General Properties of Enzymes

- Enzymes differ from ordinary chemical catalysts in reaction rate, reaction conditions, reaction specificity, and control.
- The unique physical and chemical properties of the active site limit an enzyme's activity to specific substrates and reactions.
- Some enzymes require metal ions or organic cofactors.

Catalytic Mechanisms

1. Amino acid side chains that can donate or accept protons can participate in chemical reactions as acid or base catalysts: ***Acid-base catalysis***
2. Nucleophilic groups can catalyze reactions through the transient formation of covalent bonds with the substrate: ***Covalent catalysis***
3. In metal ion catalysis, the unique electronic properties of the metal ion facilitate the reaction: ***Metal ion catalysis***
4. Enzymes accelerate reactions by bringing reacting groups together and orienting them for reaction: ***Proximity and orientation effects***
5. Transition state stabilization can significantly lower the activation energy for a reaction: ***Preferential binding of the transition state complex***

Lysozyme

- Model building indicates that binding to lysozyme distorts the substrate sugar residue.
- Lysozyme's active site Asp and Glu residues promote substrate hydrolysis by acid–base catalysis, covalent catalysis, and stabilization of an oxonium ion transition state.

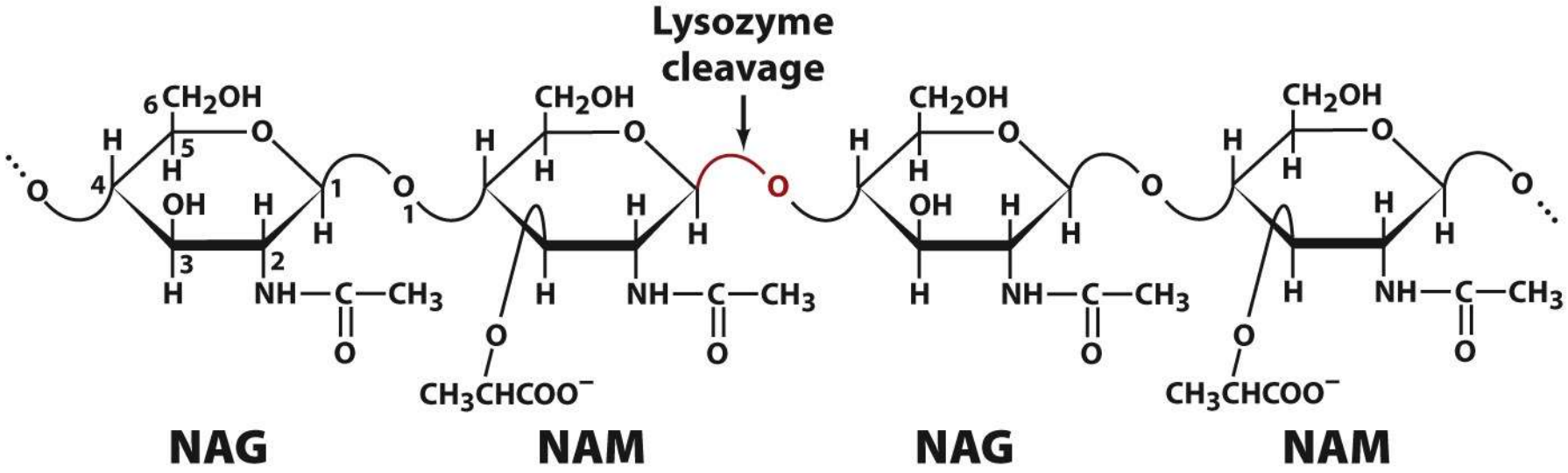
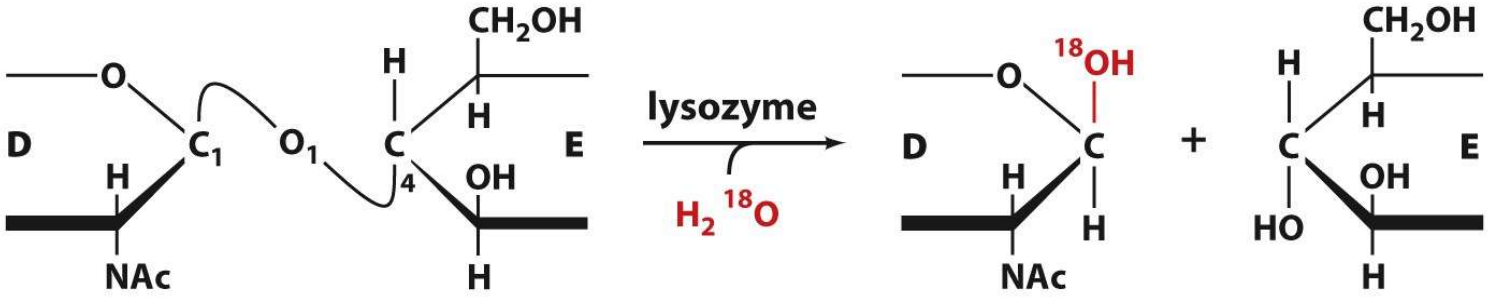


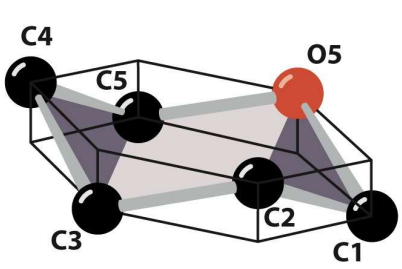
Figure 11-16

Identification of Lysozyme Cleavage Site

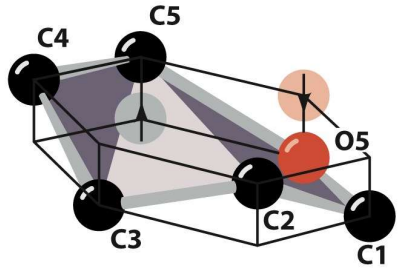


- Destroys bacterial cell walls (peptidoglycan)
- Hydrolyzing the β (1→4) glycosidic linkages from *N*-acetylmuramic acid (NAM) to *N*-acetylglucosamine (NAG)
- Also hydrolyzes β (1→4)-linked poly(NAG) (=chitin)
- Bactericidal agent or helps dispose of killed bacteria
- Hen egg white (HEW) lysozyme is the most studied.

