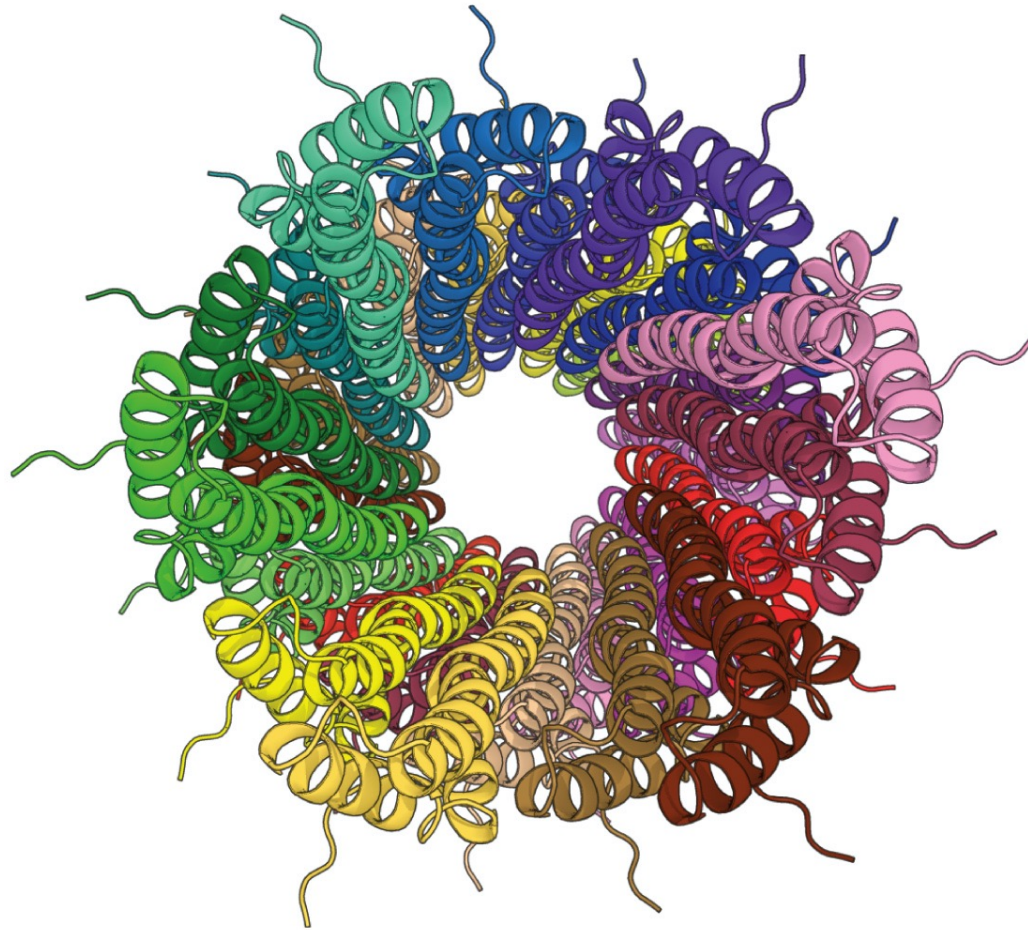


# Chapter 3 – Protein Structure and Function



**Chapter 3 Opener**  
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# Chapter 3 - Protein Structure and Function

3.1 Hierarchical Structure of Proteins

3.2 Protein Folding

3.3 Protein Binding and Enzyme Catalysis

3.4 Regulating Protein Function

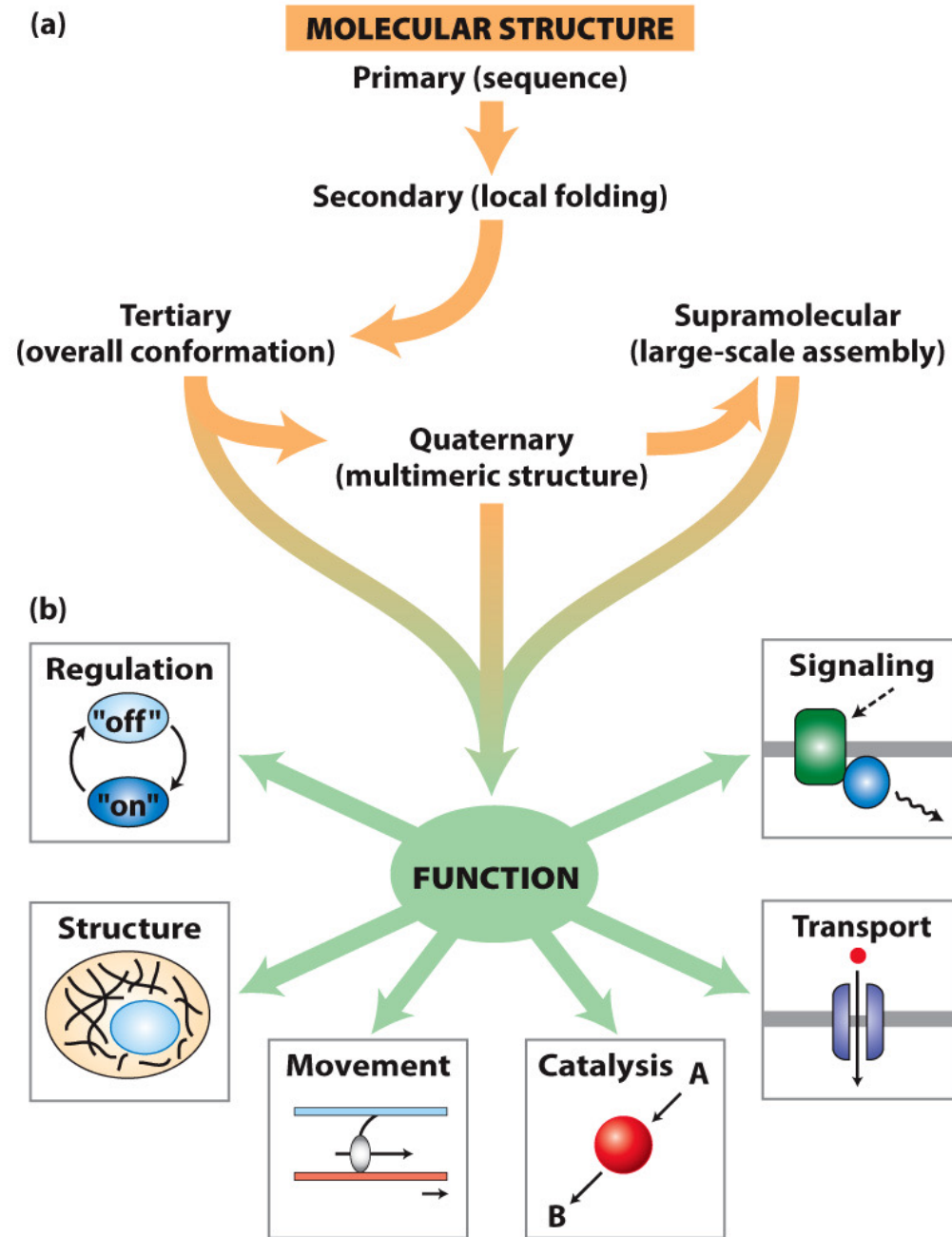
3.5 Purifying, Detecting, and Characterizing Proteins

3.6 Proteomics

# Protein Structure and Function

## 3.1 Hierarchical Structure of Proteins

- Protein sequence specifies folding into secondary and tertiary structures that either are functional units or can interact with other peptides to form quaternary structure functional units.
- Exceptional conformational flexibilities of disordered proteins contribute to their multiple functions.
- Some polypeptides with dissimilar sequences fold into similar 3D structures.
- Homologous proteins evolved from a common ancestor, have similar sequences, structures, and functions, and can be classified into families and superfamilies.



**Figure 3-1**  
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**(a) Primary structure**

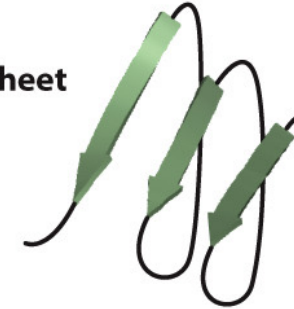
- Ala - Glu - Val - Thr - Asp - Pro - Gly -

**(b) Secondary structure**

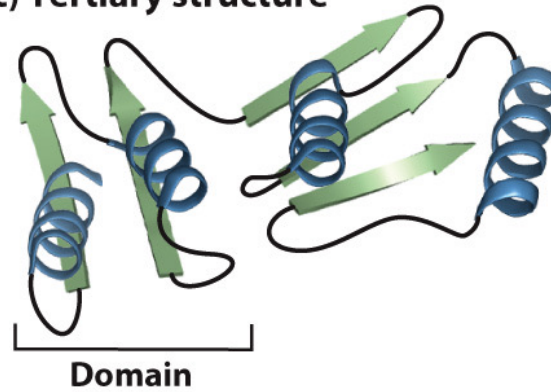
$\alpha$  helix



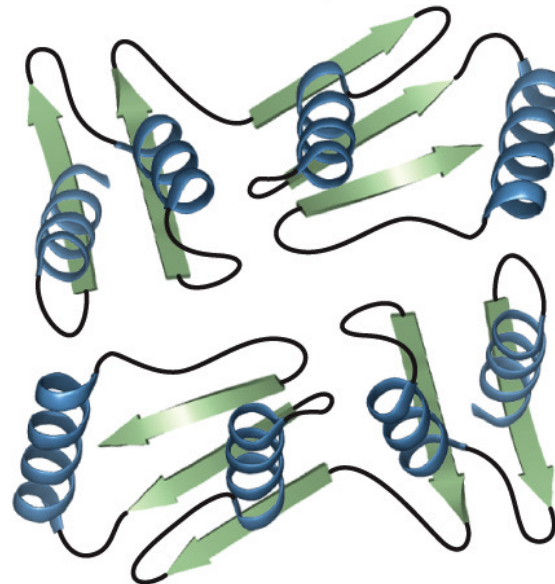
$\beta$  sheet



**(c) Tertiary structure**

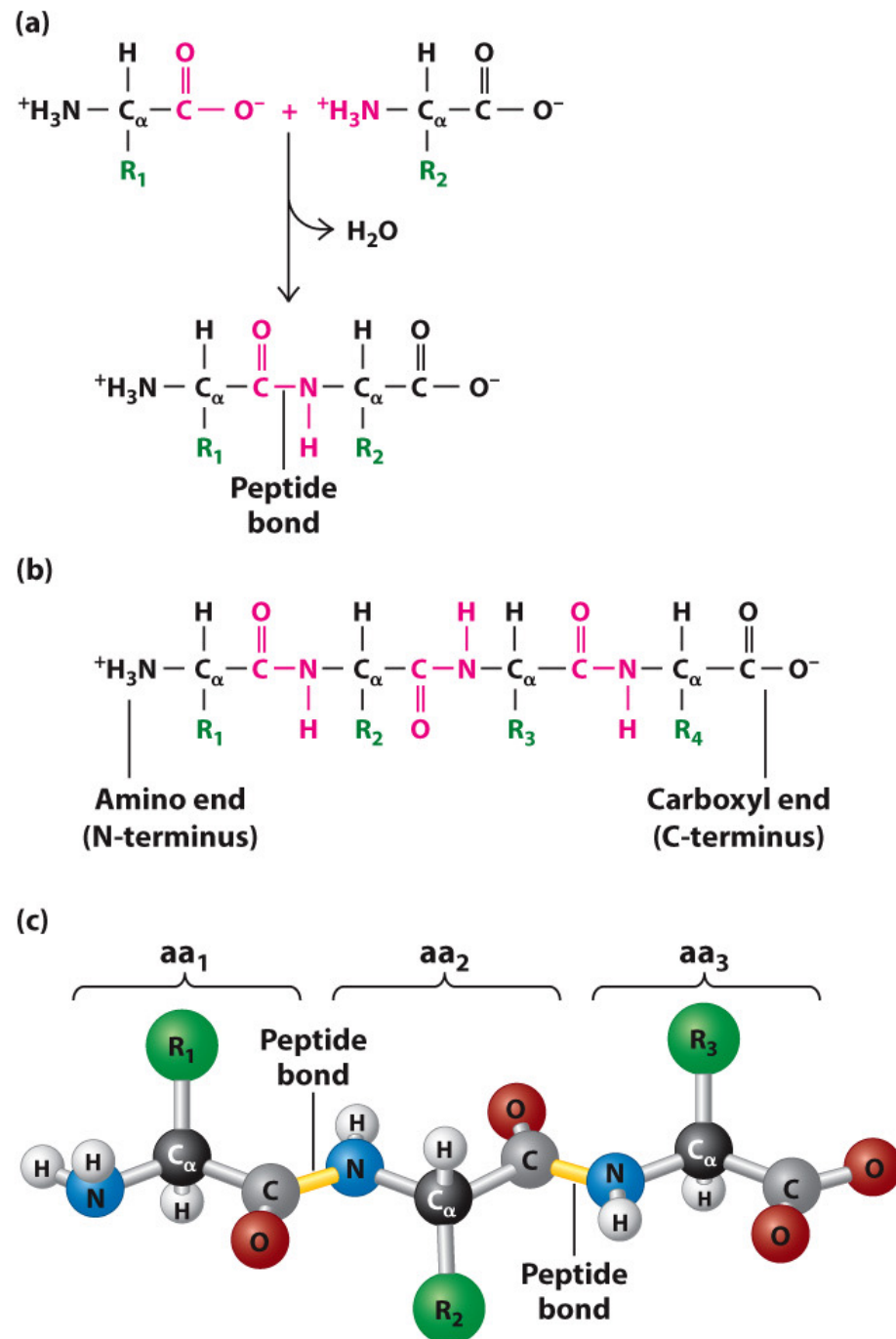


**(d) Quaternary structure**



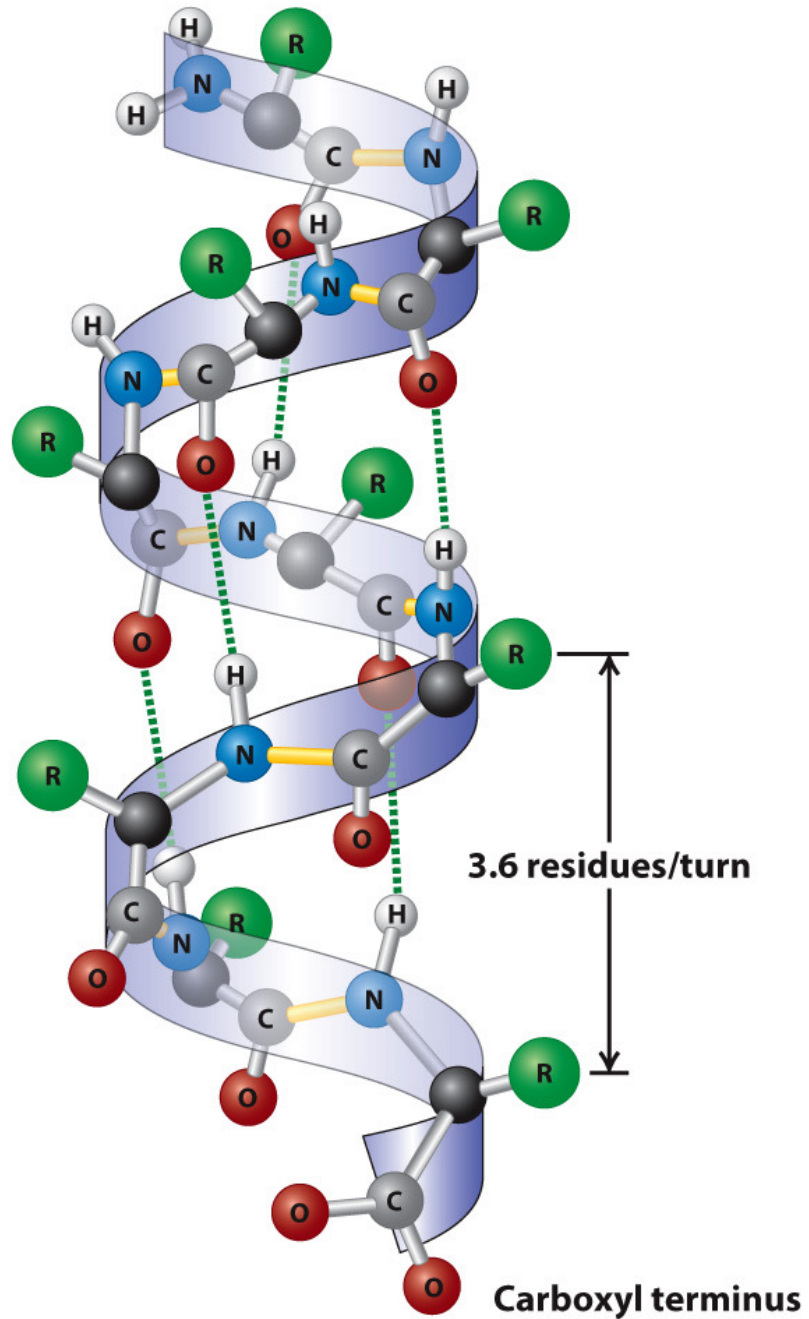
**Figure 3-2**

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**Figure 3-3**  
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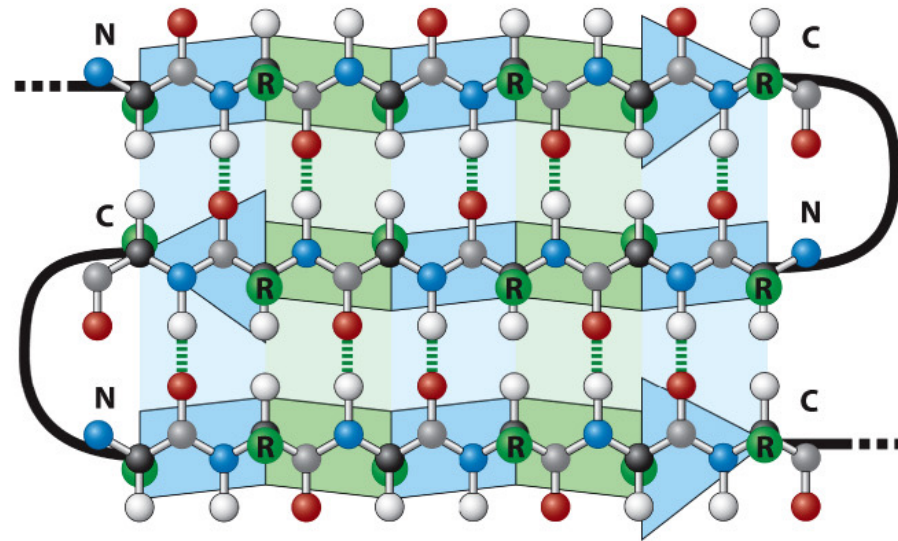
**Amino terminus**



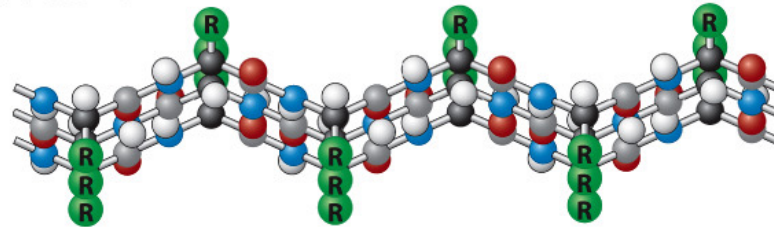
**Figure 3-4**

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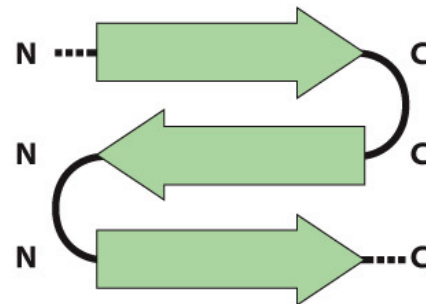
(a) Top view



