Class 11.1
Chemical Kinetics

CHEM 102H
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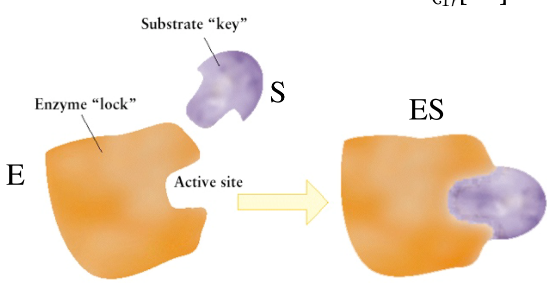
Biochemical Catalysts; Enzyme Kinetics

Substrate binds to enzyme to facilitate reaction

$E + S \rightleftharpoons ES$ forward rate = $k_{1f}[E][S]$

reverse rate = $k_{1r}[ES]$



Enzyme Kinetics

Michaelis-Menten mechanism:

$E + S \underset{k_{1r}}{\overset{k_{1f}}{\rightleftharpoons}} ES$ forward rate = $k_{1f}[E][S]$

reverse rate = $k_{1r}[ES]$

$ES \xrightarrow{k_2}$ products + E rate = $k_2[ES]$

In most cases, the substrate conc. is much greater than the enzyme. Steady-state approx. is be applied to rate of [ES] change:

$$0 = -k_{1f}[E][S] + k_{1r}[ES] + k_2[ES]$$

Enzyme Kinetics

$E + S \rightleftharpoons ES$ forward rate = $k_{1f}[E][S]$

reverse rate = $k_{1r}[ES]$

$ES \rightarrow$ products + E rate = $k_2[ES]$

To get an expression with only $[E]_0$ and [S], we can plug into the steady-state equation

([ES] = rate/ k_2 and

$[E] = [E]_0 - [ES] = [E]_0 - \text{rate}/k_2$)

$0 = -k_{1f}([E]_0 - \text{rate}/k_2)[S] + k_{1r}(\text{rate}/k_2) + \text{rate}$

$k_{1f}[E]_0[S] = \text{rate}\{(k_{1f}/k_2)[S] + (k_{1r}/k_2) + 1\}$

Enzyme Kinetics

$E + S \rightleftharpoons ES$ forward rate = $k_{1f}[E][S]$

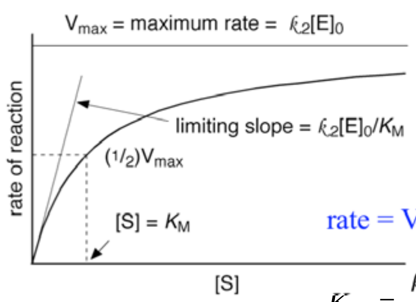
reverse rate = $k_{1r}[ES]$

$ES \rightarrow$ products + E rate = $k_2[ES]$

Finally, solve the last expression for the rate

$$\text{rate} = \frac{k_2[E]_0[S]}{K_M + [S]}; \quad K_M = \frac{k_{1r} + k_2}{k_{1f}}$$

Michaelis-Menten Kinetics

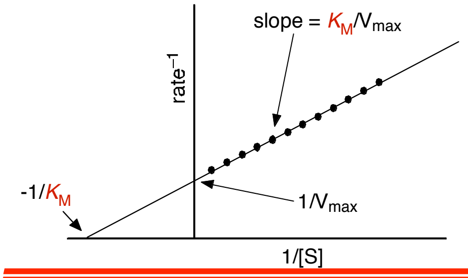


$$\text{rate} = V_0 = \frac{V_{max}[S]}{K_M + [S]};$$

$$K_M = \frac{k_{1r} + k_2}{k_{1f}}$$

Double-Reciprocal Plot

$$\frac{1}{\text{rate}} = \frac{1}{V_0} = \frac{K_M + [S]}{V_{max}[S]} = \frac{1}{V_{max}} + \frac{K_M}{V_{max}[S]}$$



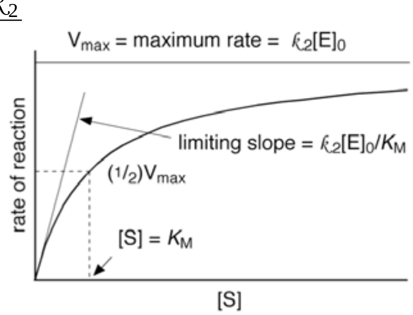
The hydrolysis of pyrophosphate to orthophosphate is an important biosynthetic process that many organisms use to as an energy source (it the same kind of reaction as occurs in the conversion of ATP into ADP). The enzyme that catalyzes this reaction is called pyrophosphatase and it obeys Michaelis-Menten kinetics. The basic enzyme unit has a molar mass of 20,000 g/mol. When purified, at its maximum reaction rate (V_{max}) such that 1.0 mg of enzyme can convert 0.112 mol/hour of pyrophosphate (which is the substrate, S) into product at 37 ˚C (body temperature).

a) How many moles of substrate are hydrolyzed per second per milligram of enzyme when the substrate concentration is much greater than K_M?

Enzyme Kinetics

$$\text{rate} = \frac{k_2[E]_0[S]}{K_M + [S]};$$

$$K_M = \frac{k_{1r} + k_2}{k_{1f}}$$



(b) If the substrate concentration is very large, then all the enzyme active sites will be filled with substrate molecules and the reaction rate will depend on the efficiency with which substrate is converted into product per enzyme molecule. At this maximum rate, how many molecules of pyrophosphate are converted into orthophosphate per second per enzyme molecule? (This number is called the *turnover number* for the enzyme. *Hint:* the number of molecules of product turned over by each molecule of enzyme is equal to the number of moles of product turned over by a mole of enzyme.)