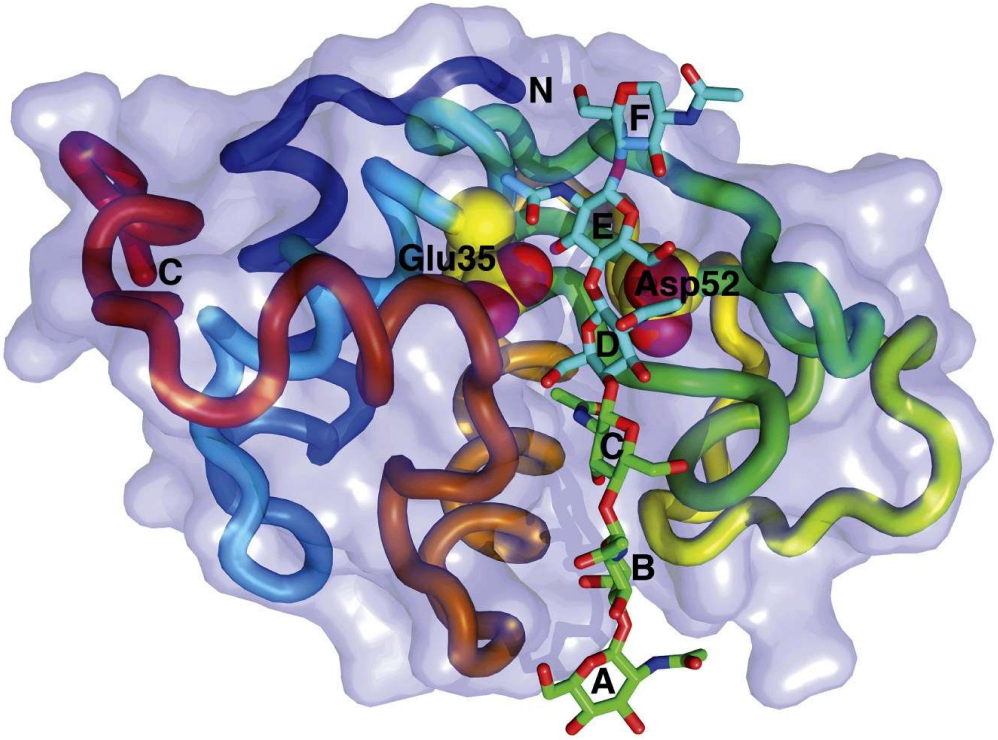
Lysozyme-Substrate Interactions



Chair conformation



Half-chair conformation



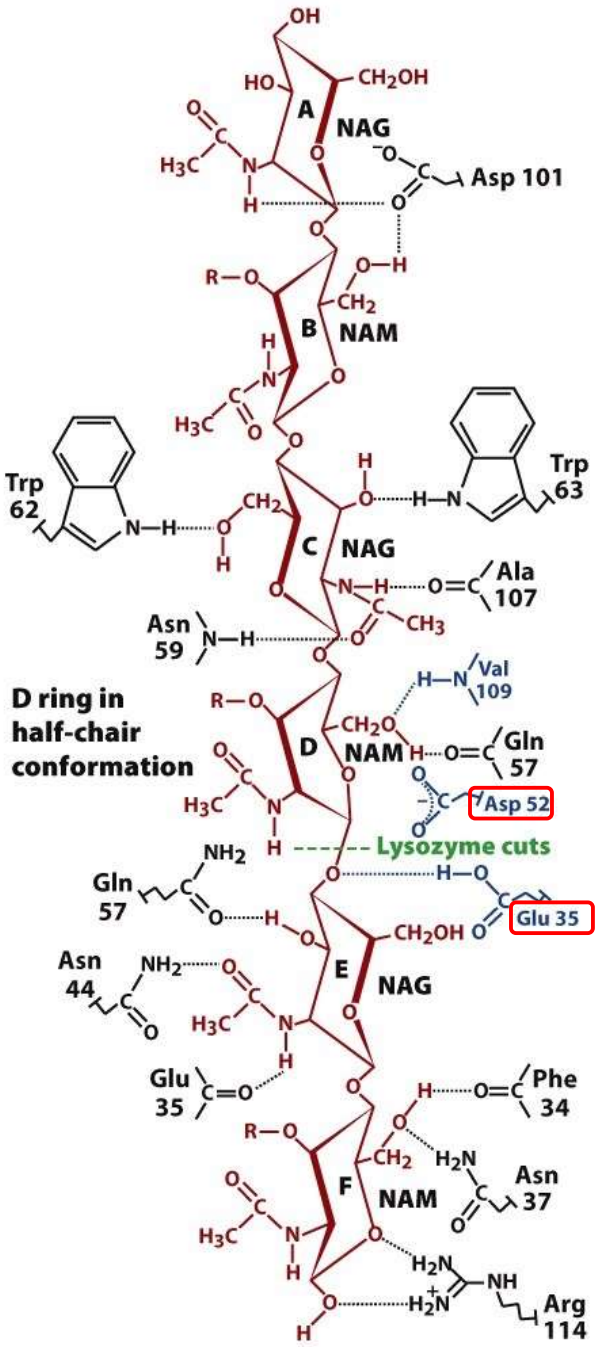


Figure 11-19

Lysozyme Reaction Mechanism

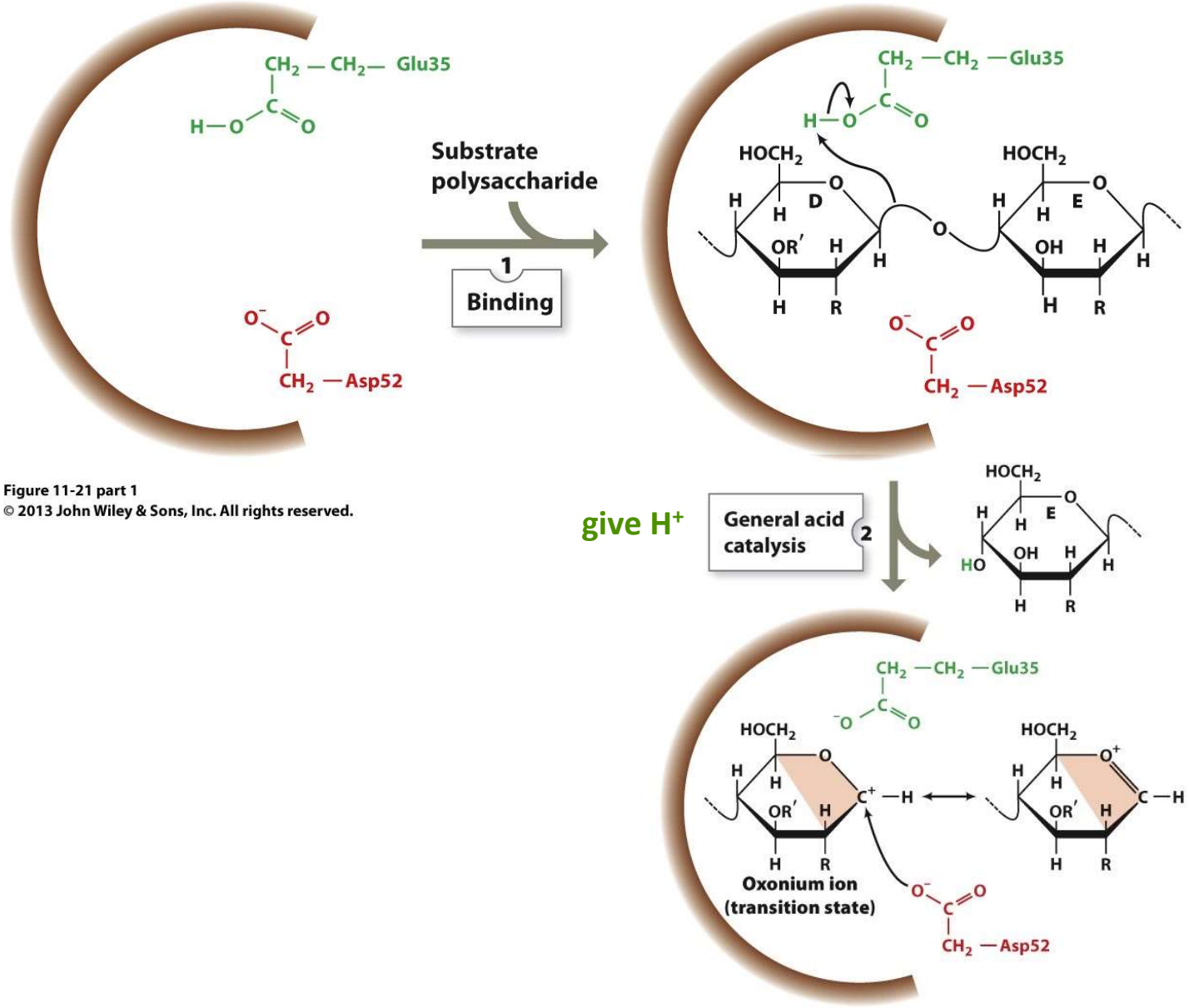
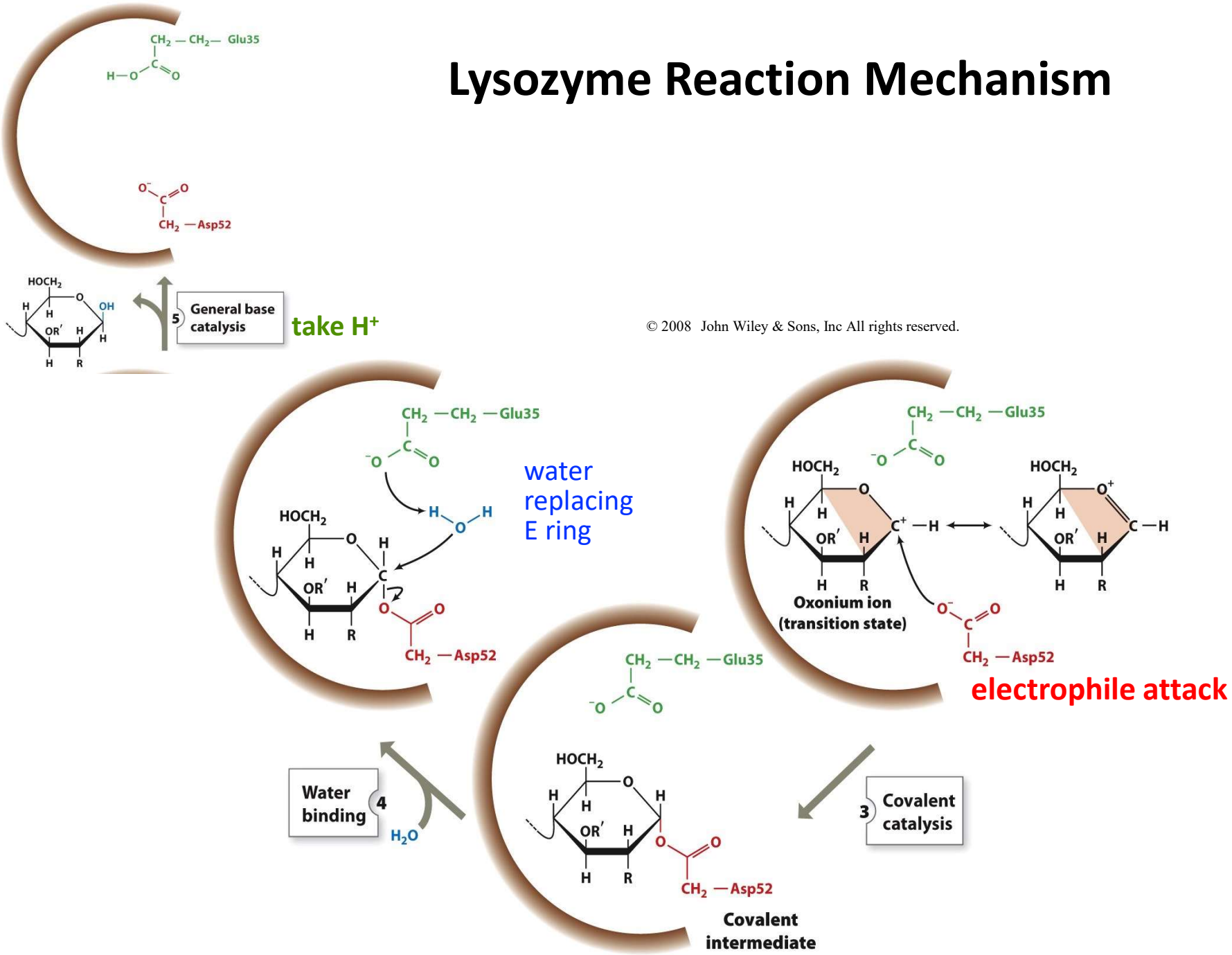
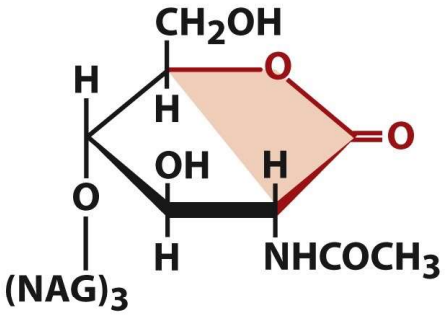


Figure 11-21 part 1

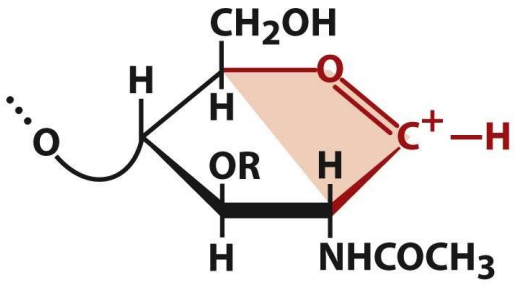