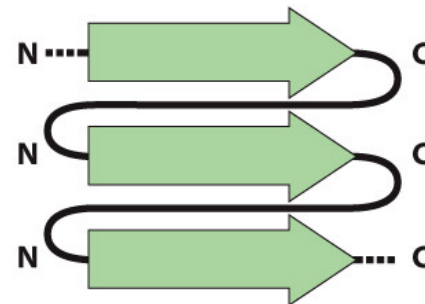
(b) Side view



(c) Anti-parallel

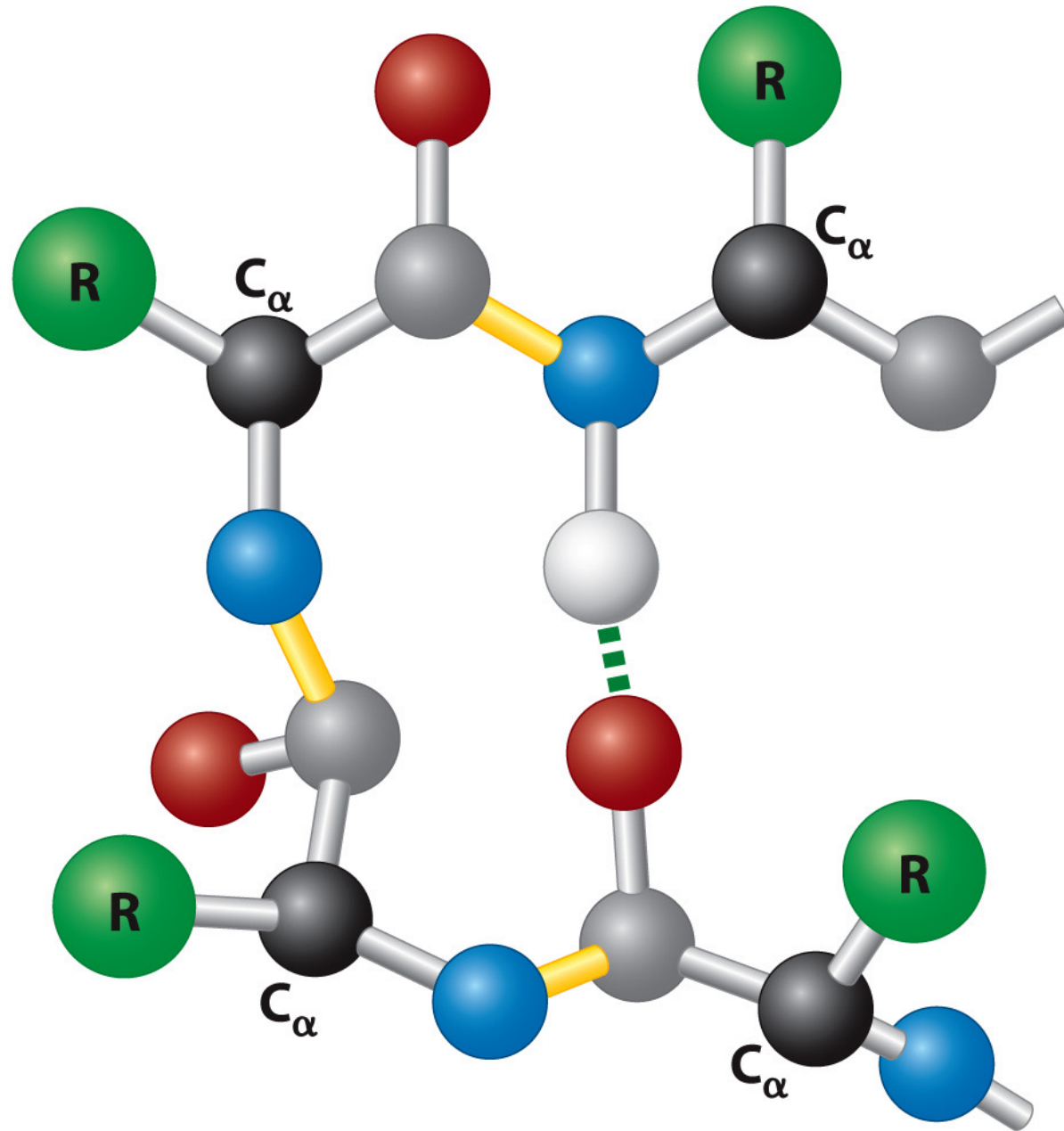


Parallel

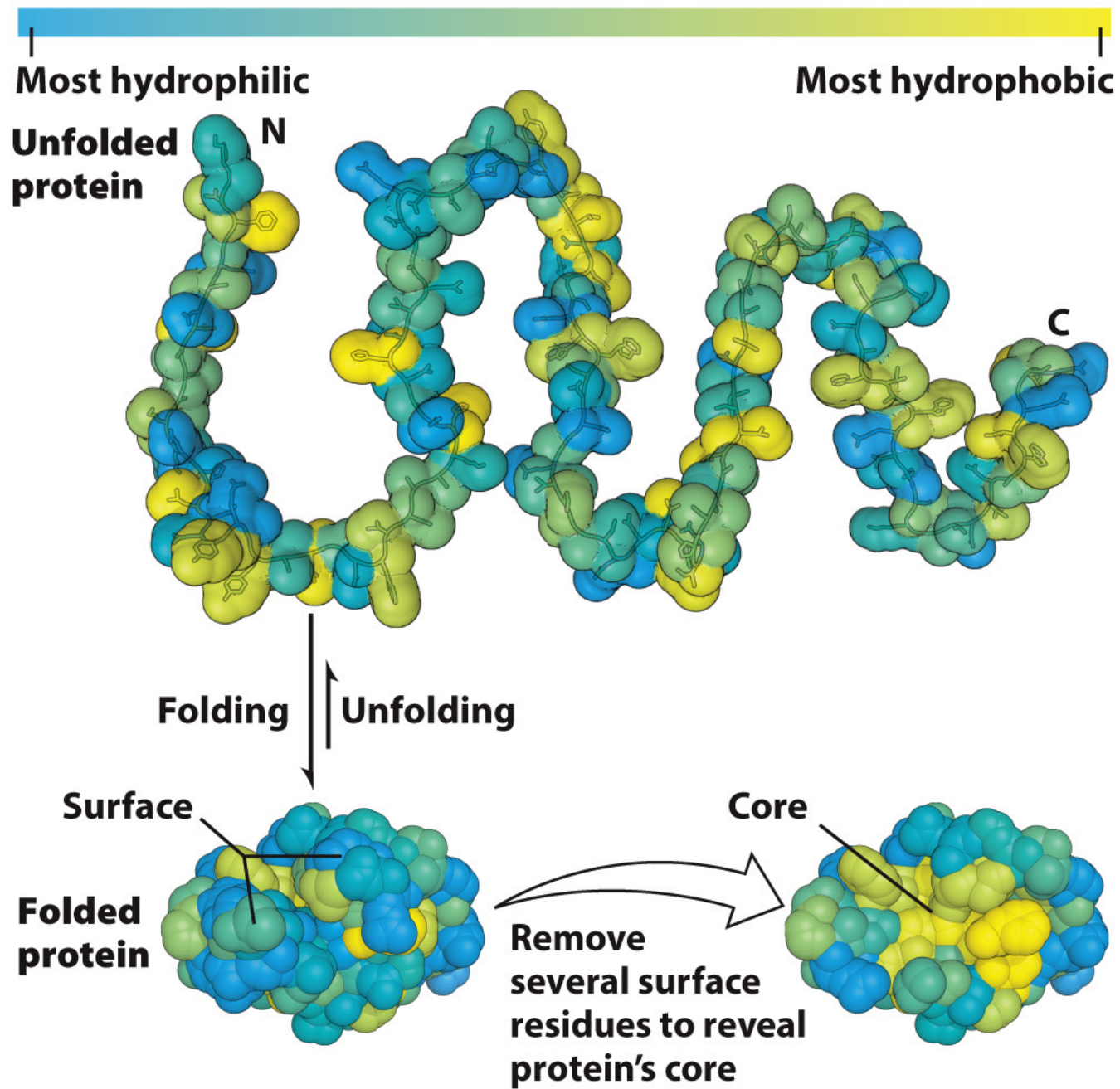


**Figure 3-5**  
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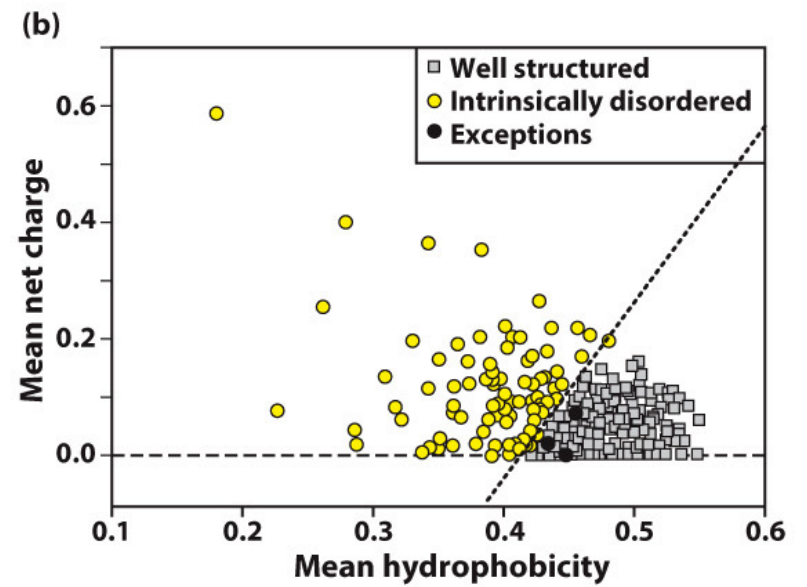
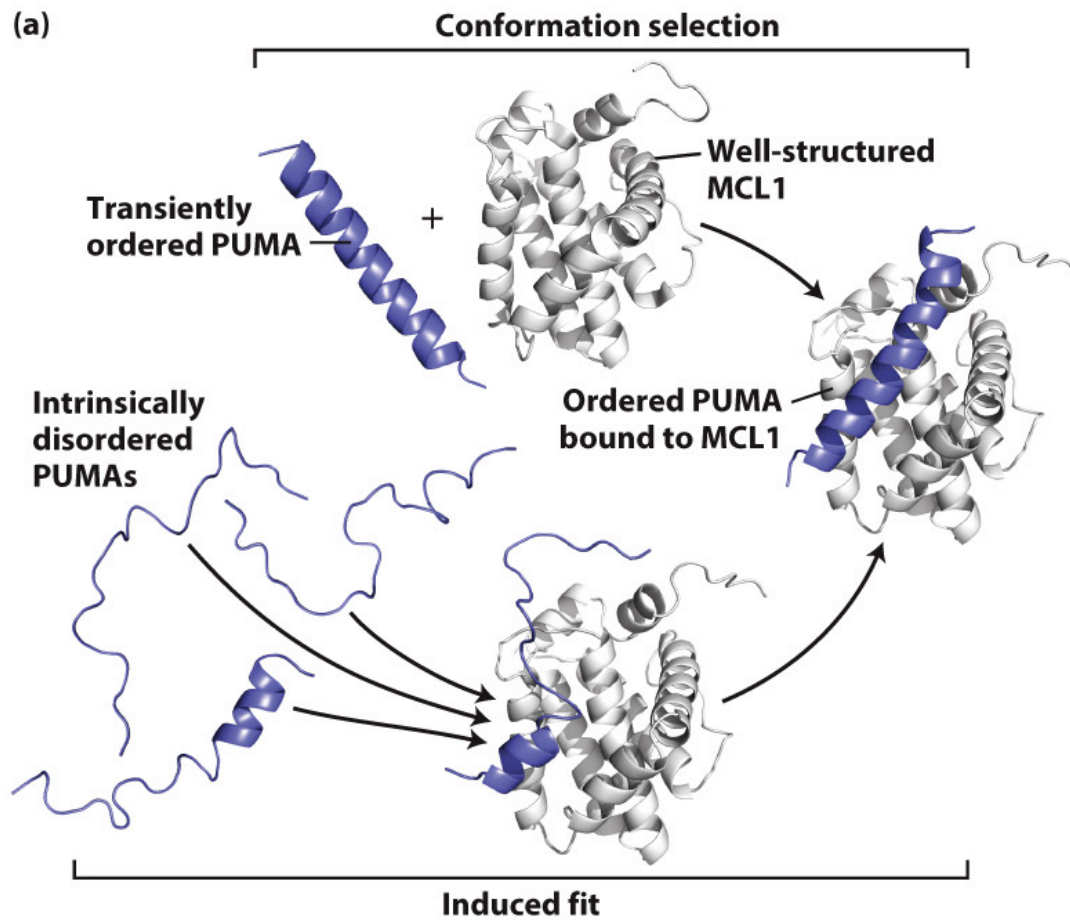




**Figure 3-6**  
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**Figure 3-7**  
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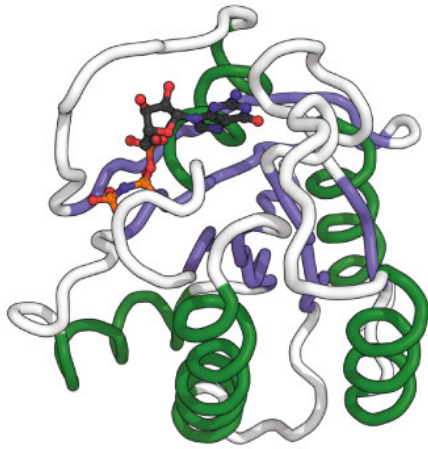


From Rogers, J. et al., "Folding and Binding of an Intrinsically Disordered Protein: Fast, but Not 'Diffusion-Limited,'" *J. Am. Chem. Soc.*, 2013, 135 (4), pp1415-1422.  
<http://pubs.acs.org/doi/pdf/10.1021/ja309527h>

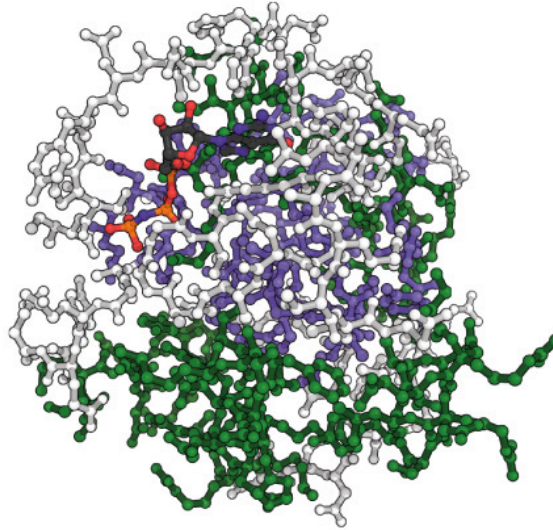
**Figure 3-8**

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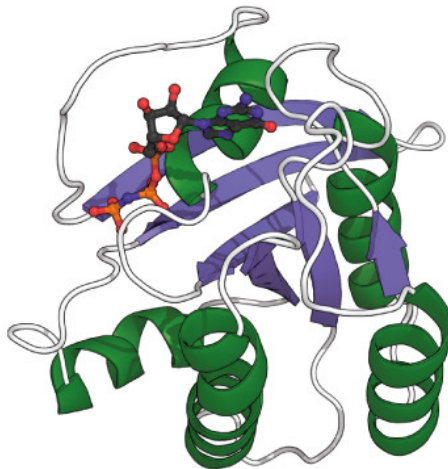
**(a) C $\alpha$  backbone trace**



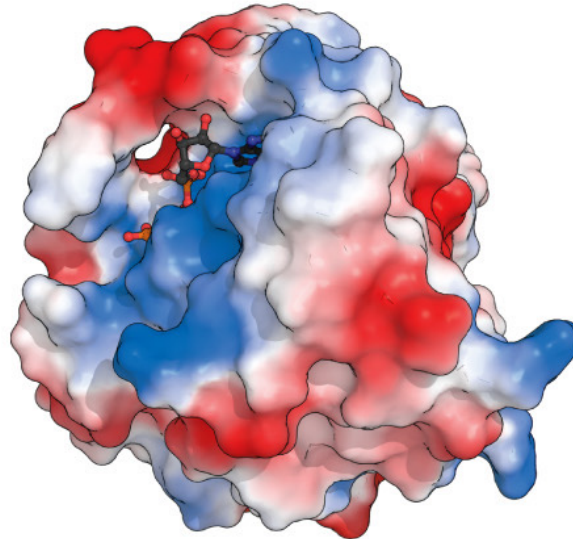
**(b) Ball-and-stick model**



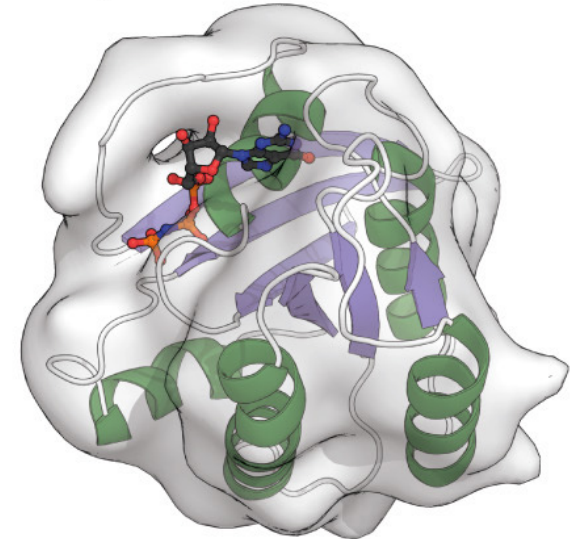
**(c) Ribbon diagram**



**(d) Water-accessible surface**

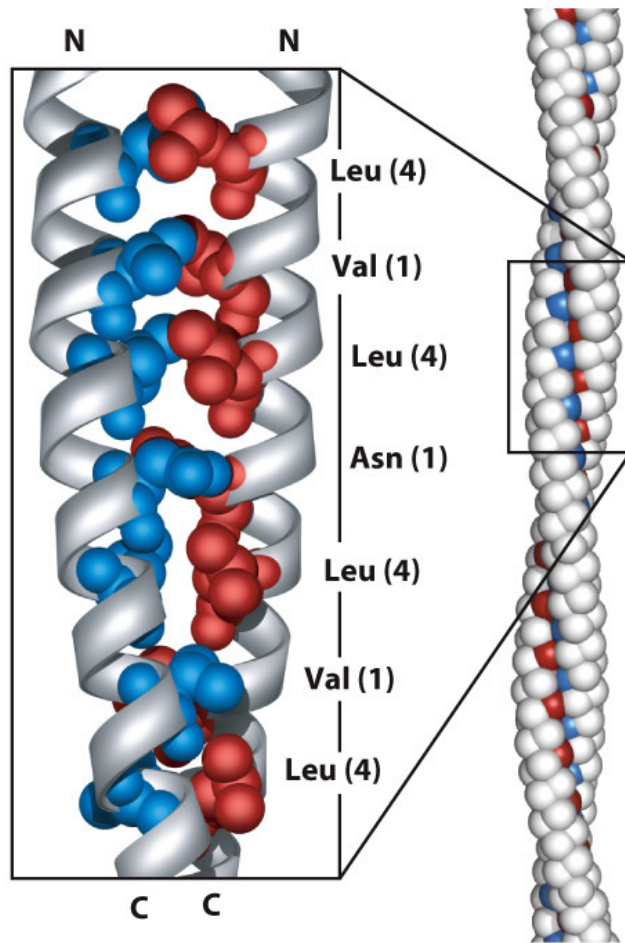


**(e) Hybrid model**

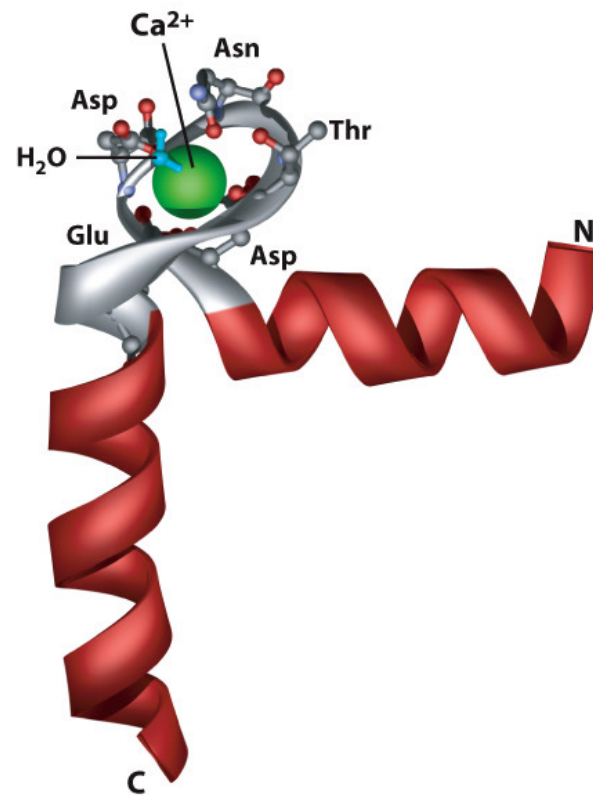


**Figure 3-9**  
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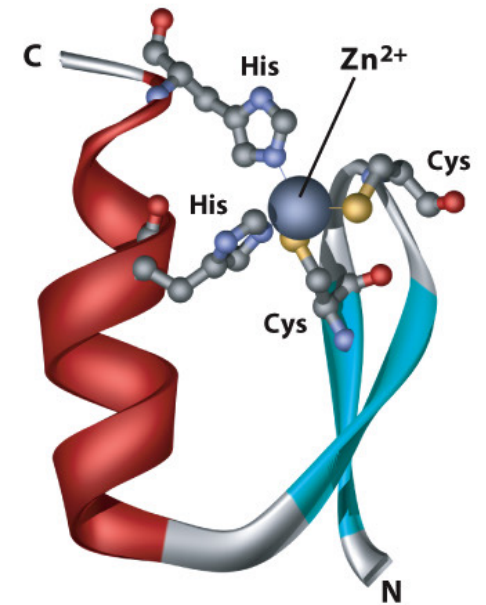
**(a) Coiled-coil motif**



