(K_1K_2K_3) + [X]^4/(K_1K_2K_3K_4)}{1 + 4[X]/K_1 + 6[X]^2/(K_1K_2) + 4[X]^3/(K_1K_2K_3) + [X]^4/(K_1K_2K_3K_4)},$$

  where $K_i = \frac{k_{-i}}{k_i}$
- This so-called Adair equation has a sigmoidal character when the later binding events have higher affinity than the earlier ones
  - Positive cooperativity: binding of the first ligand enhances the binding of later ligands

Cooperativity

- If $K_4 \ll K_1, K_2, K_3$ then the Adair equation becomes

$$Y \approx \frac{[X]^4/(K_1K_2K_3K_4)}{1+[X]^4/(K_1K_2K_3K_4)},$$

- In general form this is known as the Hill function

$$Y = \frac{([X]/K)^n}{1+([X]/K)^n} = \frac{[X]^n}{K^n+[X]^n},$$

where K is the half-saturating concentration of the ligand

Cooperativity

- Illustration of Hill functions
  - When n increases, then the sigmoidal becomes more switch-like mechanism
  - When $n = 1$, then the curve is hyperbolic

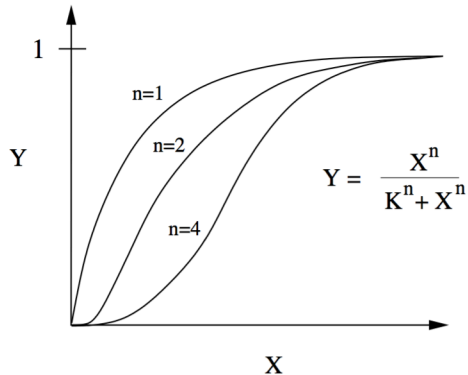

Figure: Figure 3.10 from (Ingalls, 2013)

References

- Ingalls BP, Mathematical Modeling in Systems Biology: An Introduction, MIT Press, 2013
- Darren J. Wilkinson, *Stochastic Modelling for Systems Biology*, Chapman & Hall/CRC, 2011