Lysozyme Reaction Mechanism

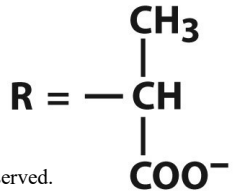


Lysozyme: The Use of Transition State Analog Inhibitor

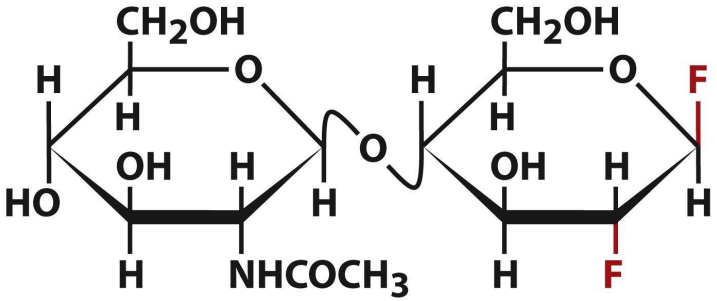


δ-Lactone analog of $(NAG)_4$

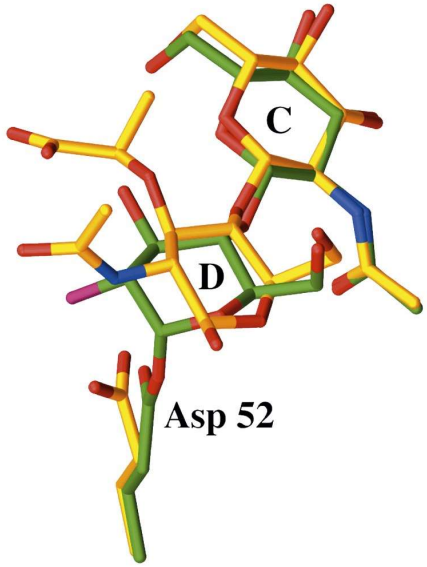




Lysozyme transition state



NAG2FGlcF



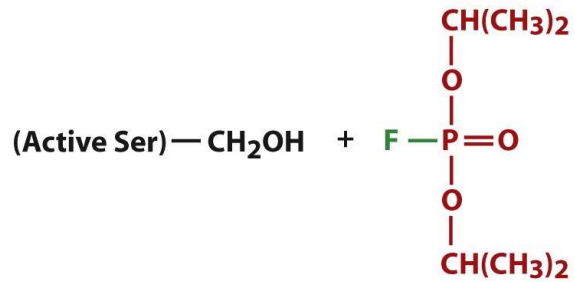
Inhibitor used to verify covalent lysozyme intermediate