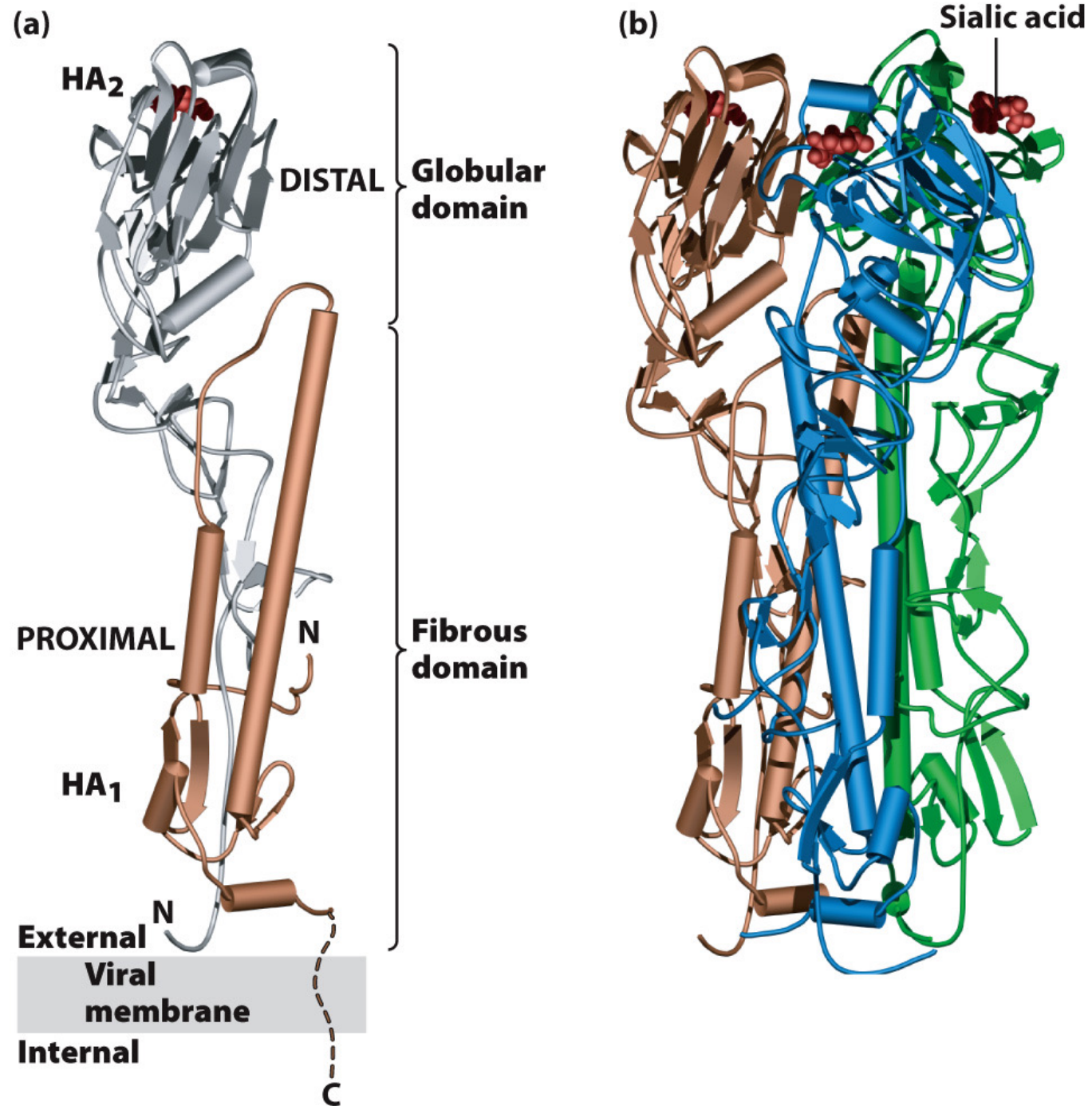
**(b) EFhand/helix-loop-helix motif**



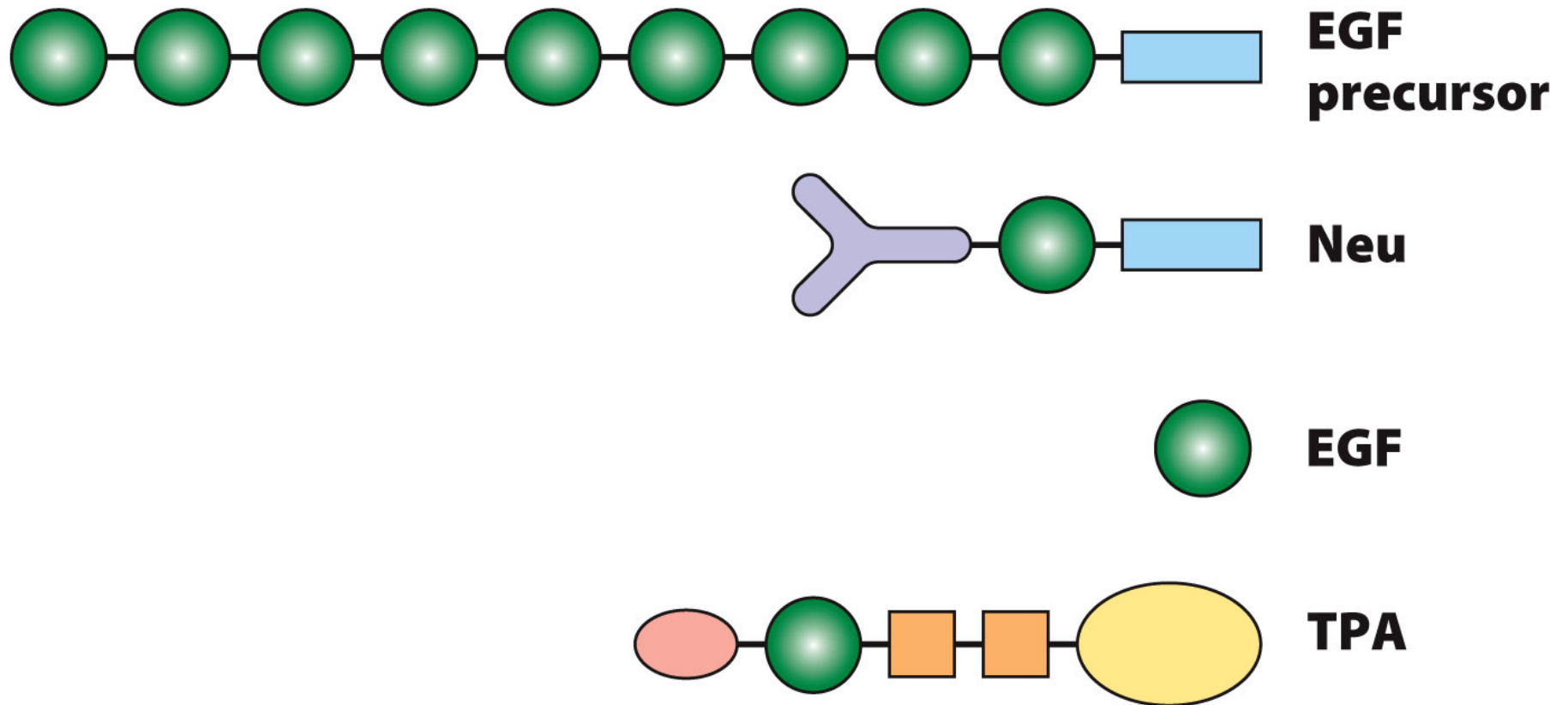
**(c) Zinc-finger motif**



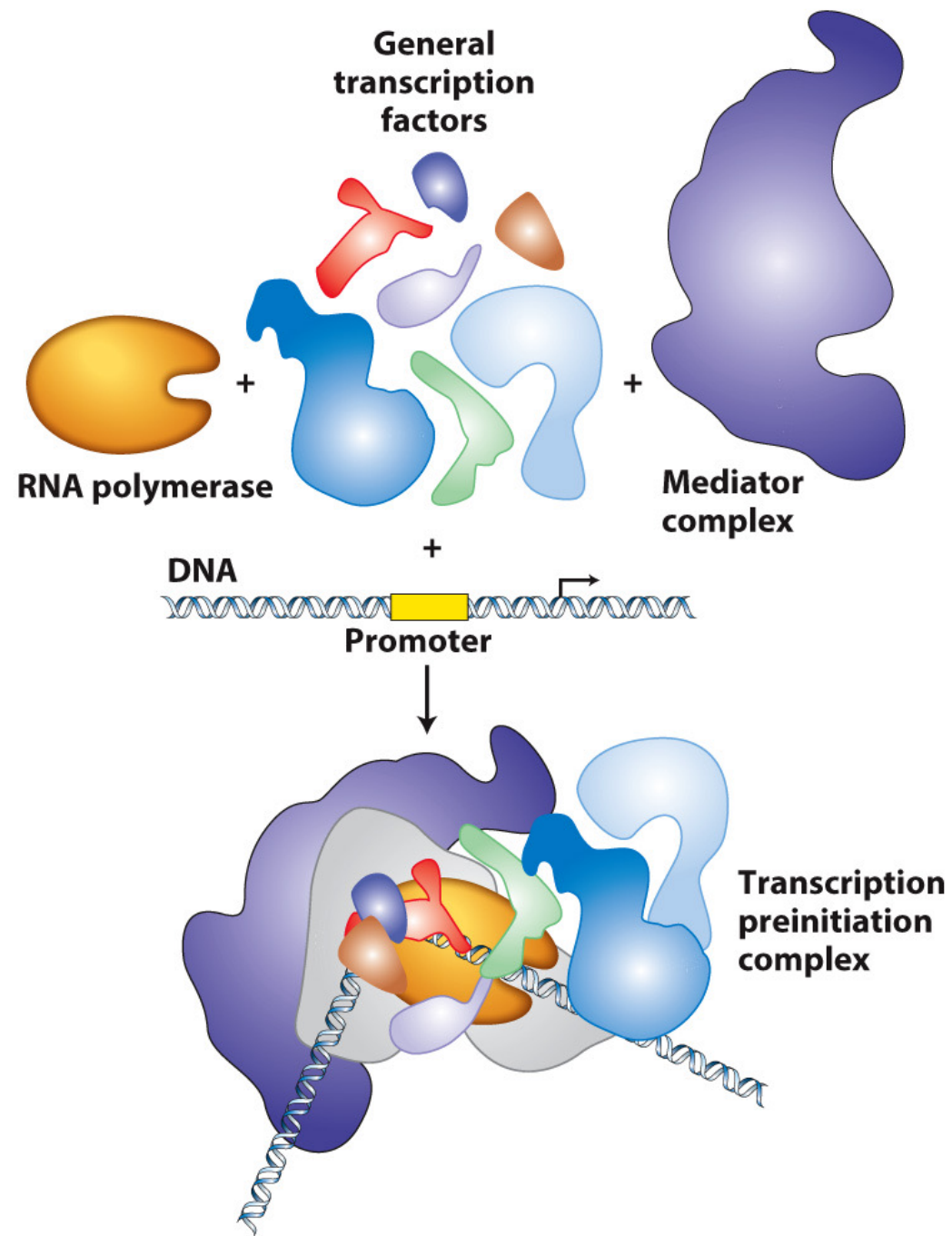
**Figure 3-10**  
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**Figure 3-11**  
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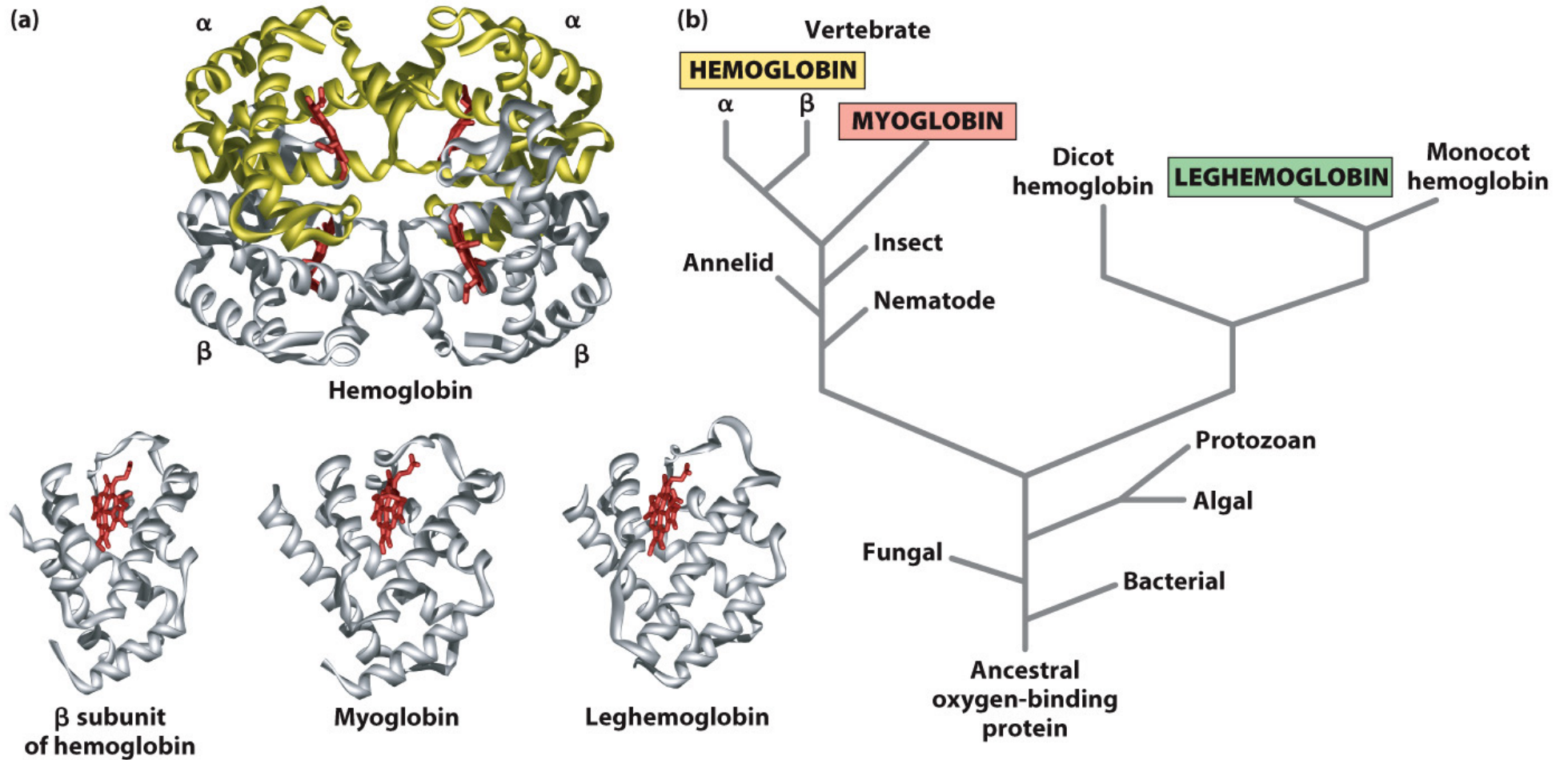


**Figure 3-12**  
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**Figure 3-13**  
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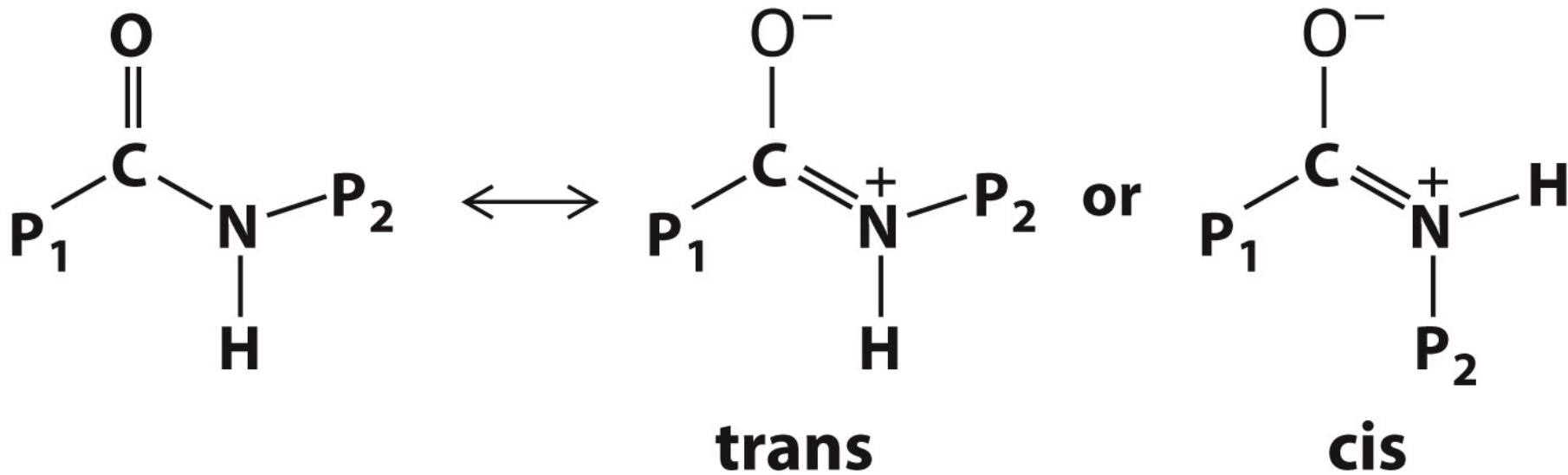


