

Chem 452 - Lecture 4

Enzymes

Part 3

Question of the Day: What are the three major types of enzyme inhibition and how can kinetics be used to distinguish between them?

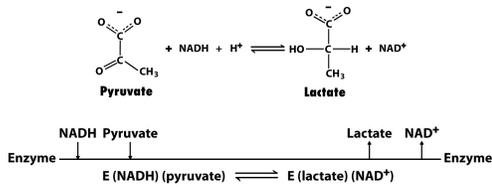
Enzyme Kinetics

† Most Reactions involve multiple substrates.

- There are three different ways that the binding substrates can occur.
 - Ordered sequential
 - Random sequential
 - Double displacement (Ping Pong)
- Determining the binding order can tell you something about the mechanism of the reaction.

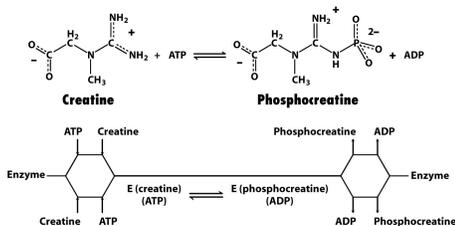
Enzyme Kinetics

† Ordered sequential



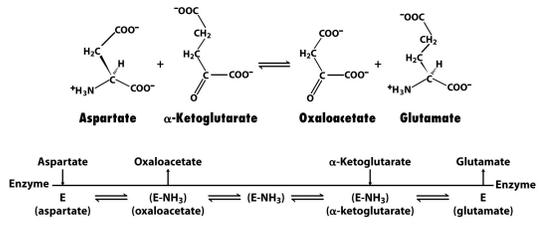
Enzyme Kinetics

† Random sequential



Enzyme Kinetics

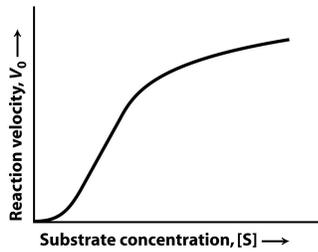
† Double displacement (Ping Pong)



Chem 452, Lecture 4 - Enzymes 5

Enzyme Kinetics

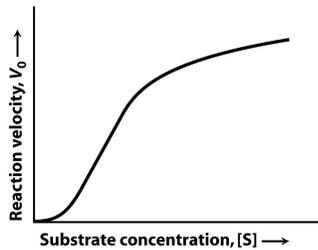
† Not all enzyme obey the Michaelis-Menten model.



Chem 452, Lecture 4 - Enzymes 6

Enzyme Kinetics

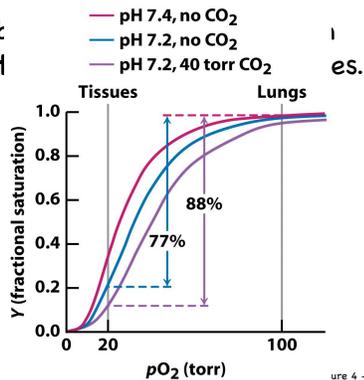
† The behavior is often seen with **allosterically regulated enzymes**.



Chem 452, Lecture 4 - Enzymes 7

Enzyme Kinetics

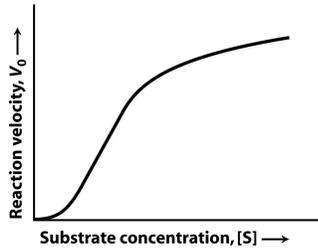
† The **allost**



ure 4 - Enzymes 7

Enzyme Kinetics

- † The behavior is often seen with **allosterically regulated** enzymes.



Chem 452, Lecture 4 - Enzymes 7

Enzyme Inhibition

- † The inhibition of enzyme activity can be physiological or not.
- † It can be reversible or irreversible.
- † Many drugs, pesticides and herbicides operate by inhibiting enzyme activity

Chem 452, Lecture 4 - Enzymes 8

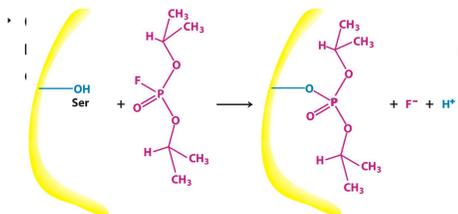
Enzyme Inhibition

- † Irreversible inhibition, while not usually physiological, can be used as a tool to study an enzyme.
- Catalytic groups at the active site are often more reactive than groups elsewhere on the enzyme.