Serine Proteases

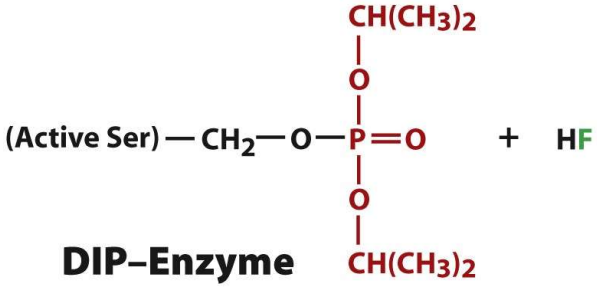
- The catalytically active **Ser**, **His**, and **Asp** residues of serine proteases were identified by chemical labeling and structural analysis.
- A binding pocket determines the substrate specificity of the various serine proteases.
- Serine proteases catalyze peptide bond hydrolysis via proximity and orientation effects, acid–base catalysis, covalent catalysis, electrostatic catalysis, and transition state stabilization.
- Zymogens are the inactive precursors of enzymes.

DIPF Irreversibly Inactivates Serine Proteases

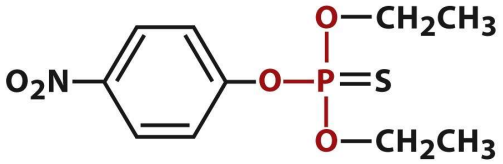
: nerve gases



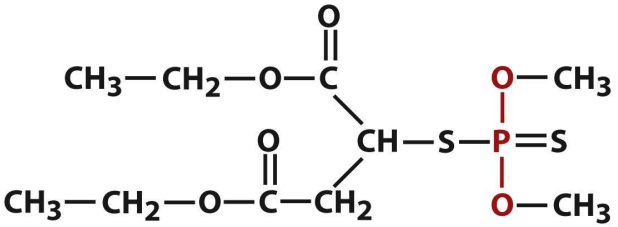
Diisopropylphospho-fluoridate (DIPF)



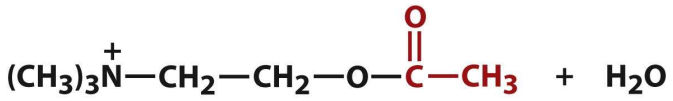
DIP–Enzyme



Parathion

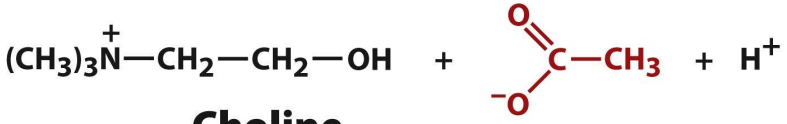


Malathion

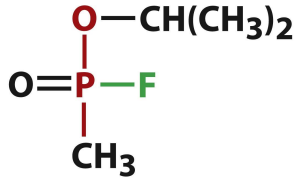


Acetylcholine

acetylcholinesterase



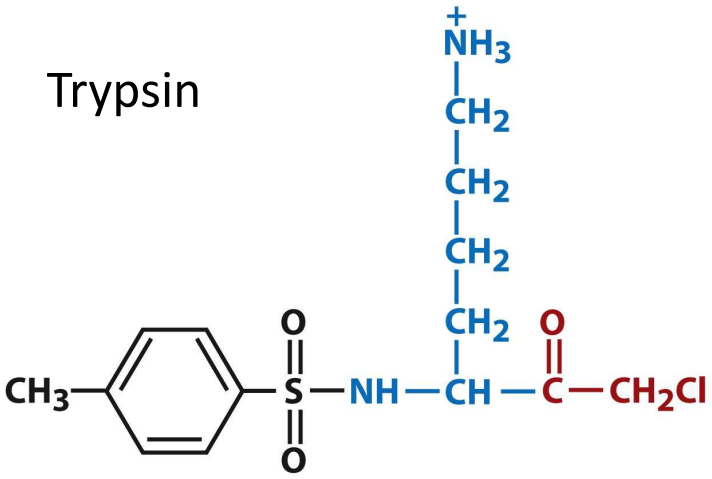
Choline



Sarin

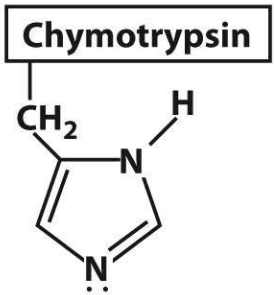
Affinity Labeling: Trypsin & Chymotrypsin

Trypsin



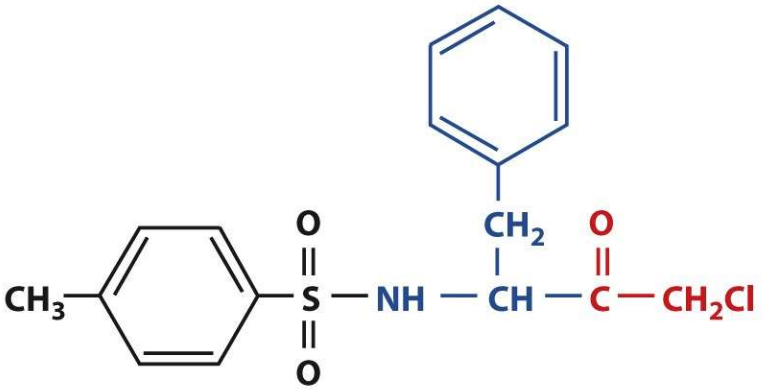
Tosyl-L-lysine chloromethylketone

Chymotrypsin



His 57

+



Tosyl-L-phenylalanine chloromethylketone (TPCK)

Chymotrypsin

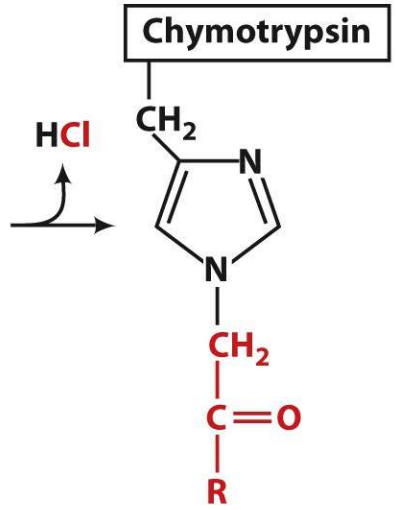


Figure 11-24

Chymotrypsin

- During digestion, dietary proteins must be broken down into small peptides by proteases.
- Chymotrypsin is one of several proteases that cuts peptides at specific locations on the peptide backbone.
- This protease is able to cleave the peptide bond adjacent to aromatic amino acids.