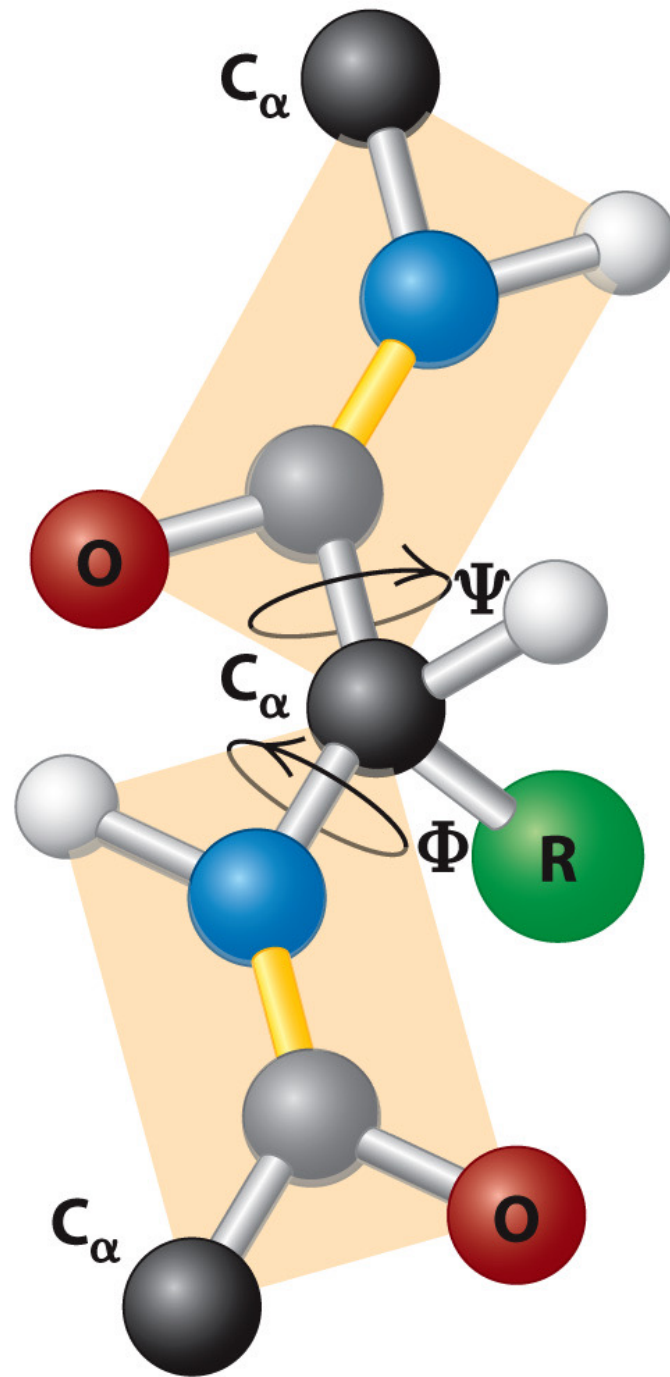


**Figure 3-14**  
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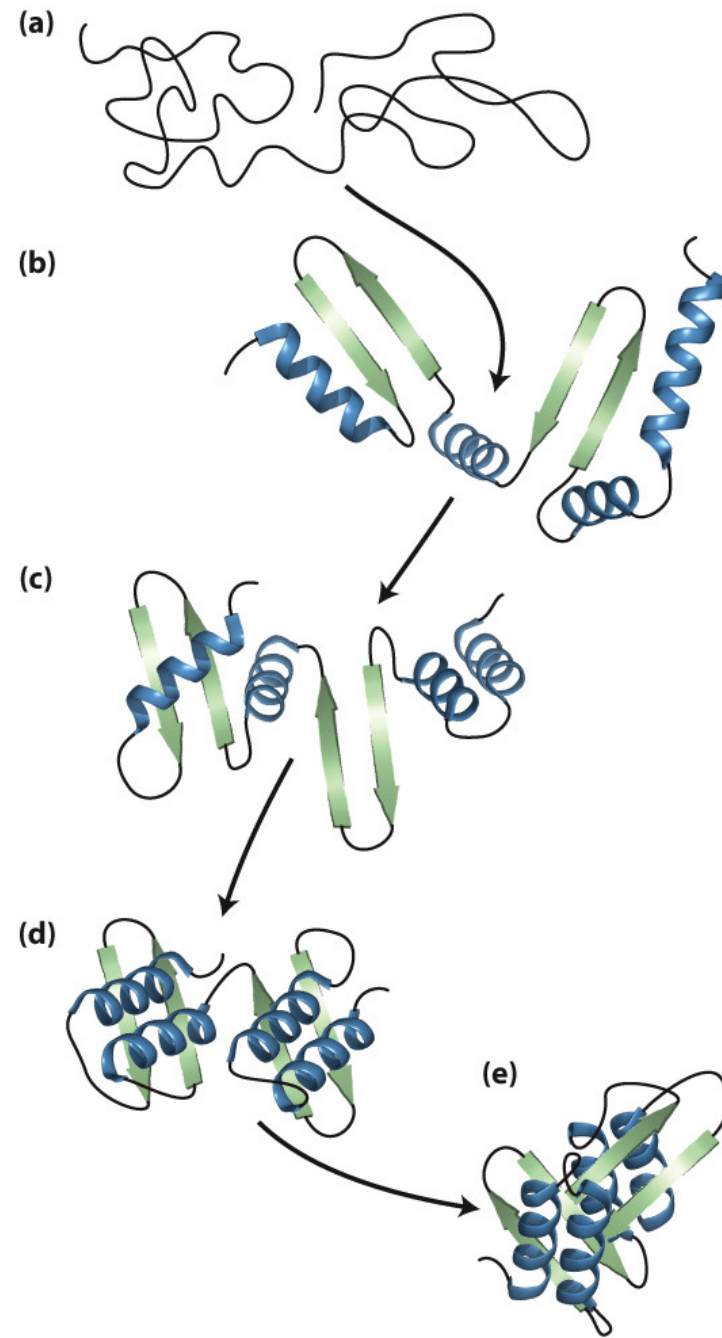
# Protein Structure and Function

- 3.2 Protein Folding
- Protein amino acid sequence determines its 3D structure and function.
- ATP-dependent molecular chaperones and chaperonins assist protein folding in vivo.
- Misfolded/denatured proteins can form well-organized amyloid fibril aggregates that can cause diseases, including Alzheimer's disease and Parkinson's disease.

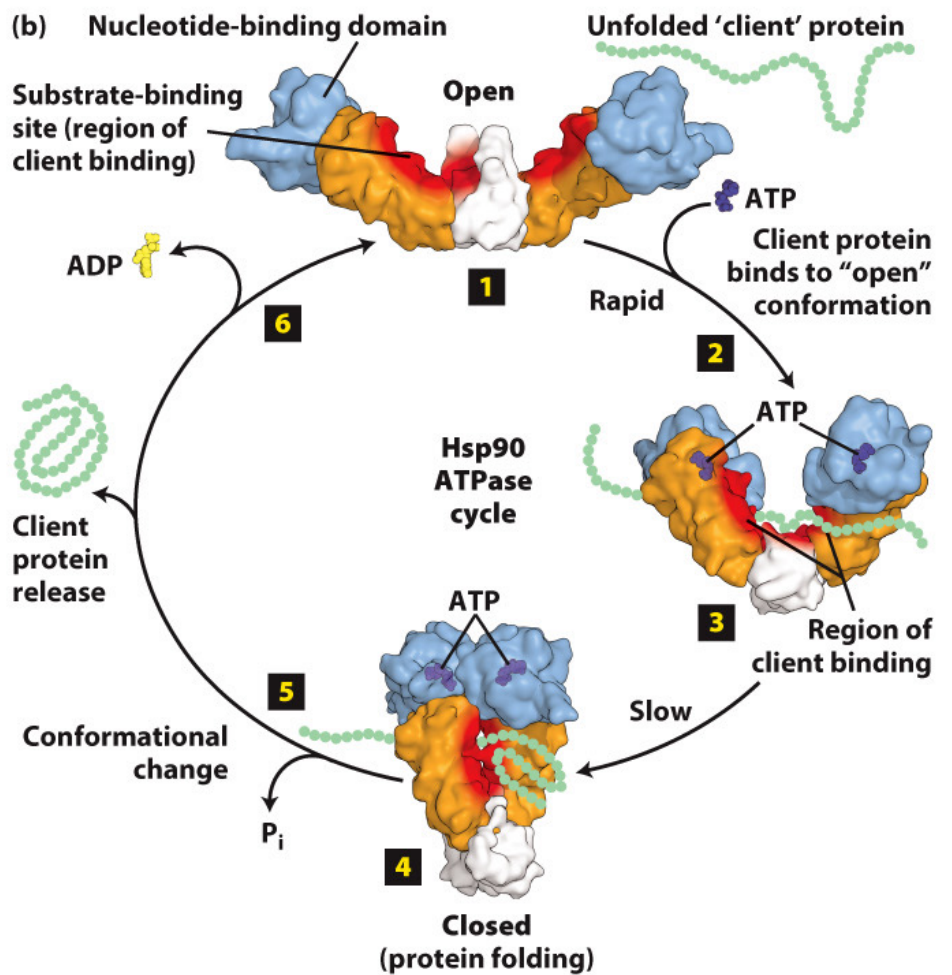
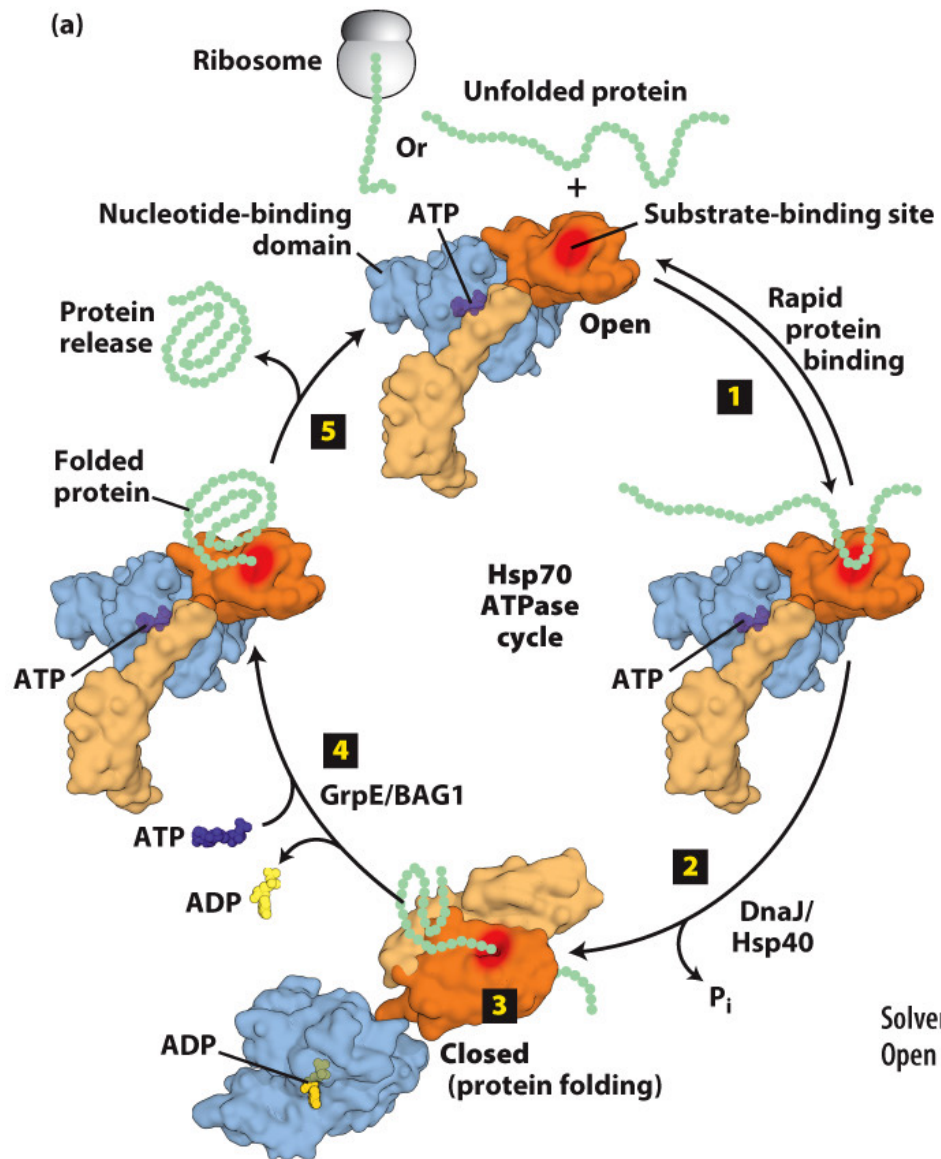




**Figure 3-15**  
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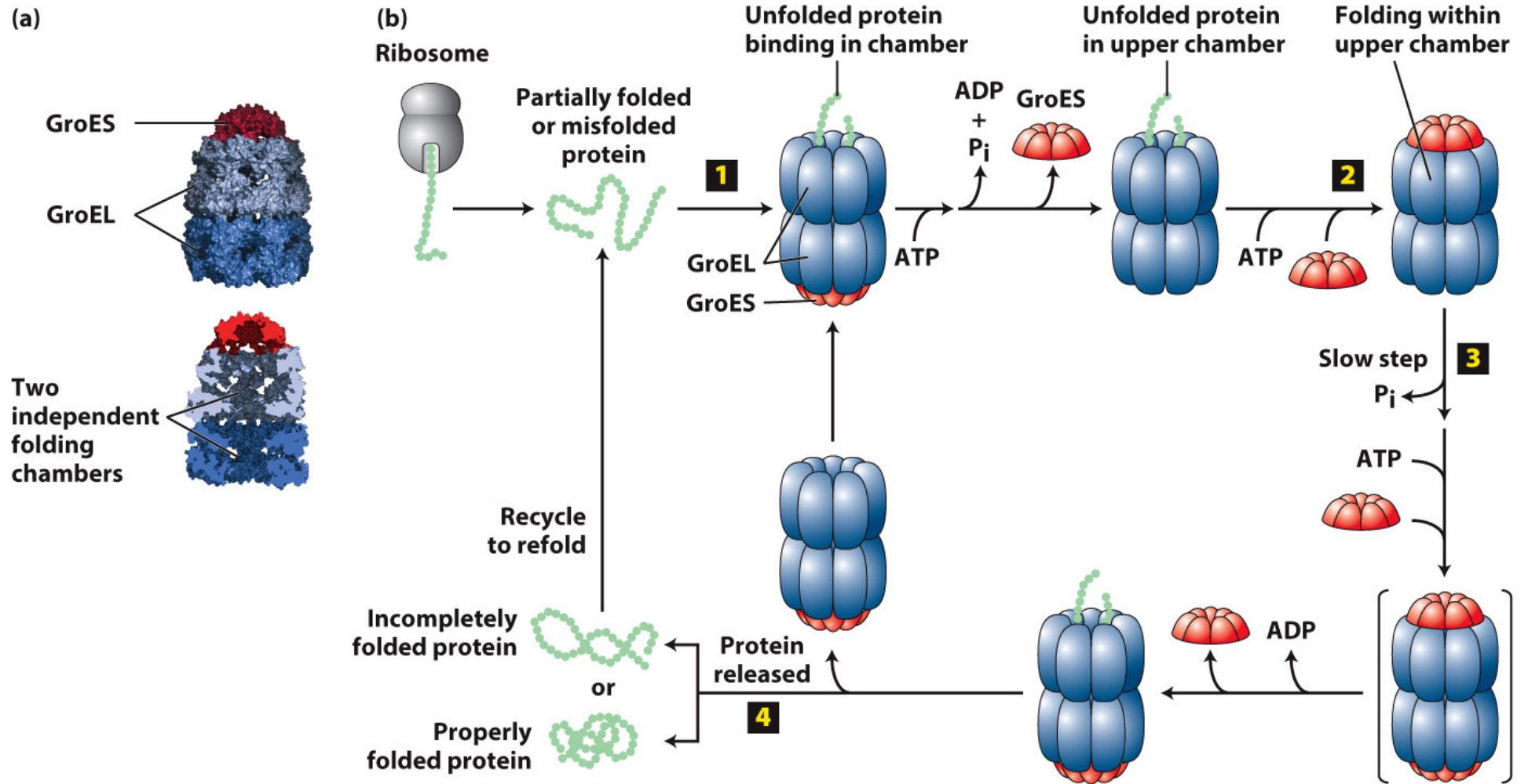


**Figure 3-16**  
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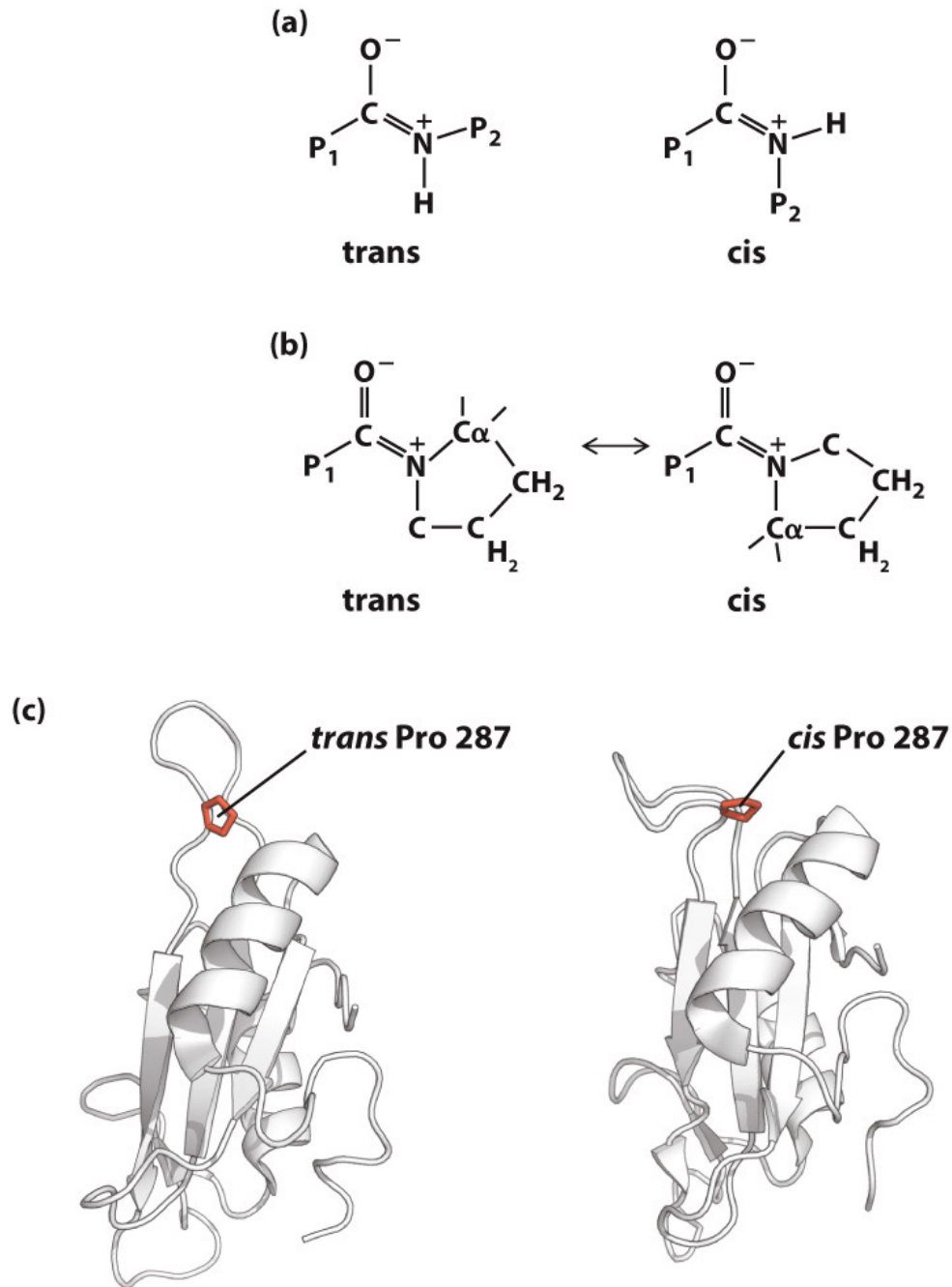


Solvent-accessible surface model of HSP90 courtesy of Elaine Kirschke and David A. Agard, UCSF. Open (ATP) PDB ID 2ior, closed (ATP) PDB ID 2cg9, closed (ADP) based on PDB ID 2cg9