Chem 452, Lecture 4 - Enzymes 9

Enzyme Inhibition

- † Irreversible inhibition, while not usually physiological, can be used as a tool to study an enzyme.



DIPF is a powerful nerve gas toxin

Chem 452, Lecture 4 - Enzymes 9

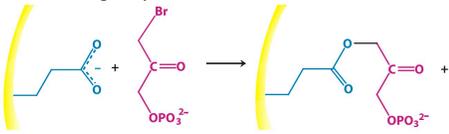
Enzyme Inhibition

- † Irreversible inhibition, while not usually physiological, can be used as a tool to study an enzyme.
- Catalytic groups at the active site are often more reactive than groups elsewhere on the enzyme.

Chem 452, Lecture 4 - Enzymes 9

Enzyme Inhibition

- † Irreversible inhibition, while not usually physiological, can be used as a tool to study an enzyme.
- Catalytic groups at the active site are often



Chem 452, Lecture 4 - Enzymes 9

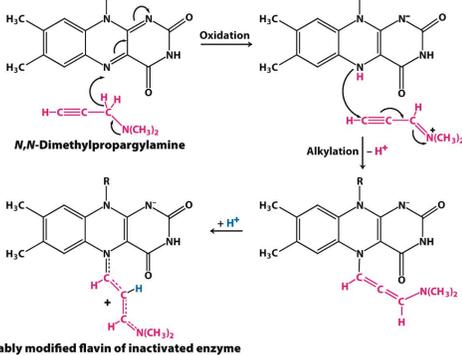
Enzyme Inhibition

- † Irreversible inhibition, while not usually physiological, can be used as a tool to study an enzyme.
- Catalytic groups at the active site are often more reactive than groups elsewhere on the enzyme.

Chem 452, Lecture 4 - Enzymes 9

Enzyme Inhibition

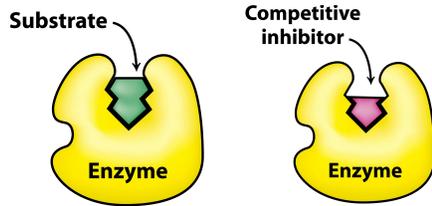
- † Flavin prosthetic group Suicide inhibitors of monoamine oxidase (MAO)



9

Enzyme Inhibition

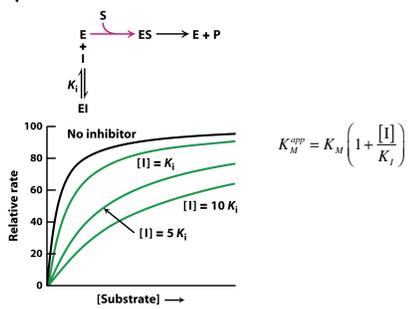
+ Competitive Inhibition



Chem 452, Lecture 4 - Enzymes 12

Enzyme Inhibition

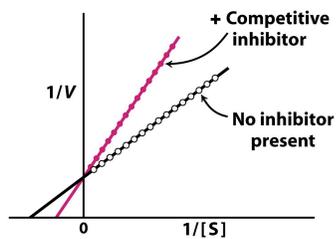
+ Competitive Inhibition



Chem 452, Lecture 4 - Enzymes 13

Enzyme Inhibition

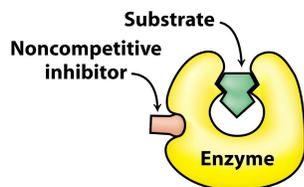
+ Competitive Inhibition



Chem 452, Lecture 4 - Enzymes 14

Enzyme Inhibition

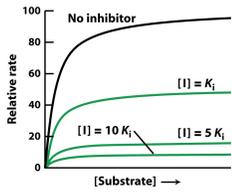
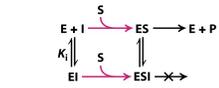
+ Noncompetitive Inhibition