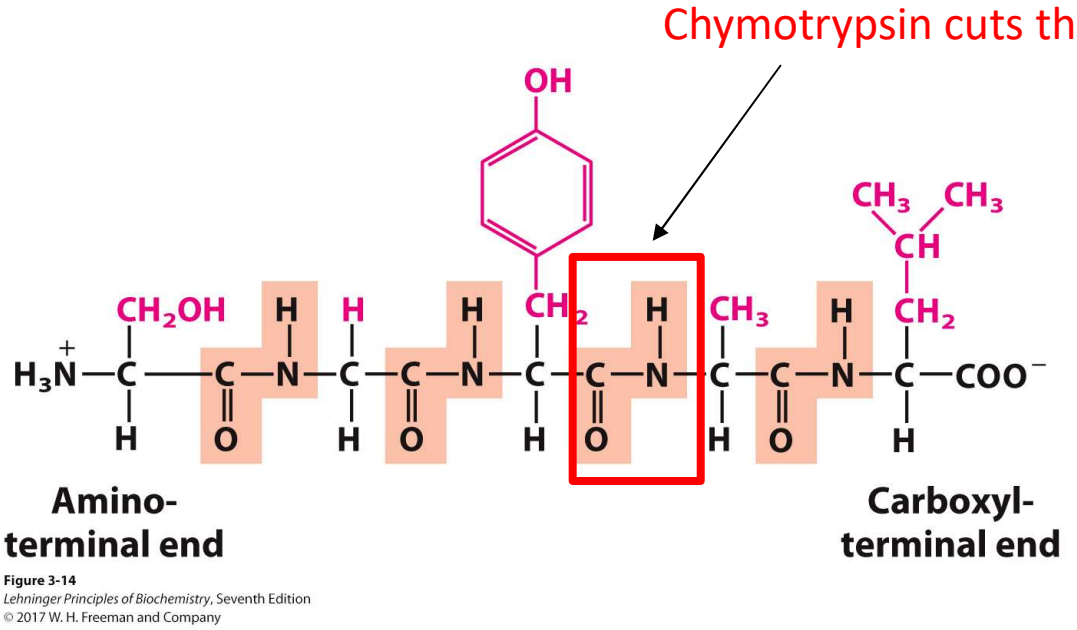


Figure 3-14
Lehninger Principles of Biochemistry, Seventh Edition

Specificity Pockets of Serine Proteases

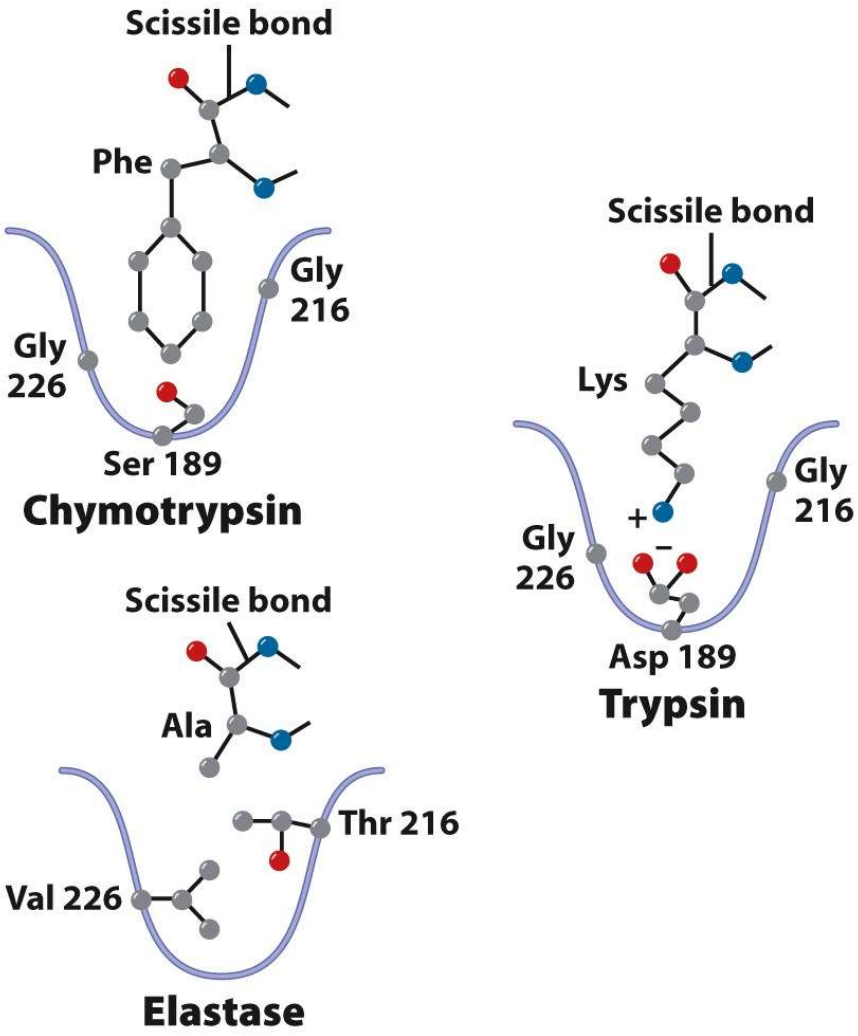