**Figure 3-17**  
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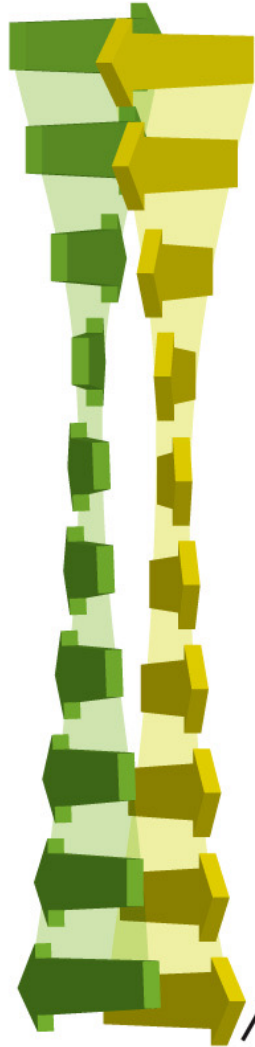
**Figure 3-18**  
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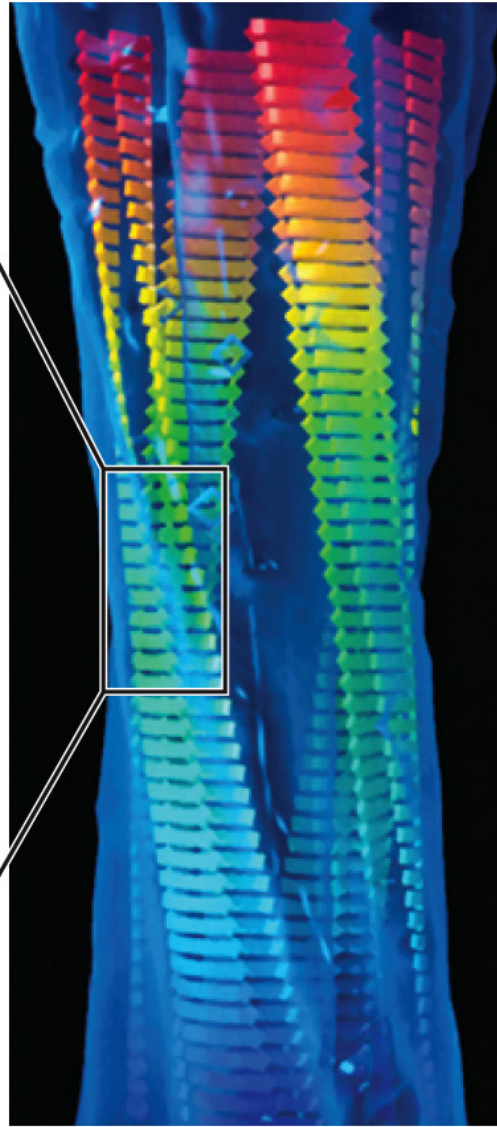
**Figure 3-19**  
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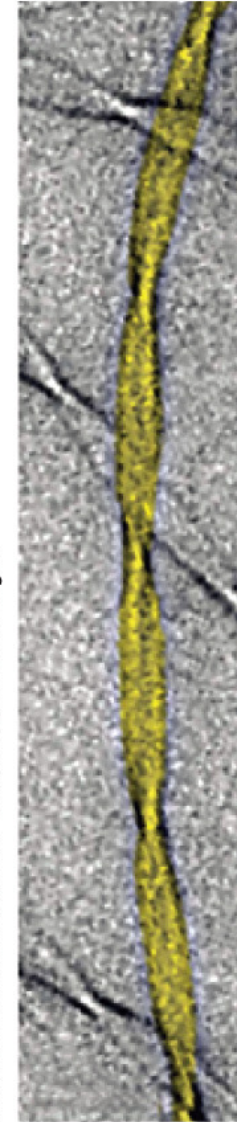
(a) Amyloid protofilament



(b) Amyloid fibrils



Republished with permission of Elsevier, from Dobson, C.M., "Protein misfolding, evolution and disease," *Trends in Biochemical Science* 1999, 24(9):329-332. Fig. 3



Reprinted by permission from Macmillan Publishers Ltd: from Knowles et al., *Nat. Rev. Mol. Cell Biol.* 2014, 15(6):384-396. Fig. 3a

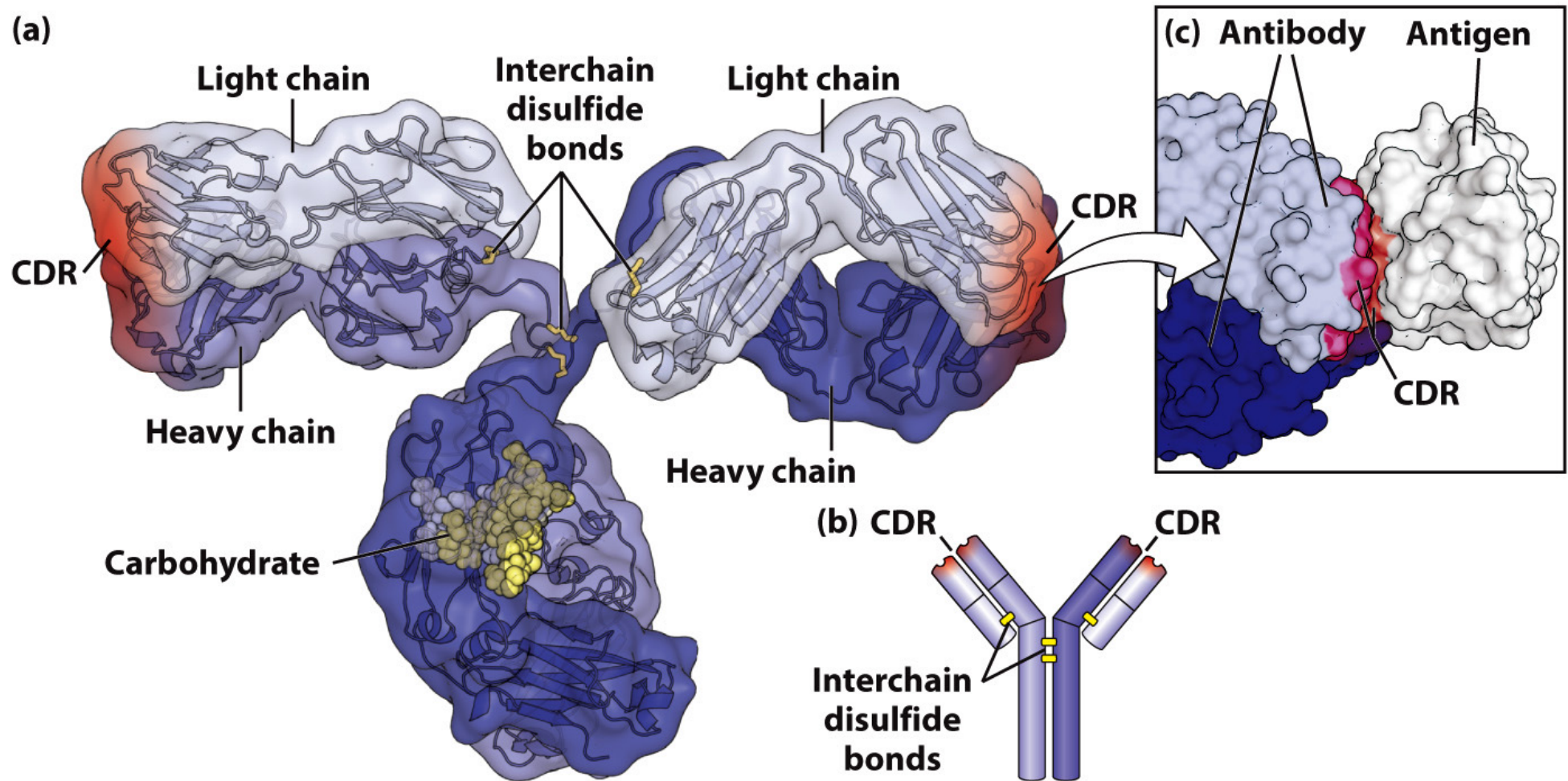
50 nm

**Figure 3-20ab**  
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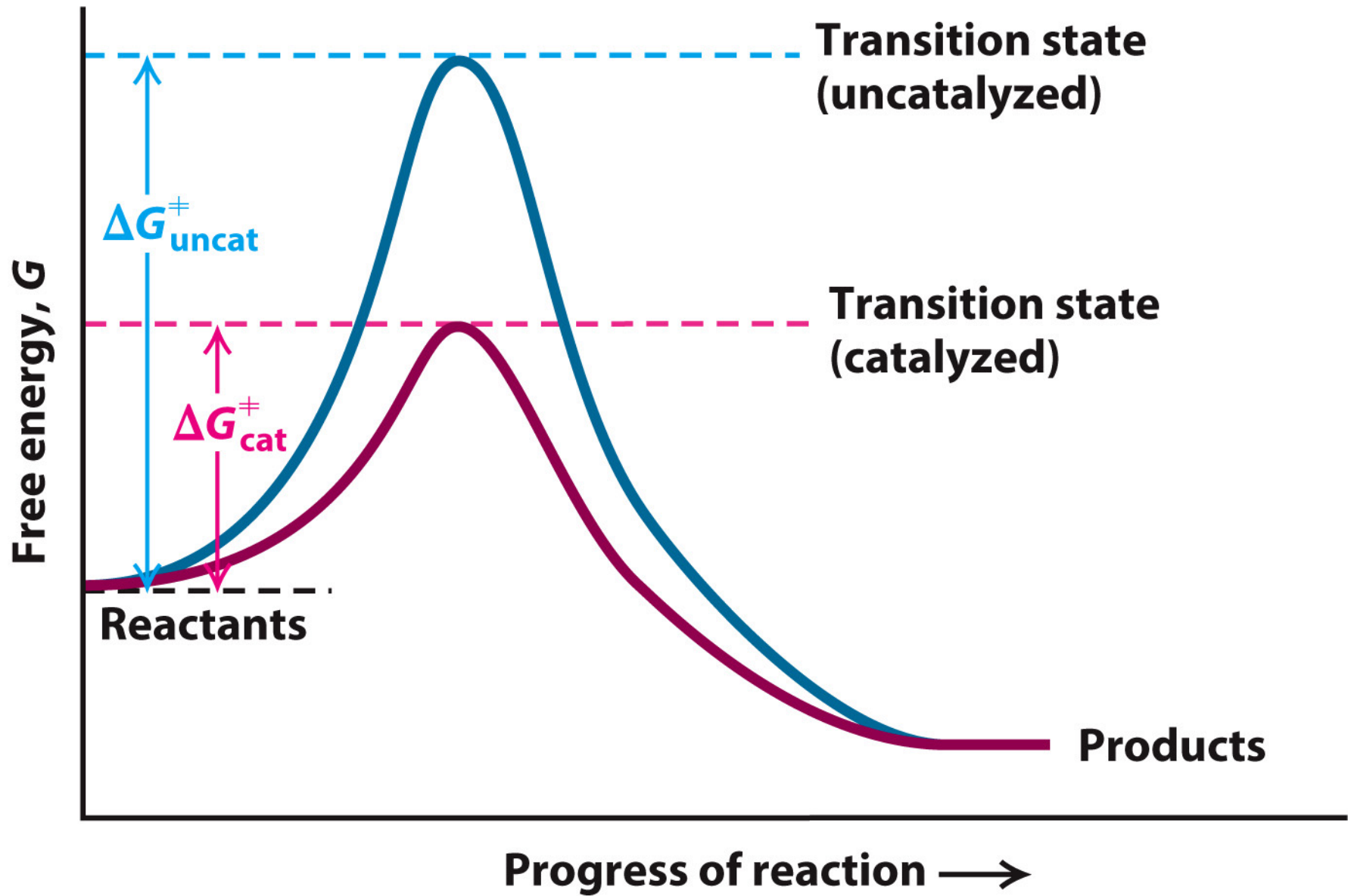
# Protein Structure and Function

## 3.3 Protein Binding and Enzyme Catalysis

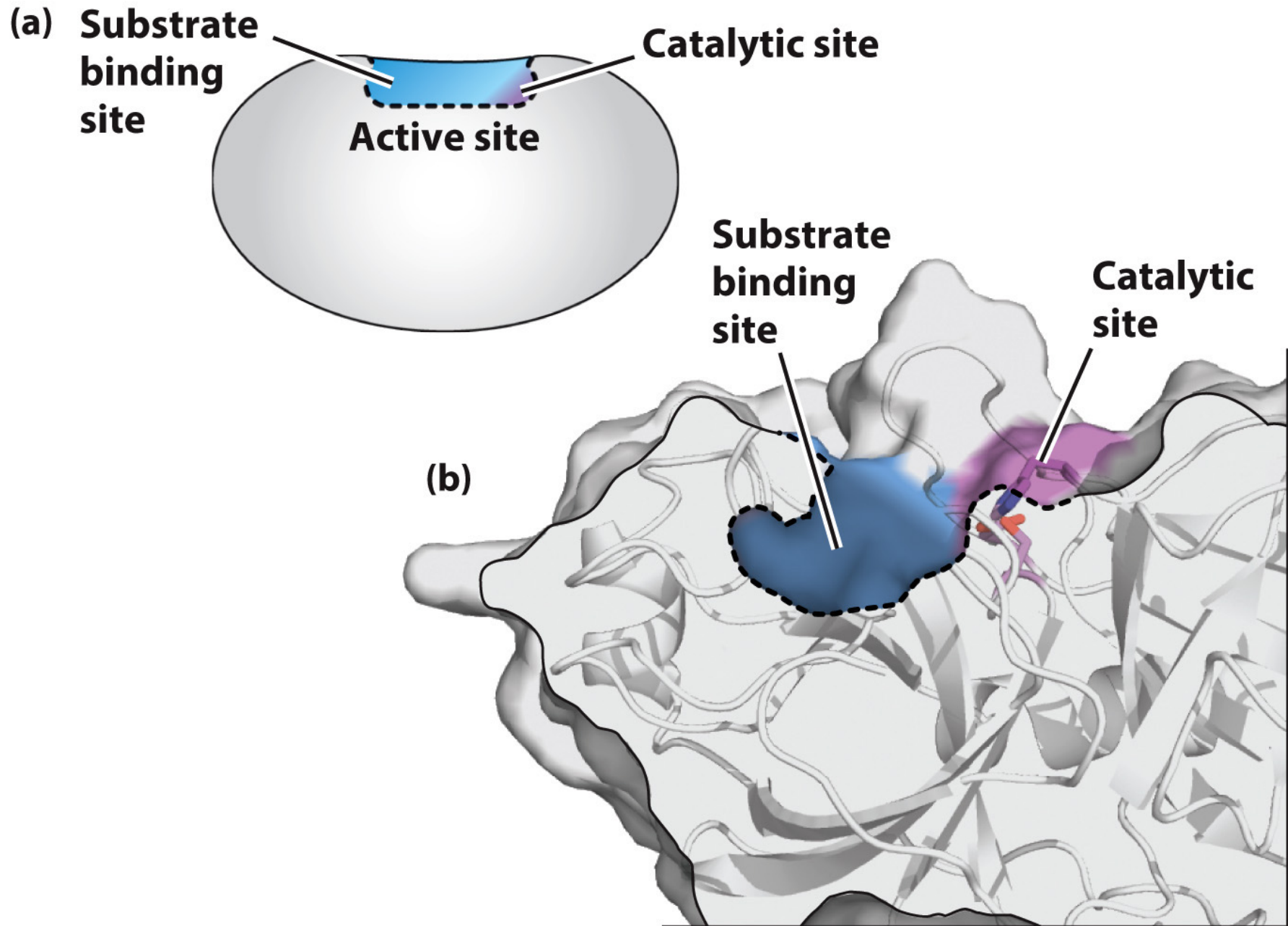
- Protein function depends on binding other molecules (ligands).
- Enzymes accelerate rates of cellular reactions by lowering activation energy and stabilizing transition-state intermediates.
- Enzymes often use acid-base catalysis mediated by one or more amino acid side chains.
- Metabolic pathway enzymes may be associated as domains of a monomeric protein, subunits of a multimeric protein, or components of a protein complex assembled on a common scaffold.