Chem 452, Lecture 4 - Enzymes 15

Enzyme Inhibition

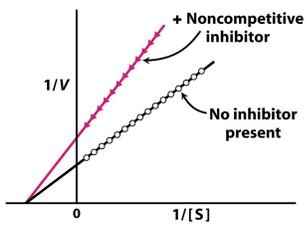
+ Noncompetitive Inhibition



$$V_{MAX}^{app} = \frac{V_{MAX}^{app}}{\left(1 + \frac{[I]}{K_i}\right)}$$

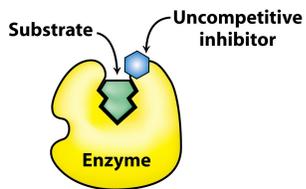
Enzyme Inhibition

+ Noncompetitive Inhibition



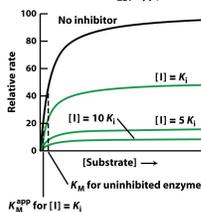
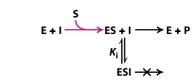
Enzyme Inhibition

+ Uncompetitive Inhibition



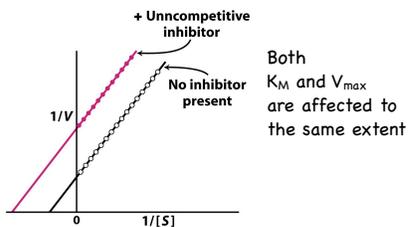
Enzyme Inhibition

+ Uncompetitive Inhibition



Enzyme Inhibition

+ Uncompetitive Inhibition



Chem 452, Lecture 4 - Enzymes 20

Problem

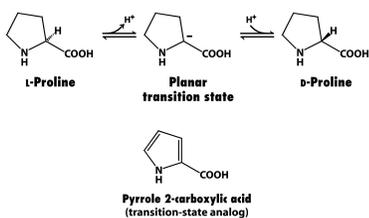
- C) Ibuprofen is an inhibitor of the enzyme *prostaglandin endoperoxide synthase*. By inhibiting the synthesis of *prostaglandins*, ibuprofen reduces both inflammation and pain. Using the data below, determine the type of inhibition that ibuprofen exerts on prostaglandin endoperoxide synthase

[S] {mM}	v_o {mM/min}	v_o {mM/min} /w Ibuprofen
0.5	23.5	16.67
1	32.2	25.25
1.5	36.9	30.49
2.5	41.8	37.04
3.5	44	38.91

21

Enzyme Inhibition

- + Some inhibitors are transition state analogues instead of substrate analogues.



Chem 452, Lecture 4 - Enzymes 22

Thermodynamics

"I think that enzymes are molecules that are complementary in structure to the activated complexes of the reactions that they catalyze, that is, to the molecular configuration that is intermediate between the reacting substance and the products of reaction for these catalyzed processes. the attraction of the enzyme molecule for the activated complex would thus lead to a decrease in its energy and hence to the decrease in the energy of activation of the reaction and to the increase in the rate of the reaction."

- Linus Pauling (Nature 161 (1948):707-709)

Chem 452, Lecture 4 - Enzymes 23

Synthetic Enzymes

† Antibody enzymes (Abzymes)

- Antibodies raised to transition state analogues exhibit enzymatic activity



Antibodies raised to this compound have ferrochelatase activity ($\approx 2,500$ x the uncatalyzed reaction)

Chem 452, Lecture 4 - Enzymes 24

Enzyme Classification

† Enzymes are classified based on the types of reactions they catalyze

TABLE 8.8 Six major classes of enzymes

Class	Type of reaction	Example	Chapter
1. Oxidoreductases	Oxidation-reduction	Lactate dehydrogenase	16
2. Transferases	Group transfer	Nucleoside monophosphate kinase (NMP kinase)	9
3. Hydrolases	Hydrolysis reactions (transfer of functional groups to water)	Chymotrypsin	9
4. Lyases	Addition or removal of groups to form double bonds	Fumarase	17
5. Isomerases	Isomerization (intramolecular group transfer)	Triose phosphate isomerase	16
6. Ligases	Ligation of two substrates at the expense of ATP hydrolysis	Aminoacyl-tRNA synthetase	30

Nomenclature Committee of the International Union of Biochemistry and Molecular Biology (NC-IUBMB)

<http://www.chem.qmul.ac.uk/iubmb/enzyme/>

Chem 452, Lecture 4 - Enzymes 25

Next up

† Catalytic Strategies (Chapter 9)

- Protease reaction (Hydrolysis rxn)
- Carbonic anhydrase (Hydration rxn)
- Restriction endonuclease (Hydrolysis rxn)
- Myosin ATPase

Chem 452, Lecture 4 - Enzymes 26