Figure 11-27

Active Site Residues in Serine Proteases

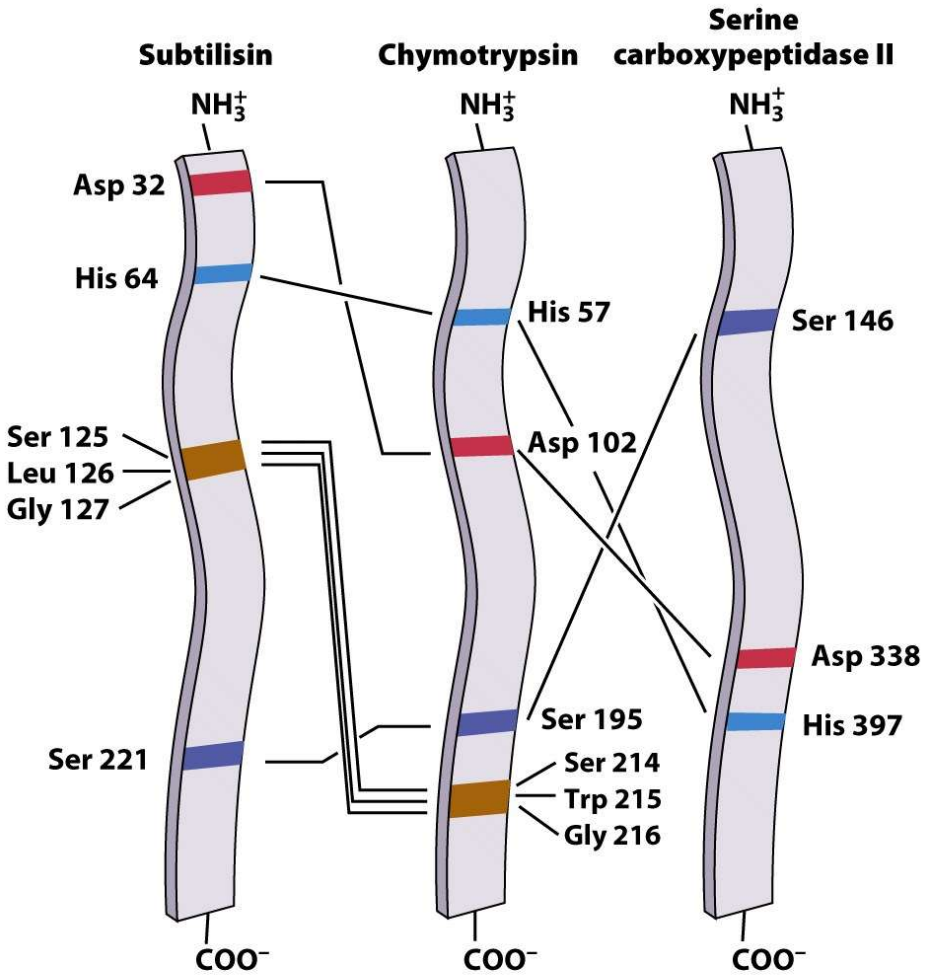


Figure 11-28

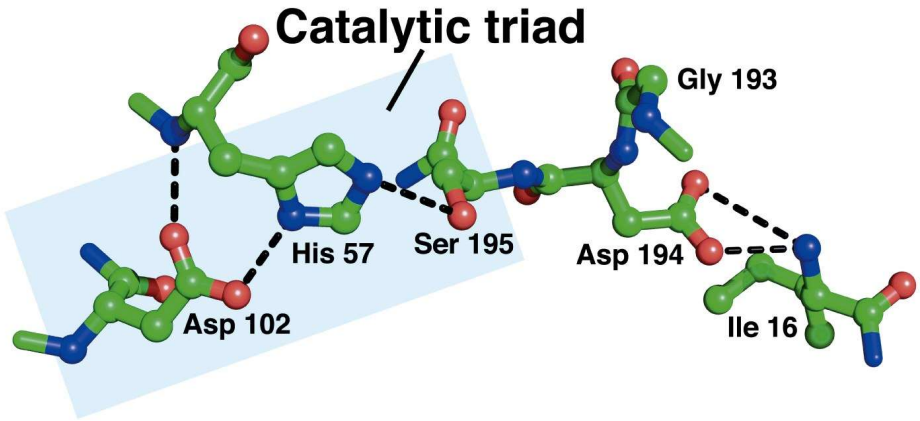
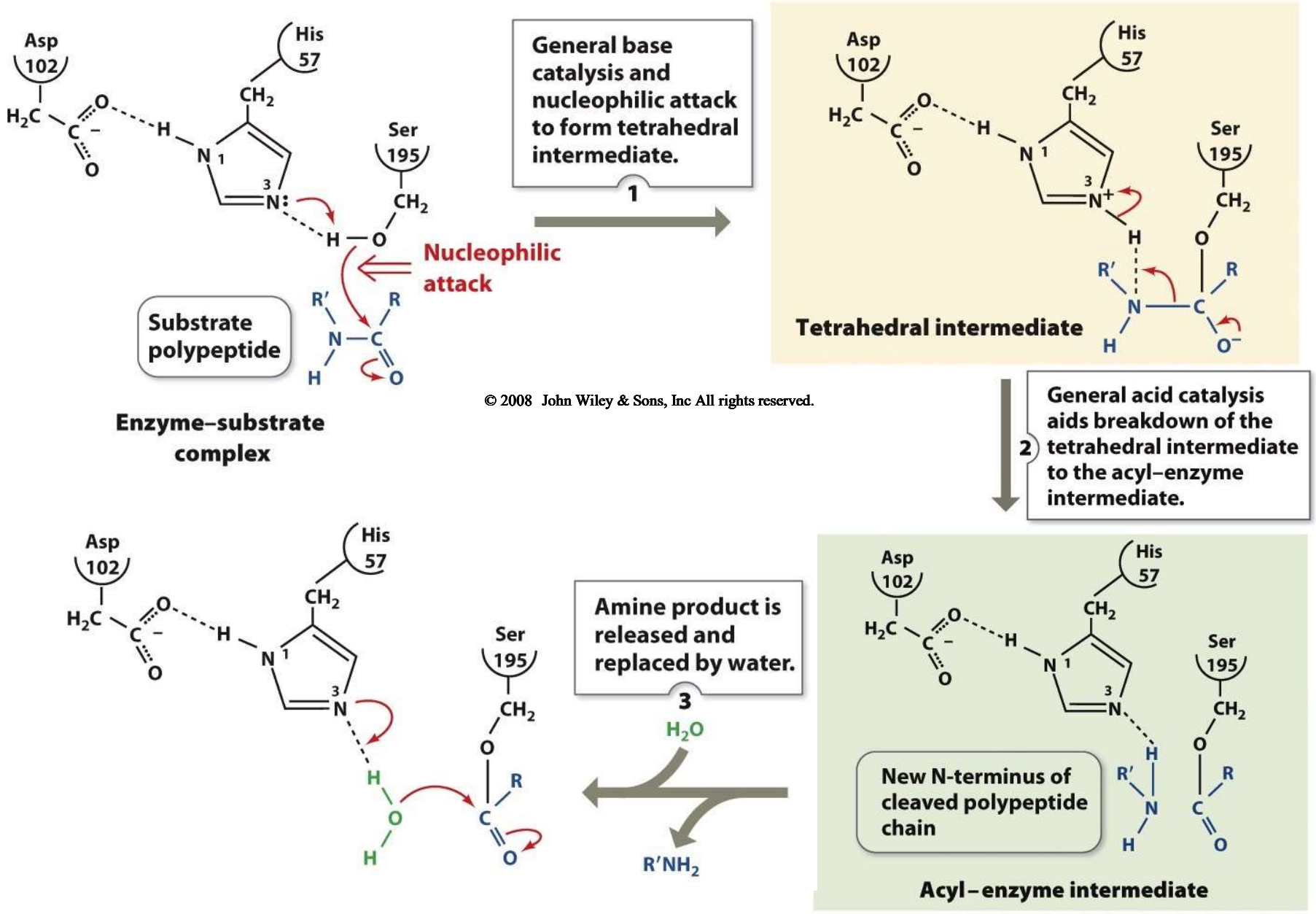