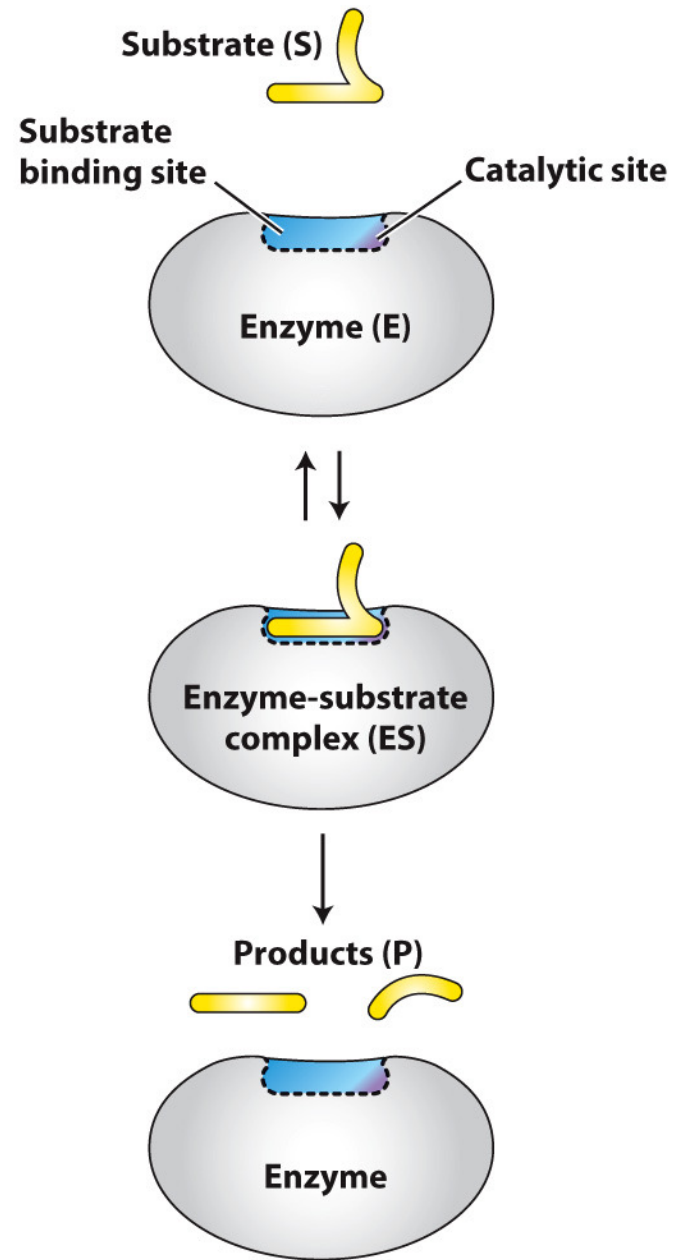
**Figure 3-21**  
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**Figure 3-22**  
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**Figure 3-23**  
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**Figure 3-25**  
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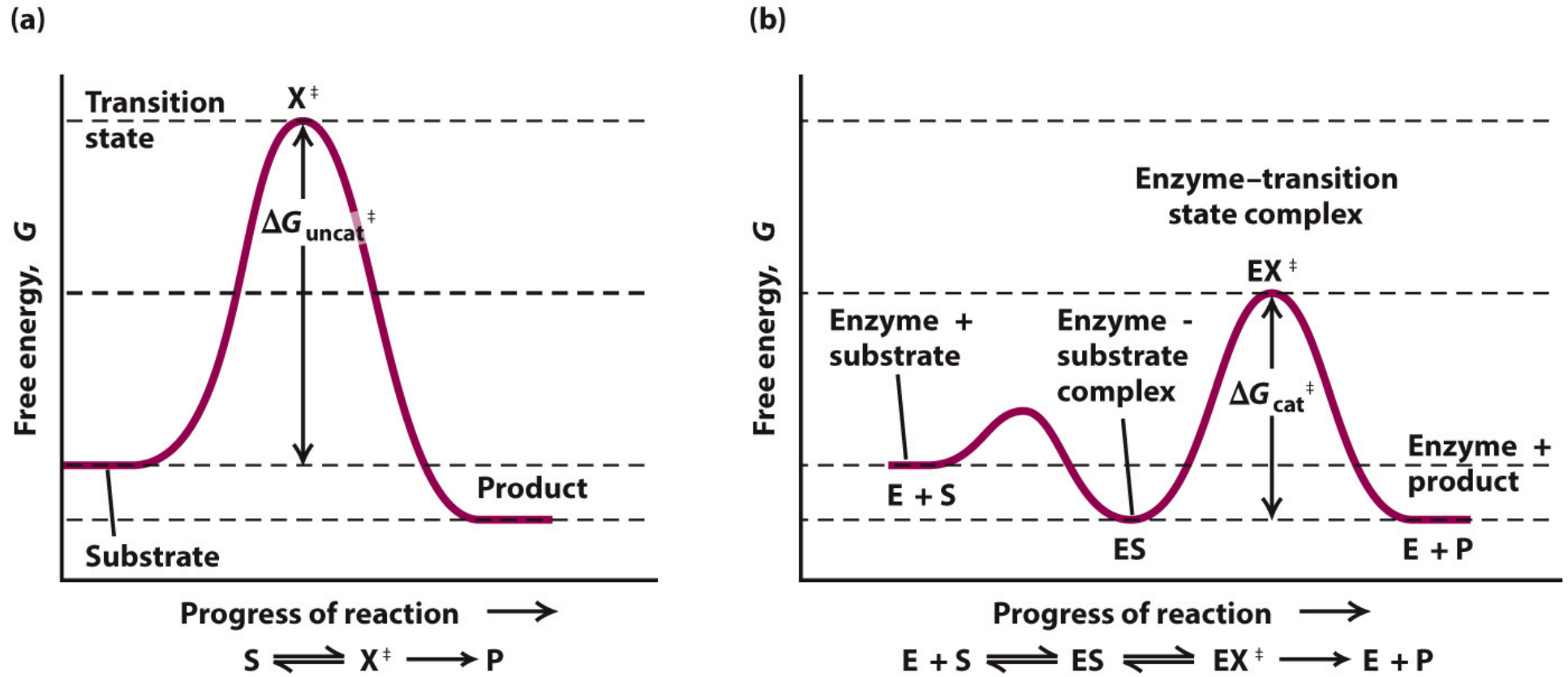
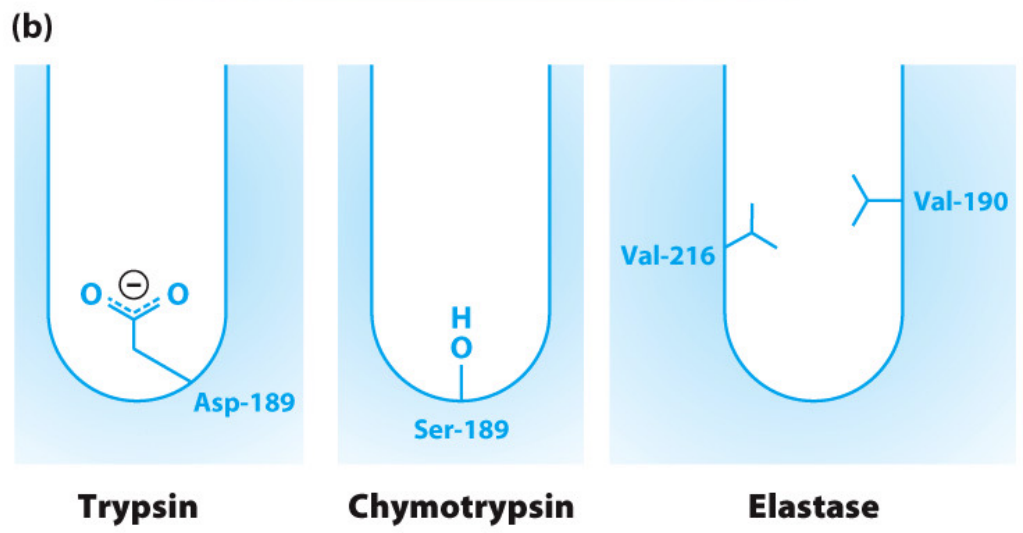
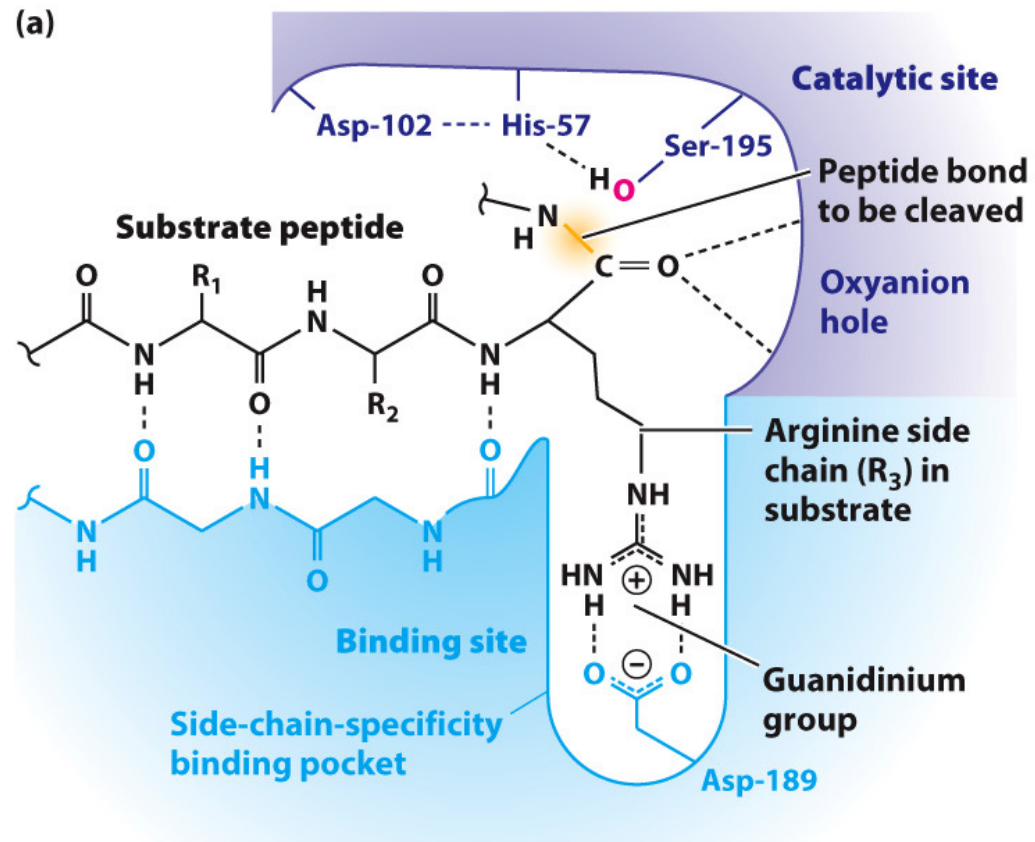
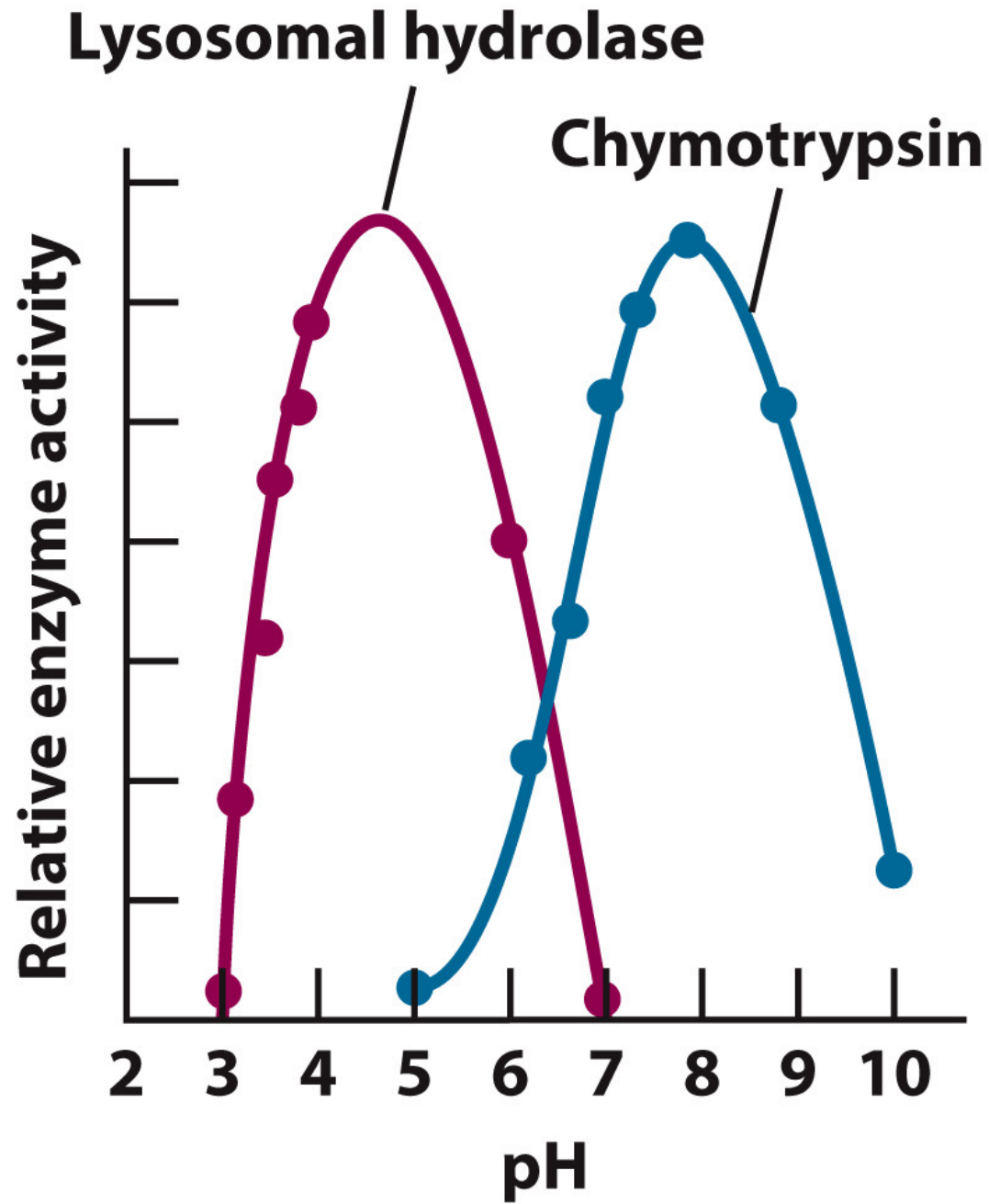


Figure 3-26  
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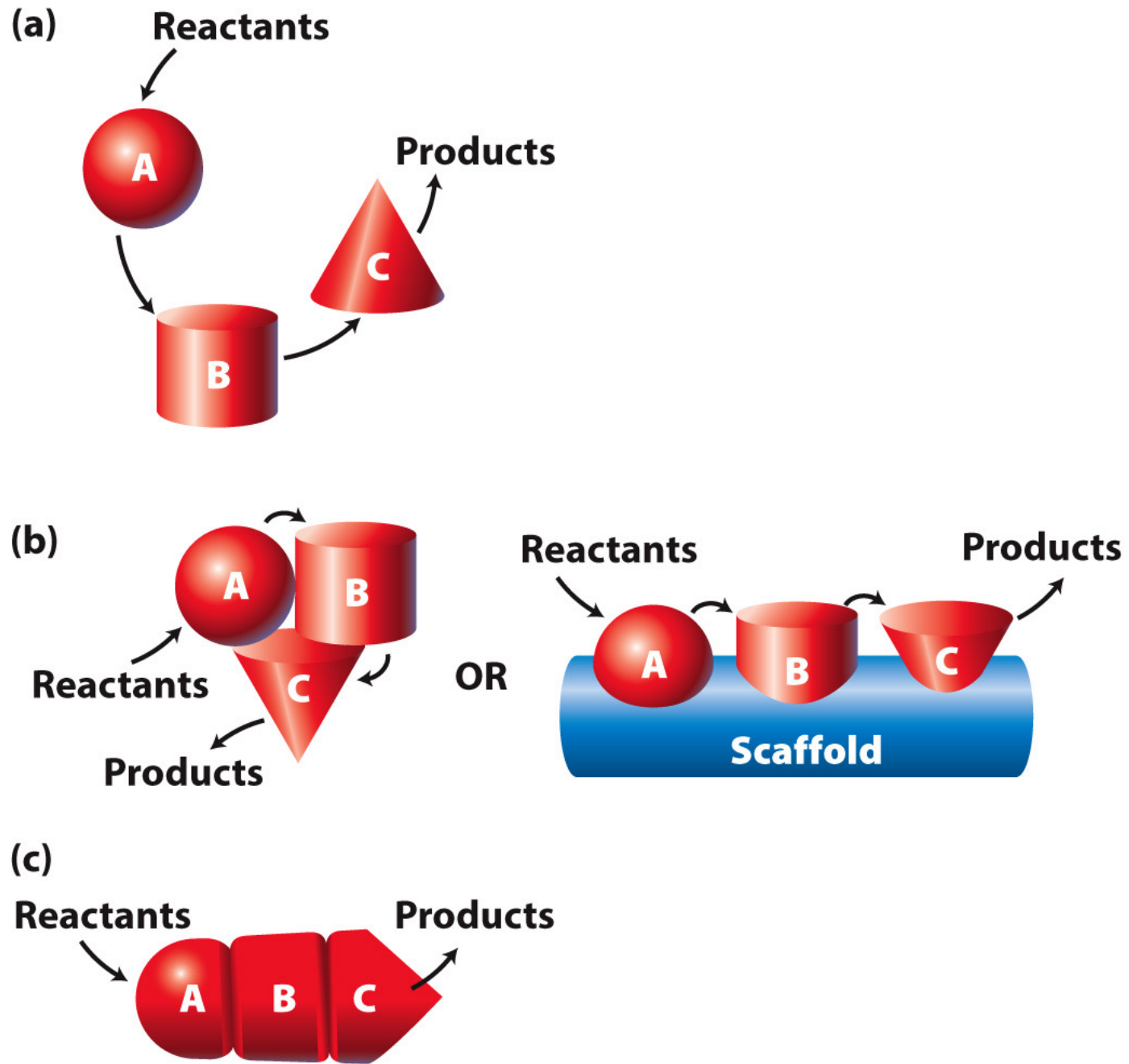


**Figure 3-27**  
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**Figure 3-29**  
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**Figure 3-30**  
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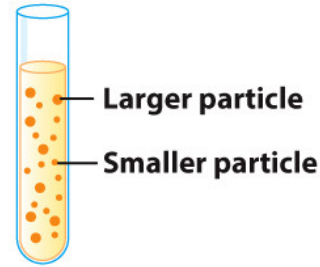
# Protein Structure and Function

## 3.5 Purifying, Detecting, and Characterizing Proteins

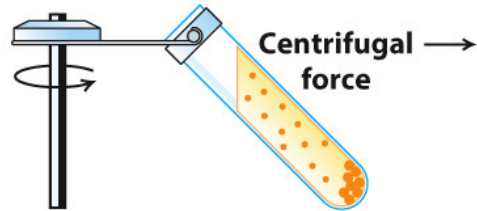
- Proteins can be isolated from other cell components on the basis of a variety of physical and chemical properties.
- Proteins can be detected and quantified by various assays and specific antibody recognition.
- Tagging with various types of markers can be used to investigate protein synthesis, location, processing, and stability.
- X-ray crystallography, cryoelectron microscopy, and NMR spectroscopy reveal 3D structures of proteins.

**(a) Differential centrifugation**

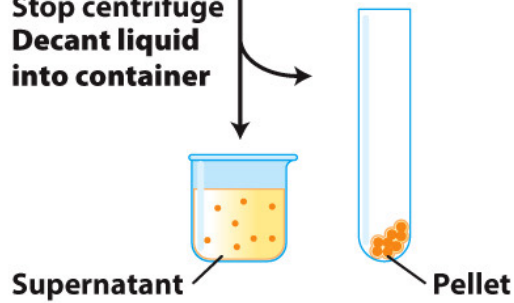
**1** Sample is poured into tube



**2** Centrifuge  
Particles settle  
according to  
mass

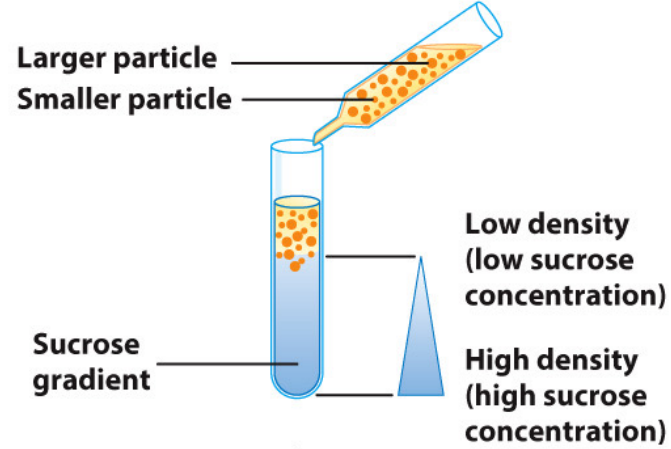


**3** Stop centrifuge  
Decant liquid  
into container

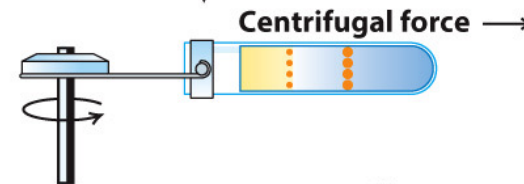


**(b) Rate-zonal centrifugation**

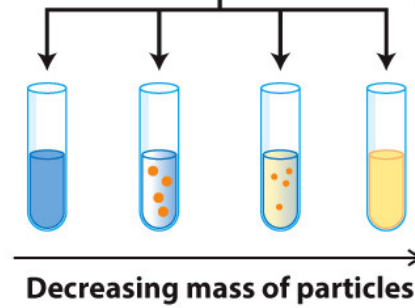
**1** Sample is layered on top of density gradient



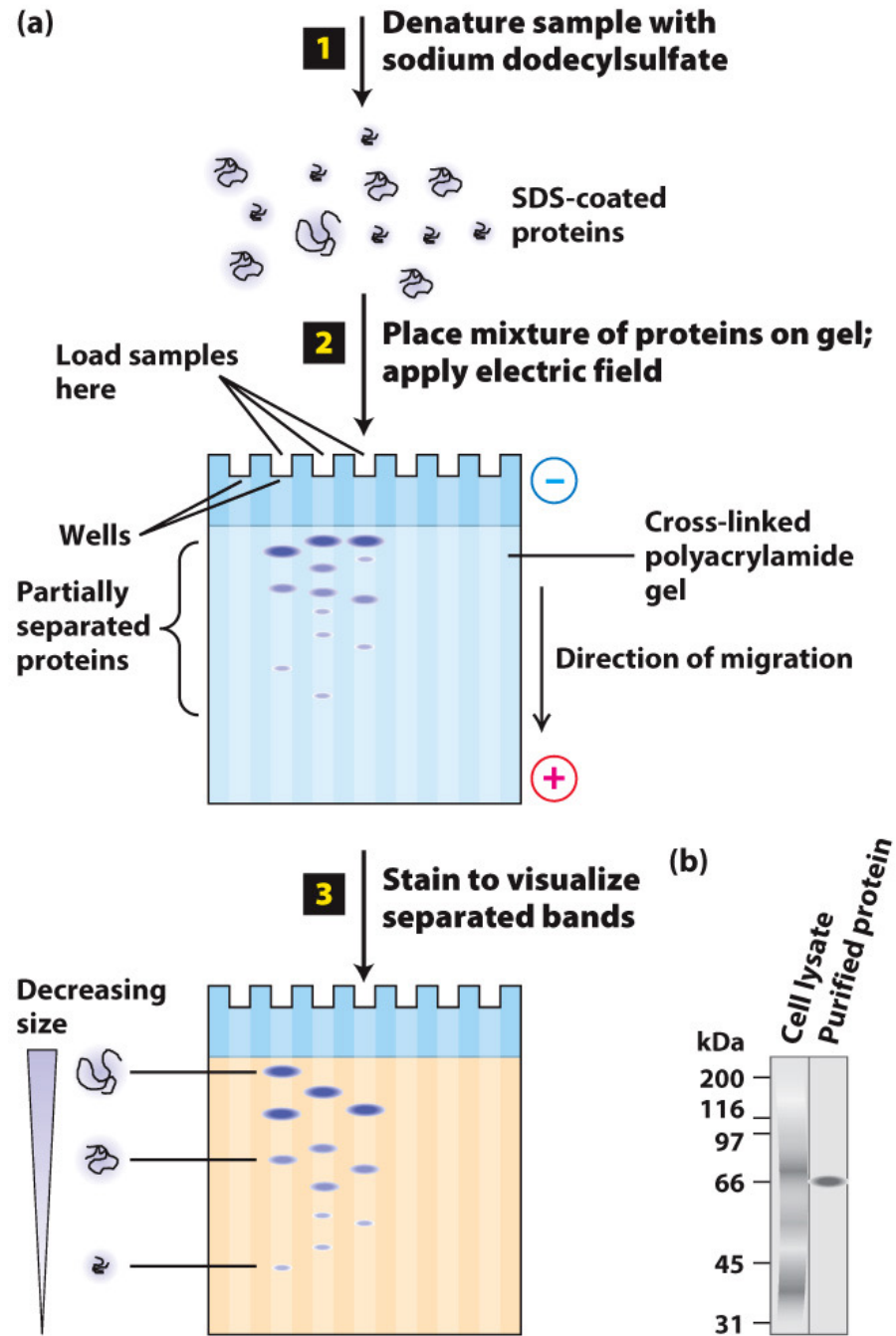
**2** Centrifuge  
Particles settle  
according to  
mass



**3** Stop centrifuge  
Collect fractions  
and do assay



**Figure 3-37**  
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**Figure 3-38**  
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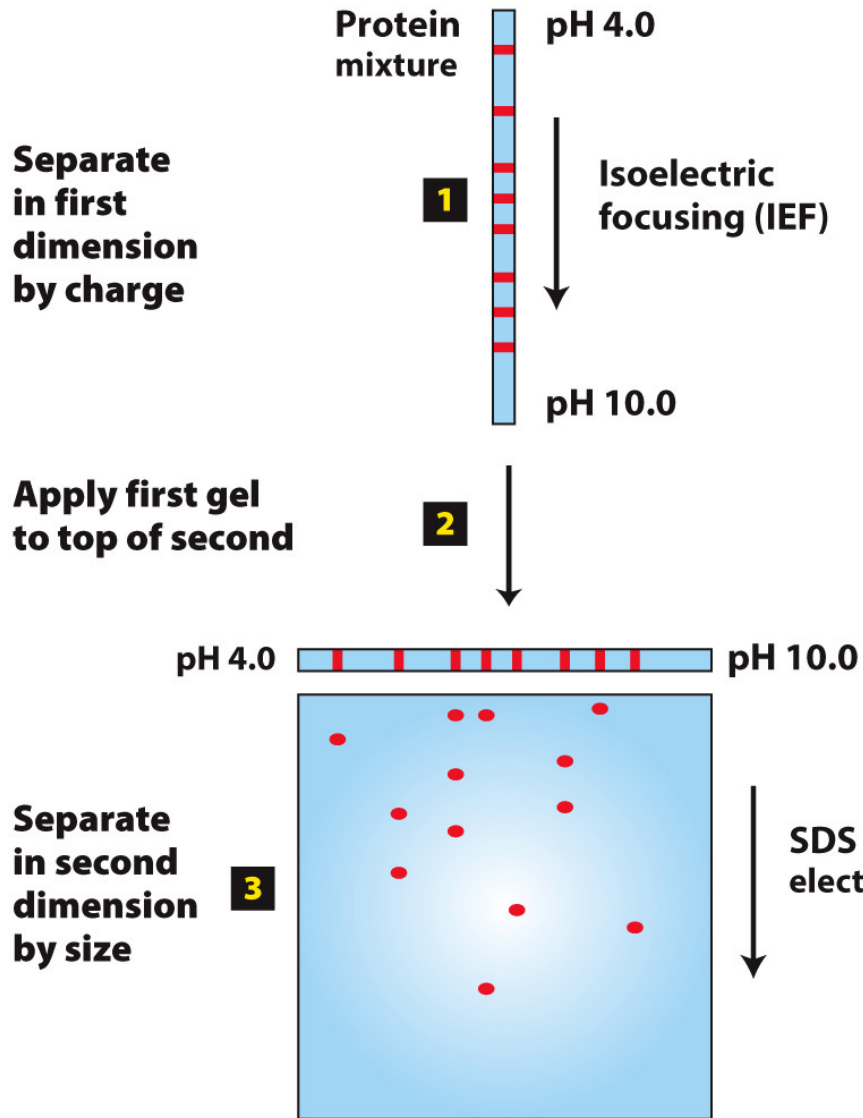


Figure 3-39a  
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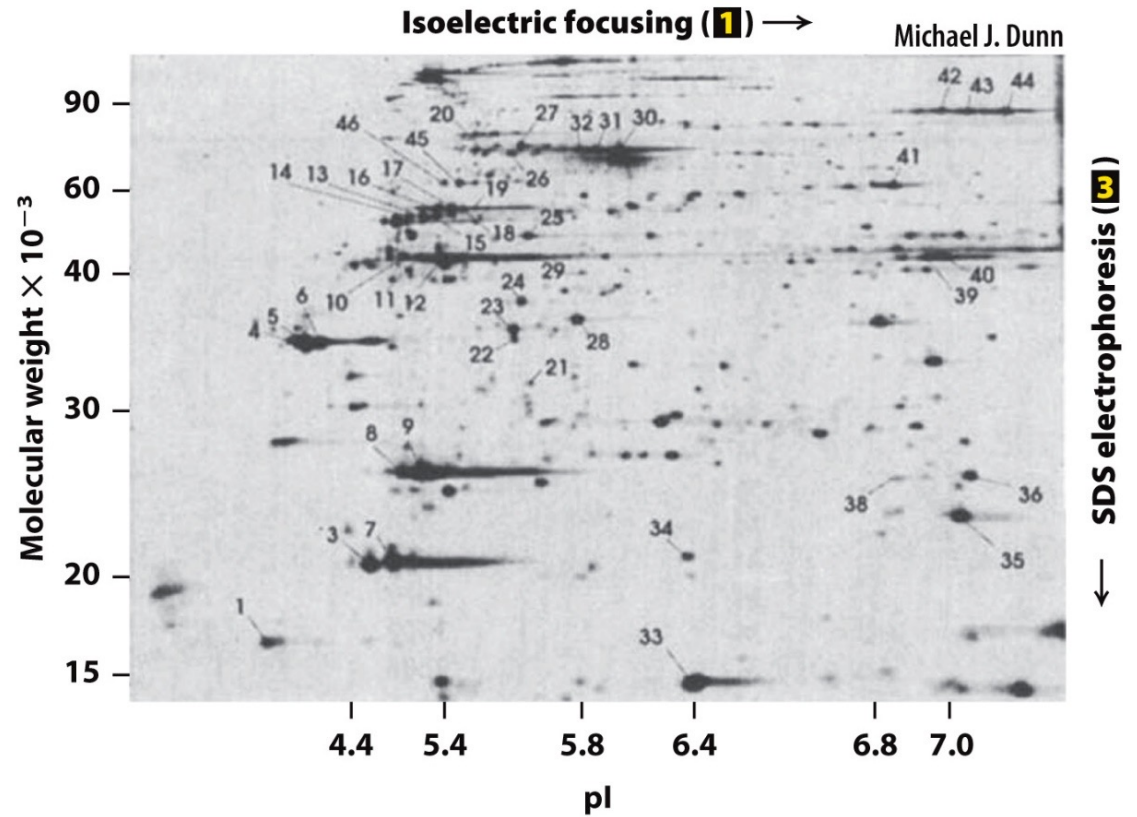
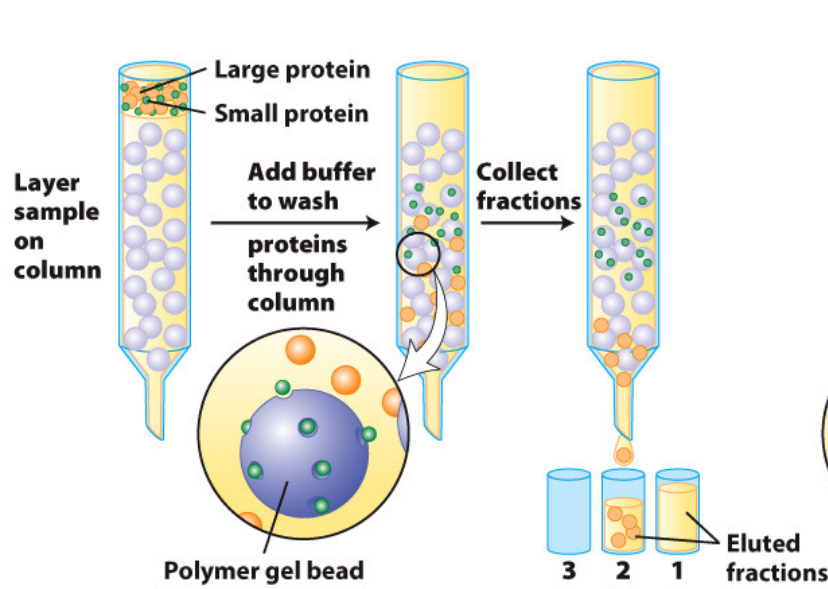
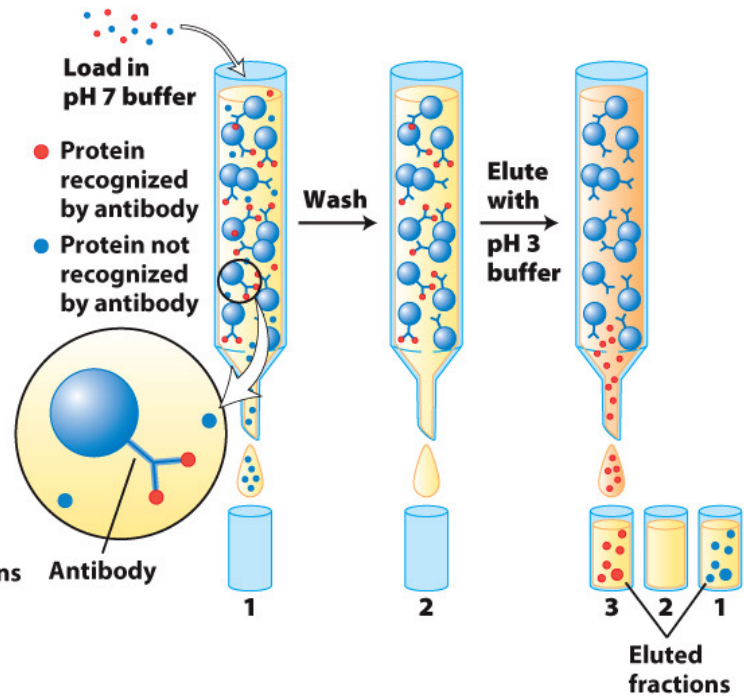


Figure 3-39b  
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(a) Gel filtration chromatography



(c) Antibody-affinity chromatography



(b) Ion-exchange chromatography

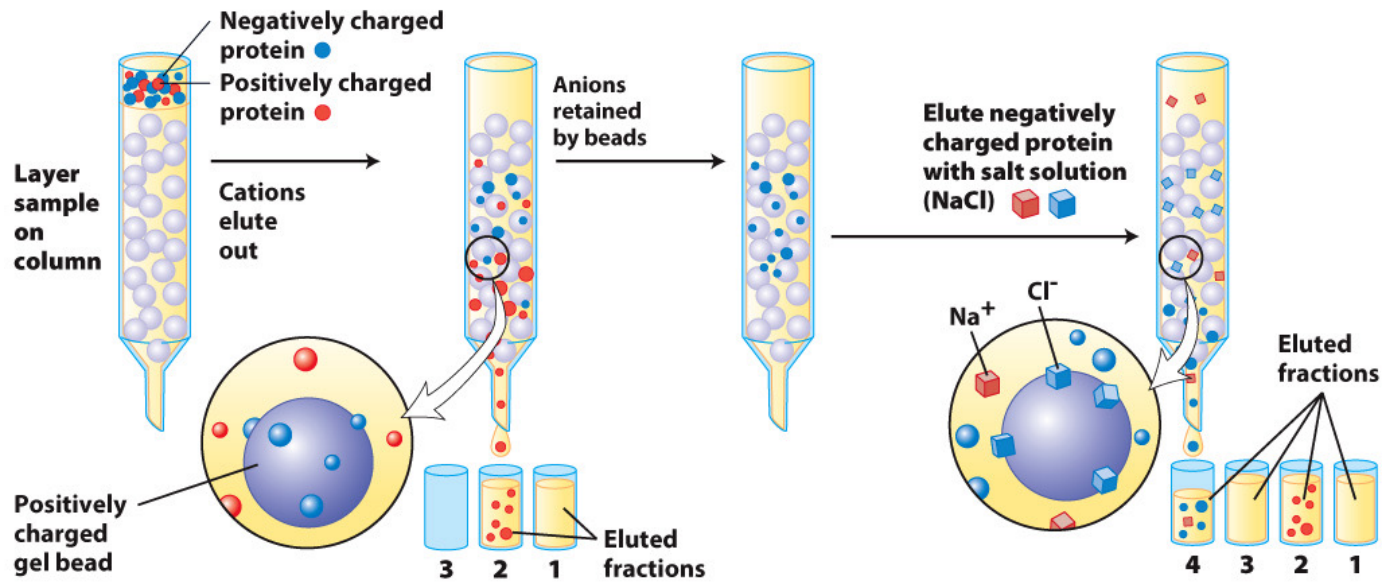
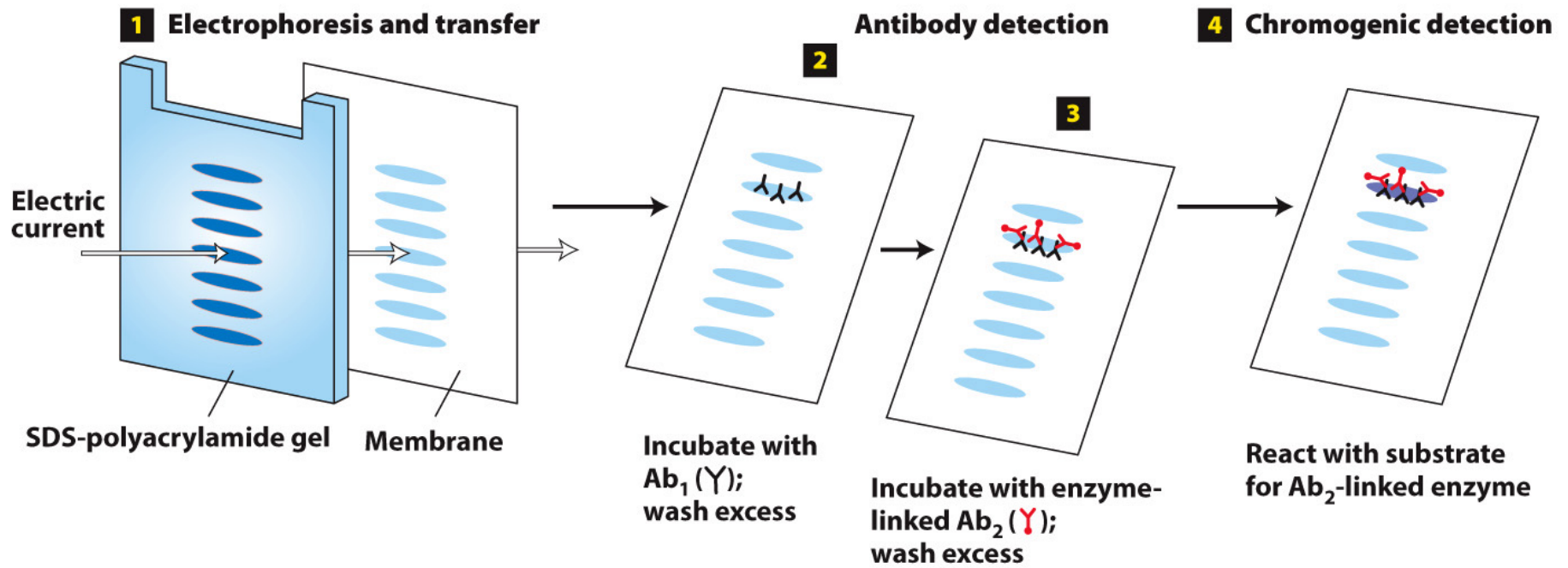


Figure 3-40

## General method of immunoblotting



**Figure 3-41a**  
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**TABLE 3-1 Radioisotopes Commonly Used in Biological Research**

<b>Isotope</b>	<b>Half-Life</b>
<b>Phosphorus-32</b>	<b>14.3 days</b>
<b>Iodine-125</b>	<b>60.4 days</b>
<b>Sulfur-35</b>	<b>87.5 days</b>
<b>Tritium (hydrogen-3)</b>	<b>12.4 years</b>
<b>Carbon-14</b>	<b>5730.4 years</b>

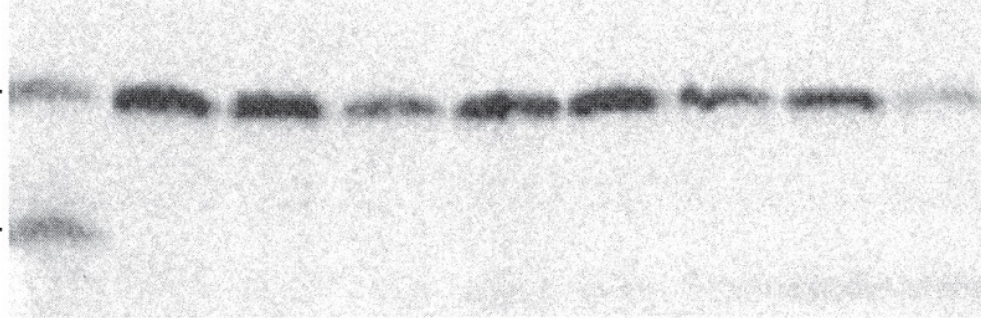
**Table 3-1**  
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(a)

Pulse (h)	0.5								
Chase (h)	0	.5	1	2	4	6	8	12	24

Normal protein

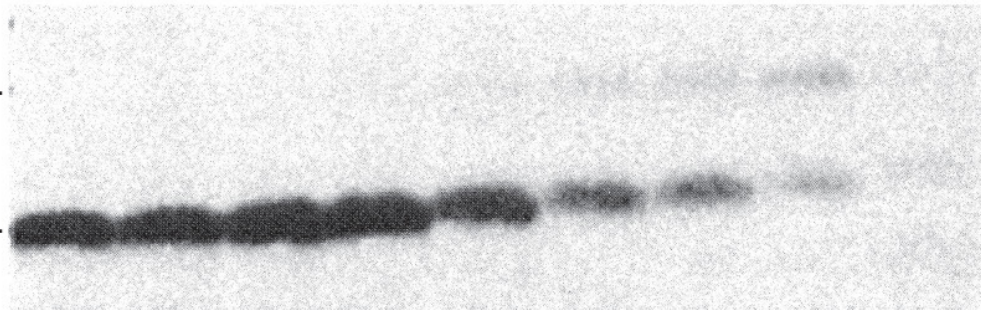
*m* —  
*p* —



(b)