Figure 11-26

chymotrypsin

Mechanism of Serine Proteases



Mechanism of Serine Proteases

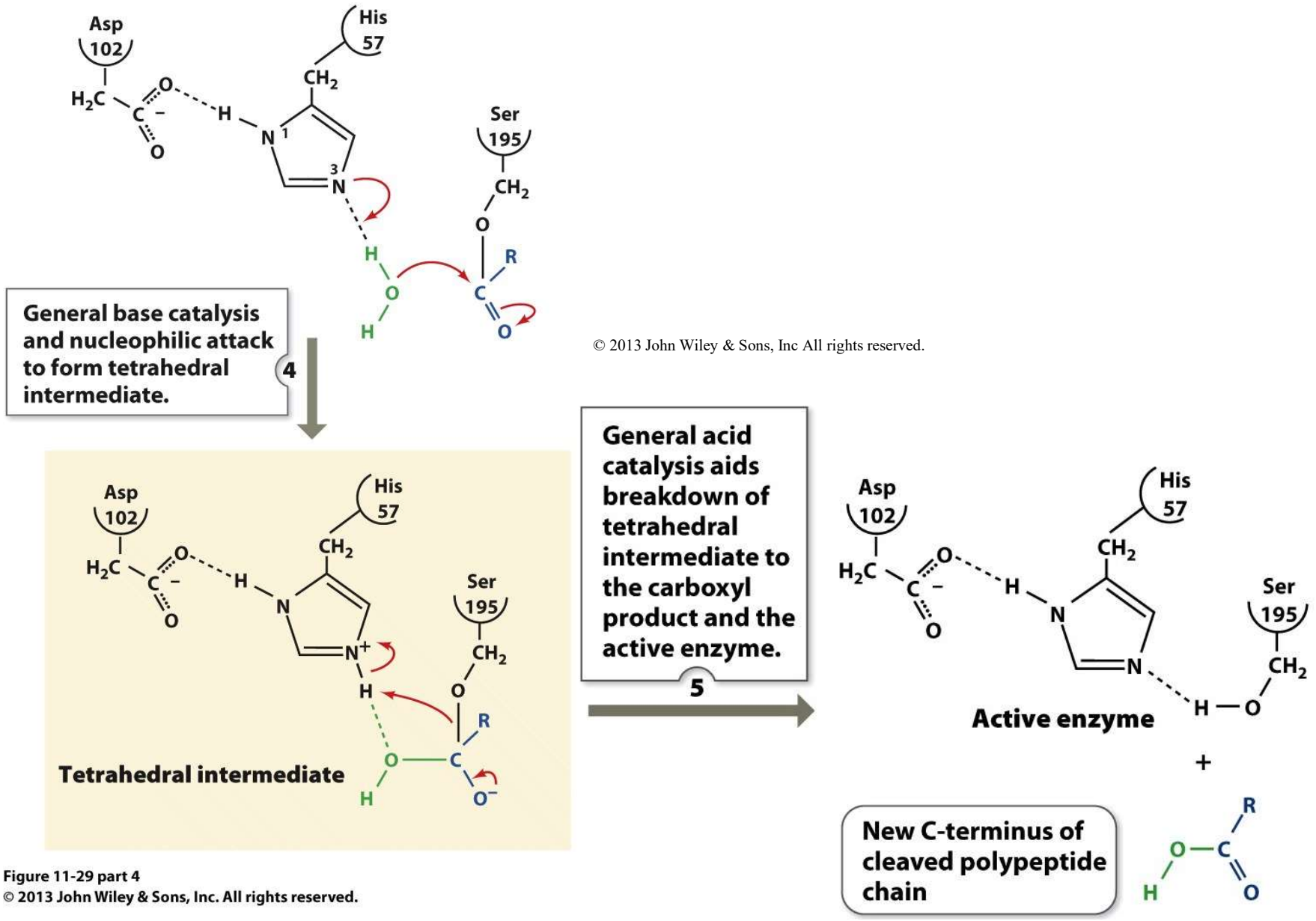
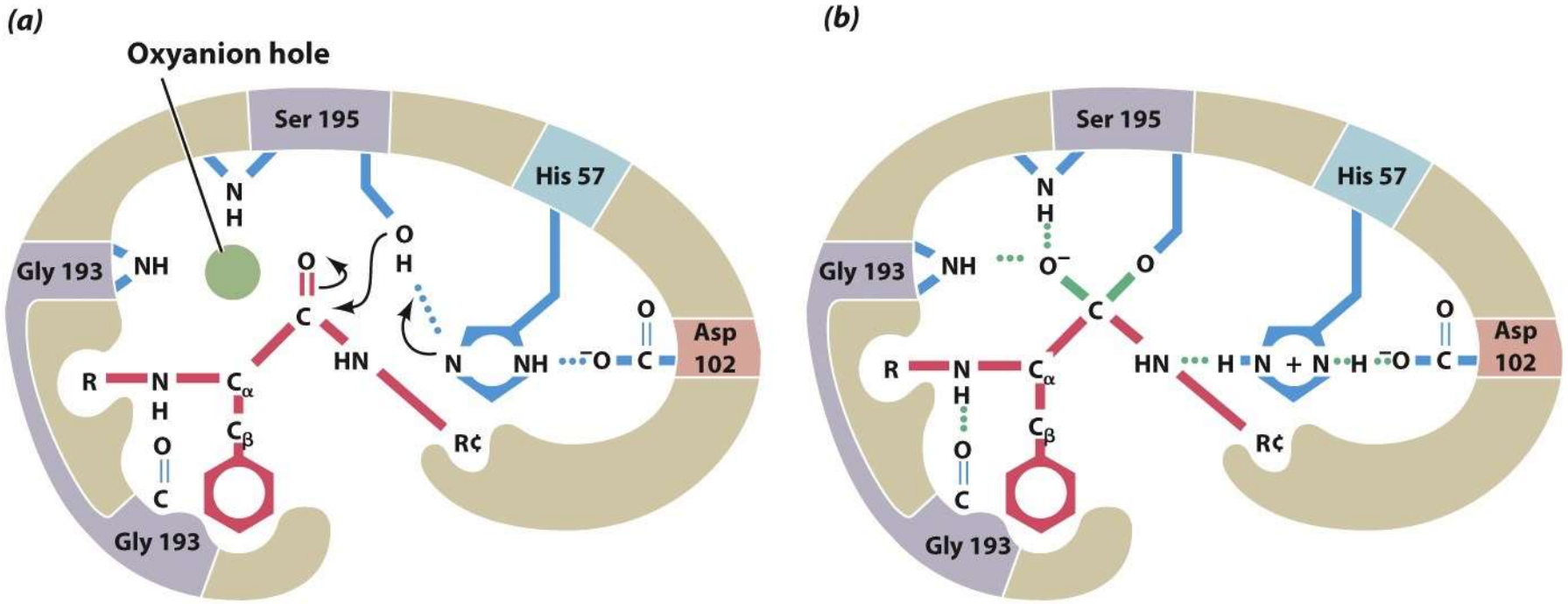
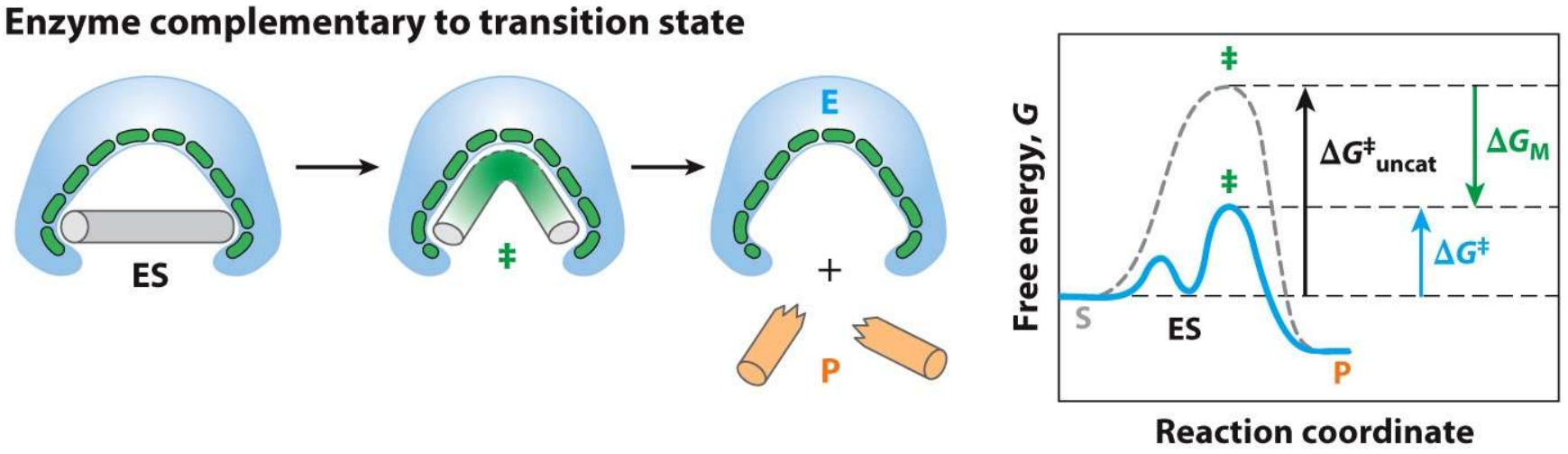


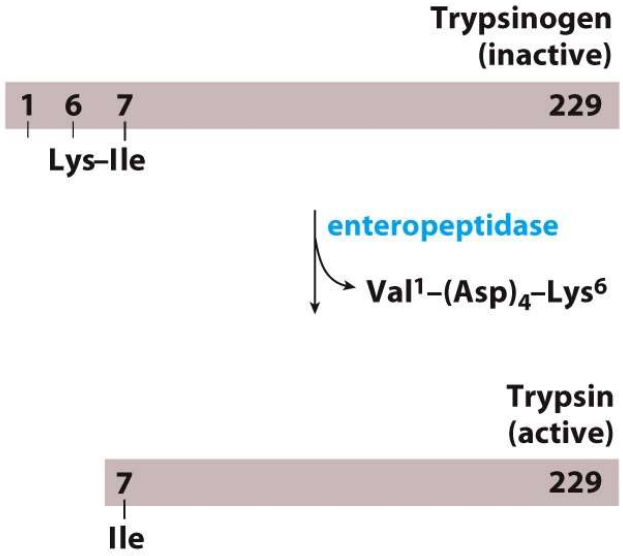
Figure 11-29 part 4

TS Stabilization in Serine Proteases

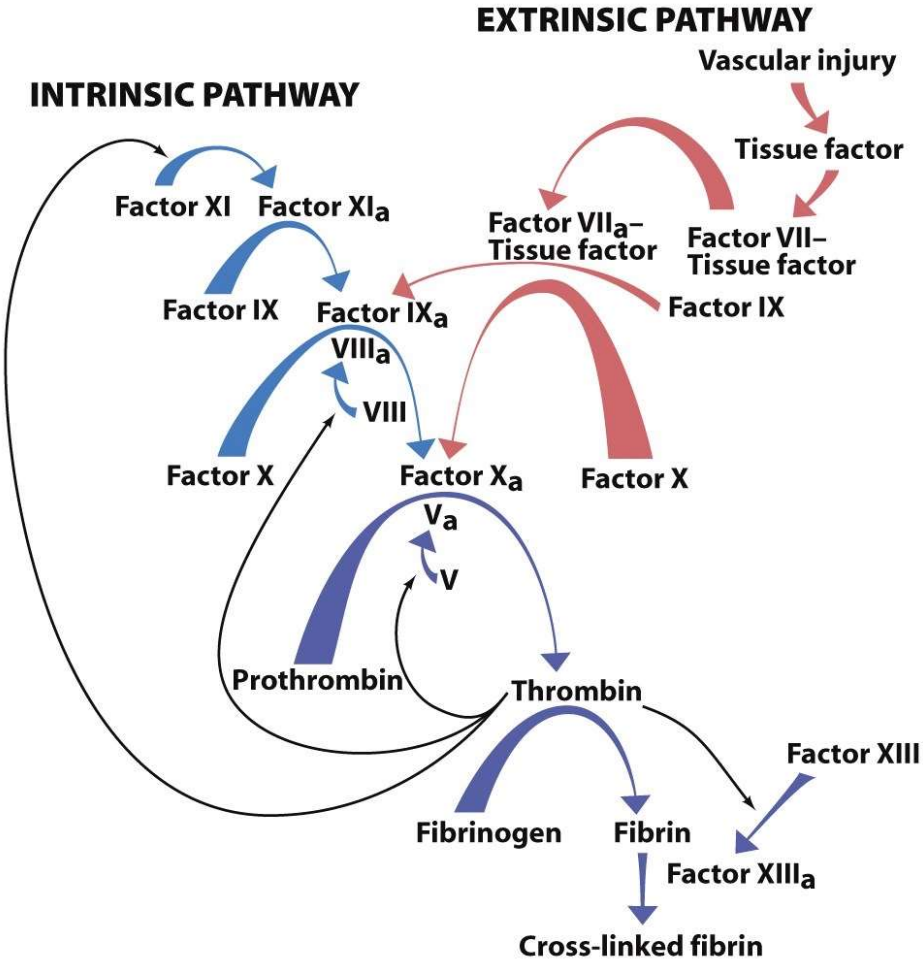




Zymogens are activated by irreversible covalent modification: blood coagulation cascade



Nelson and Cox, Lehninger Principles of biochemistry 7th edition (2017), International edition
Figure 6-39 (p232)



Specificity Pockets of Serine Proteases

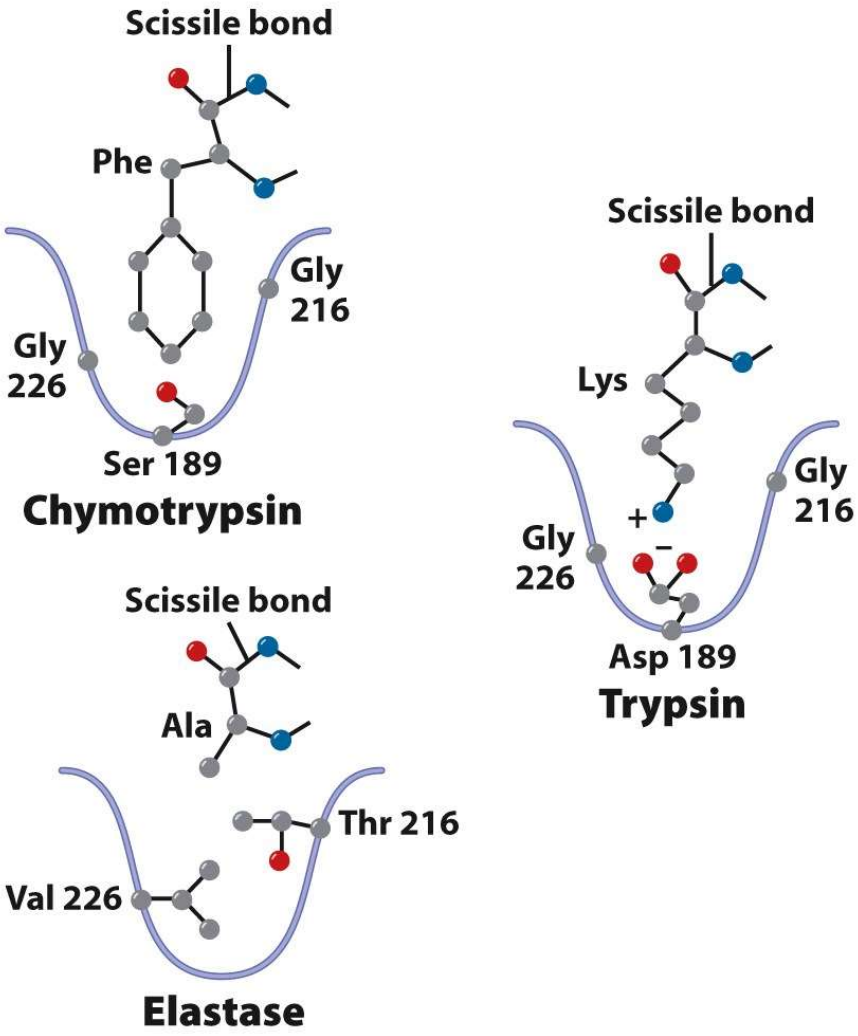


Figure 11-27