Mutant protein

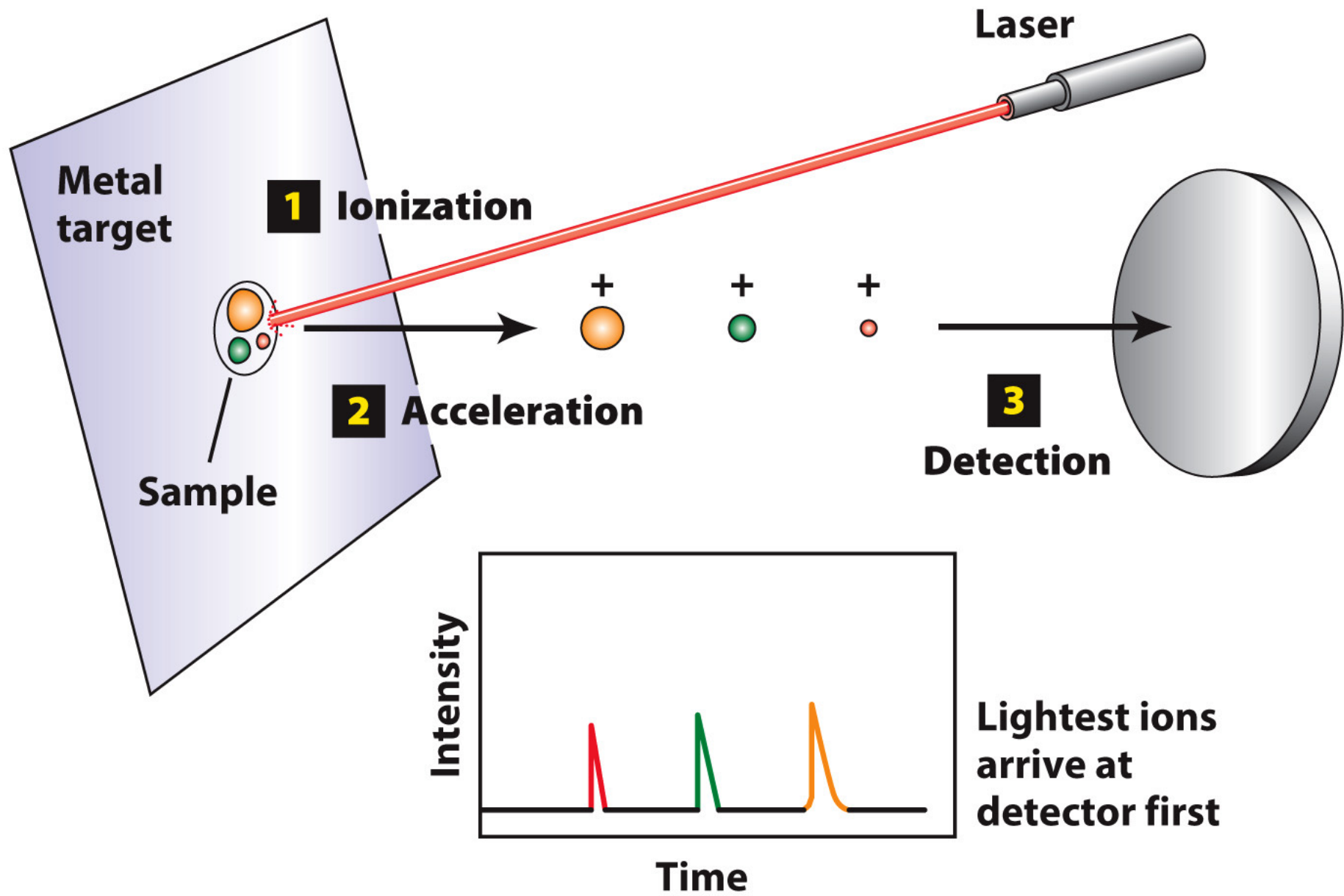
*m* —  
*p* —



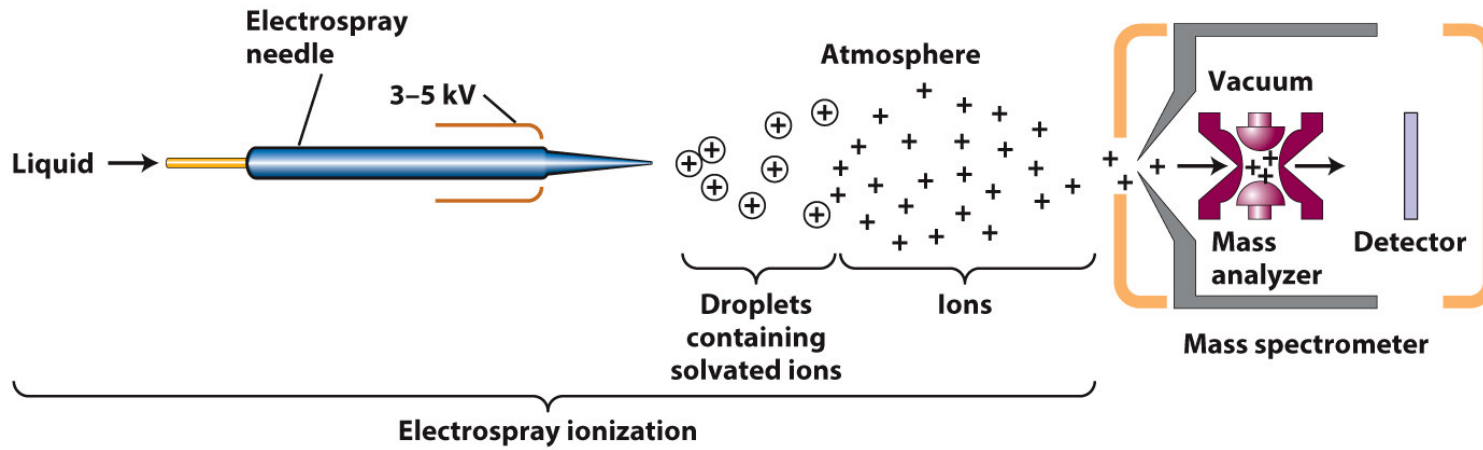
**Precursor protein (*p*) is converted to mature protein (*m*) by post-translational carbohydrate addition**

© Kozarsky et al., *The Journal of Cell Biology*. 102: 1567–1575. doi:10.1083/jcb.102.5.1567

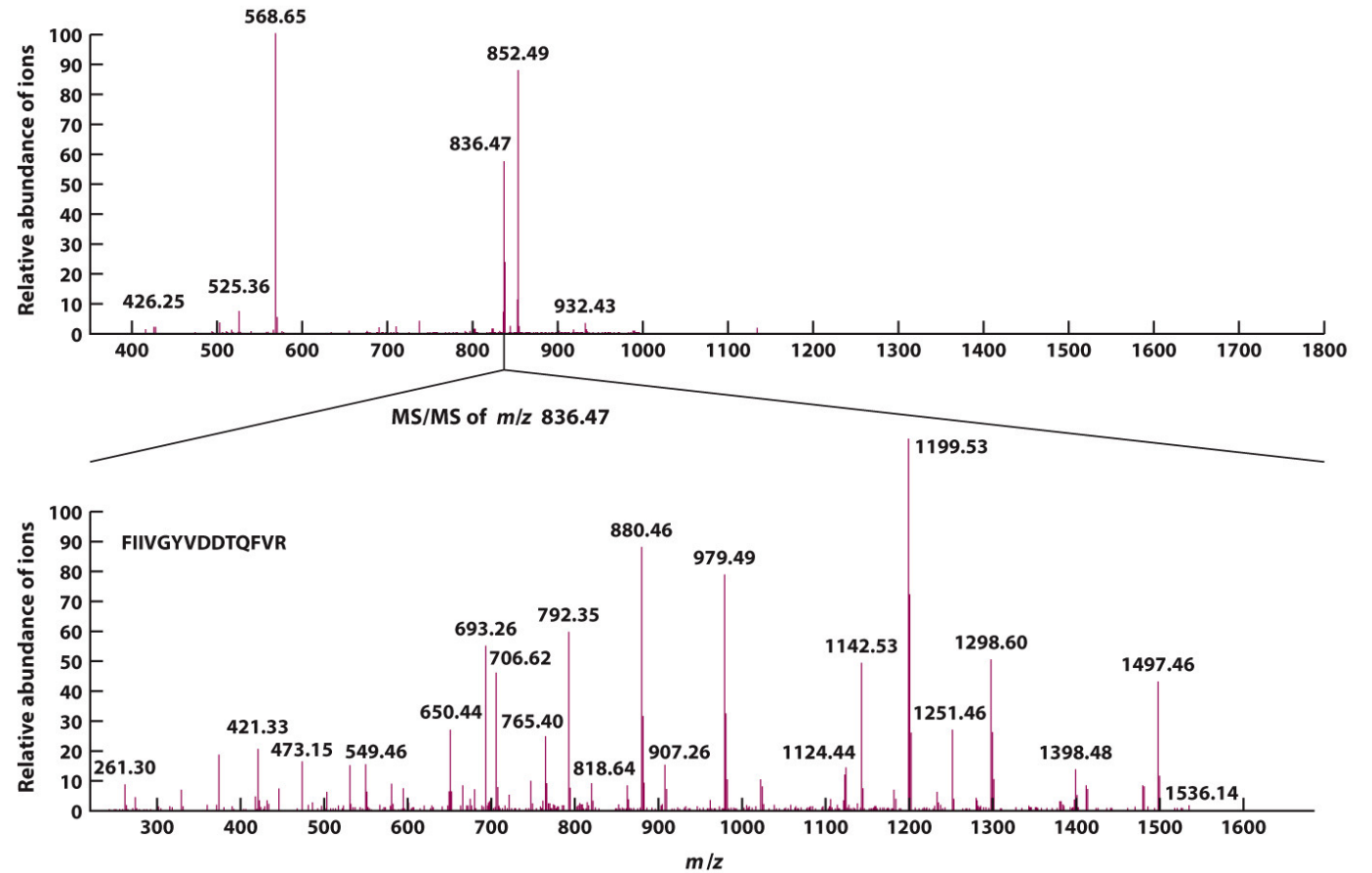
Figure 3-42  
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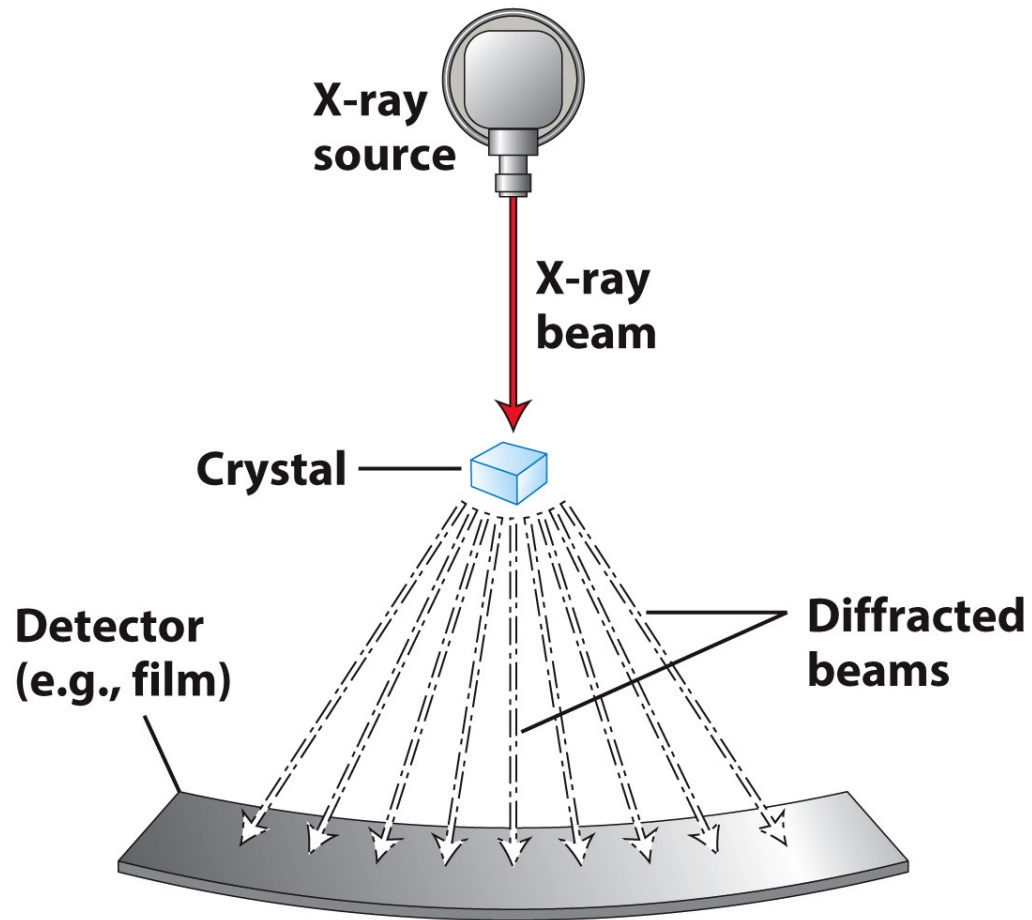
**Figure 3-43**  
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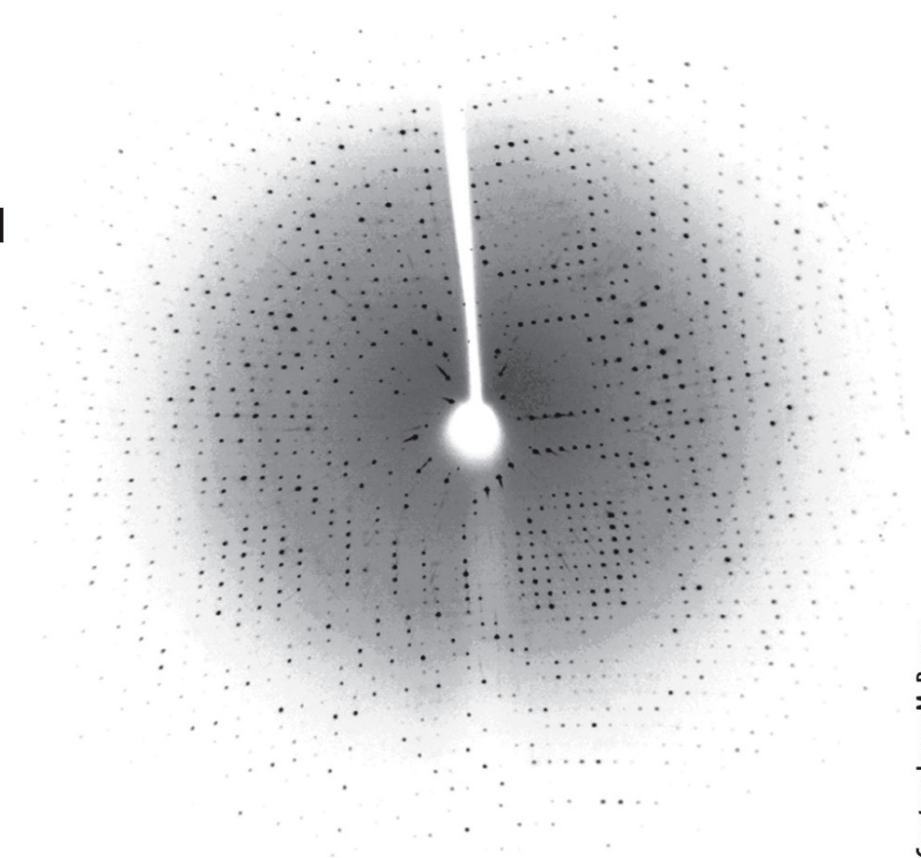
**Figure 3-44a**  
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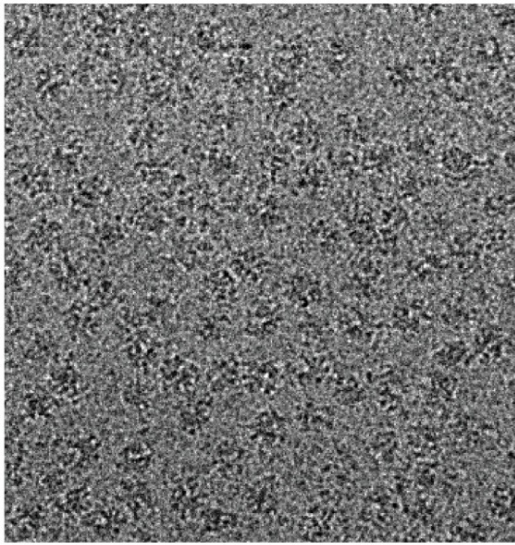
**Figure 3-44b**  
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**Figure 3-45a**  
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**Figure 3-45b**  
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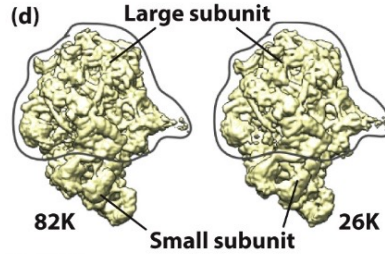
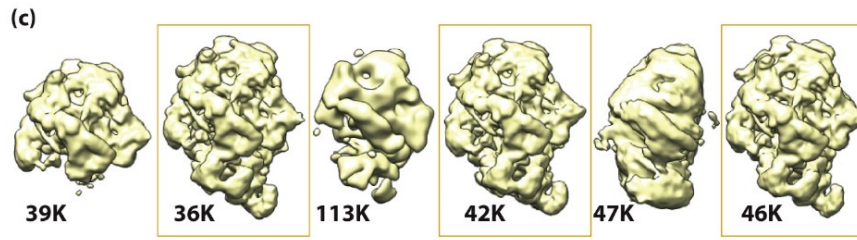
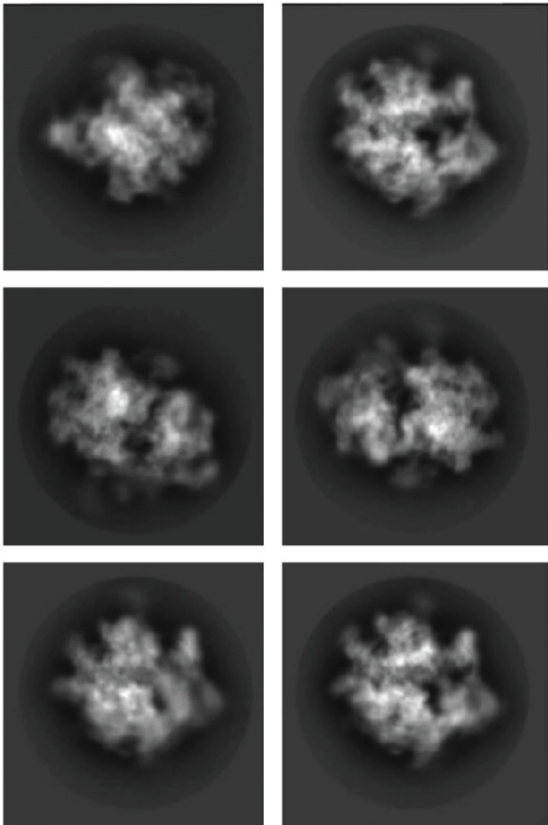


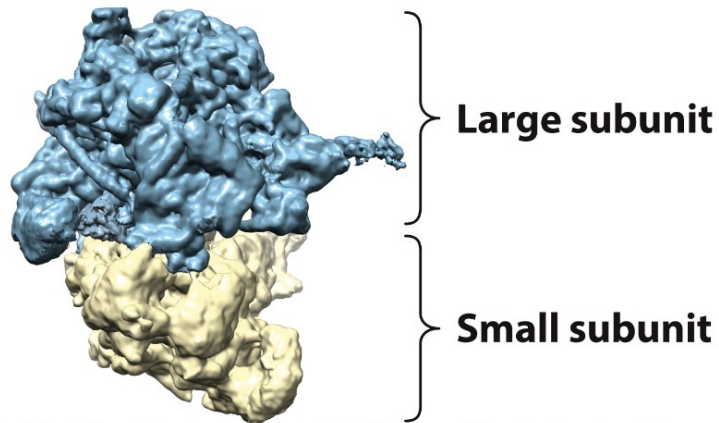
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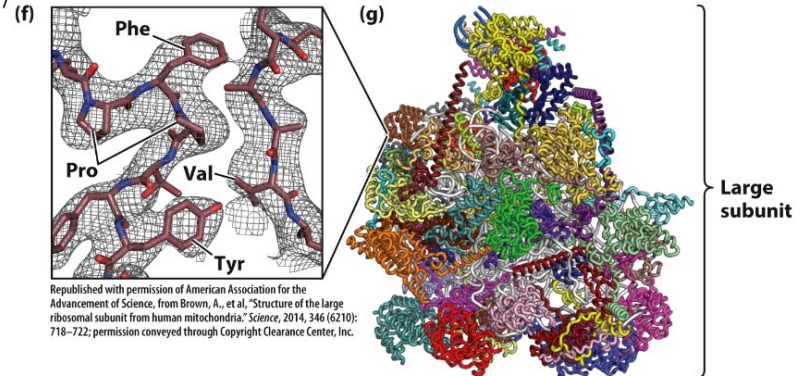
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