Hierachy

Protein structures can be organized into four levels of hierarchies with increasing complexity. These levels are primary structure, secondary structure, tertiary structure, and quaternary structure.

- **Primary structure**: A linear amino acid sequence of a protein is the primary structure. This is the simplest level with amino acid residues linked together through peptide bonds.
- **Secondary structure**: Most proteins have segments of their polypeptide chains repeatedly coiled or folded in patterns that contribute to the protein's overall shape. These coils and folds, collectively referred to as secondary structure, are the result of hydrogen bonds between the repeating constituents of the polypeptide backbone, between main chain atoms of the C=O group and the NH group of different residues. Individually, these hydrogen bonds are weak, but because they are repeated many times over a relatively long region of the polypeptide chain, they can support a particular shape for that part of the protein.
    - ✓ One such secondary structure is the **α helix**, a delicate coil held together by hydrogen bonding between every fourth amino acid.
    - ✓ The other main type of secondary structure is the **β pleated sheet**. Two or more regions of the polypeptide chain lying side by side are connected by hydrogen bonds between parts of the two parallel polypeptide backbones. Pleated sheets make up the core of many globular proteins and dominate some fibrous proteins.
- The **tertiary structure**, which is the three-dimensional arrangement of various secondary structural elements and connecting regions. The tertiary structure can be described as the complete three-dimensional assembly of all amino acids of a single polypeptide chain. Superimposed on the patterns of secondary structure is a protein's tertiary structure. While secondary structure involves interactions between backbone constituents, tertiary structure is the overall shape of a polypeptide resulting from interactions between the sidechains (R groups) of the various amino acids. One type of interaction that contributes to tertiary structure is called a hydrophobic interaction. As a polypeptide folds into its functional shape, amino acids with hydrophobic (nonpolar) side chains usually end up in clusters at the core of the protein, out of contact with water. Thus, what we call a hydrophobic interaction is actually caused by the action of water molecules, which exclude nonpolar substances as they form hydrogen bonds with each other and with hydrophilic parts of the protein. Once nonpolar amino acid side chains are close together, van der Waals interactions help hold them together. Meanwhile. hydrogen bonds between polar side chains and ionic bonds between positively and negatively charged side chains also help stabilize tertiary structure. These are all weak interactions, but their cumulative effect helps

give the protein a unique shape. The shape of a protein may be reinforced further by covalent bonds caned disulfide bridges. Disulfide bridges form where two cysteine monomers, amino acids with sulfhydryl groups (-SH) on their side chains, are brought close together by the folding of the protein

- Beyond the tertiary structure is the **quaternary structure**, which refers to the association of several polypeptide chains into a protein complex, which is maintained by noncovalent interactions. In such a complex, individual polypeptide chains are called monomers or subunits. Intermediate between secondary and tertiary structures, a level of supersecondary structure is often used, which is defined as two or three secondary structural elements forming a unique functional domain, a recurring structural pattern conserved in evolution. Quaternary structure is the overall protein structure that results from the aggregation of these polypeptide subunits.

Stabilizing Forces

Protein structures from secondary to quaternary are maintained by noncovalent forces. These include electrostatic interactions, van der Waals forces, and hydrogen bonding.

- ✓ **Electrostatic interactions** are a significant stabilizing force in a protein structure. They occur when excess negative charges in one region are neutralized by positive charges in another region. The result is the formation of salt bridges between oppositely charged residues. The electrostatic interactions can function within a relatively long range (15 Å).
- ✓ **Hydrogen bonds** are a particular type of electrostatic interactions similar to dipole–dipole interactions involving hydrogen from one residue and oxygen from another. Hydrogen bonds can occur between main chain atoms as well as side chain atoms. Hydrogen from the hydrogen bond donor group such as the N–H group is slightly positively charged, whereas oxygen from the hydrogen bond acceptor group such as the C=O group is slightly negatively charged. When they come within a close distance (< 3 Å), a partial bond is formed between them, resulting in a hydrogen bond. Hydrogen bonding patterns are a dominant factor in determining different types of protein secondary structures.
- ✓ **Van der Waals forces** also contribute to the overall protein stability. These forces are instantaneous interactions between atoms when they become transient dipoles. A transient dipole can induce another transient dipole nearby. The dipoles of the two atoms can be reversed a moment later. The oscillating dipoles result in an attractive force. The van der Waals interactions are weaker than electrostatic and hydrogen bonds and thus only have a secondary effect on the protein structure.
- ✓ In addition to these common stabilizing forces, **disulfide bridges**, which are covalent bonds between the sulfur atoms of the cysteine residue, are also important in maintaining some protein structures. For certain types of proteins that contain metal

ions as prosthetic groups, noncovalent interactions between amino acid residues and the metal ions may play an important structural role.

Secondary structures

- **α-helices**

An α-helix has a main chain backbone conformation that resembles a corkscrew. Nearly all known α-helices are right-handed, exhibiting a rightward spiral form. In such a helix, there are 3.6 amino acids per helical turn. The structure is stabilized by hydrogen bonds formed between the main chain atoms of residues i and i + 4. The hydrogen bonds are nearly parallel with the helical axis. The average φ and ψ angles are 60◦ and 45◦, respectively, and are distributed in a narrowly defined region in the lower left region of a Ramachandran plot. Hydrophobic residues of the helix tend to face inside and hydrophilic residues of the helix face outside. Thus, every third residue along the helix tends to be a hydrophobic residue. Ala, Gln, Leu, and Met are commonly found in an α-helix, but not Pro, Gly, and Tyr. These rules are useful in guiding the prediction of protein secondary structures.

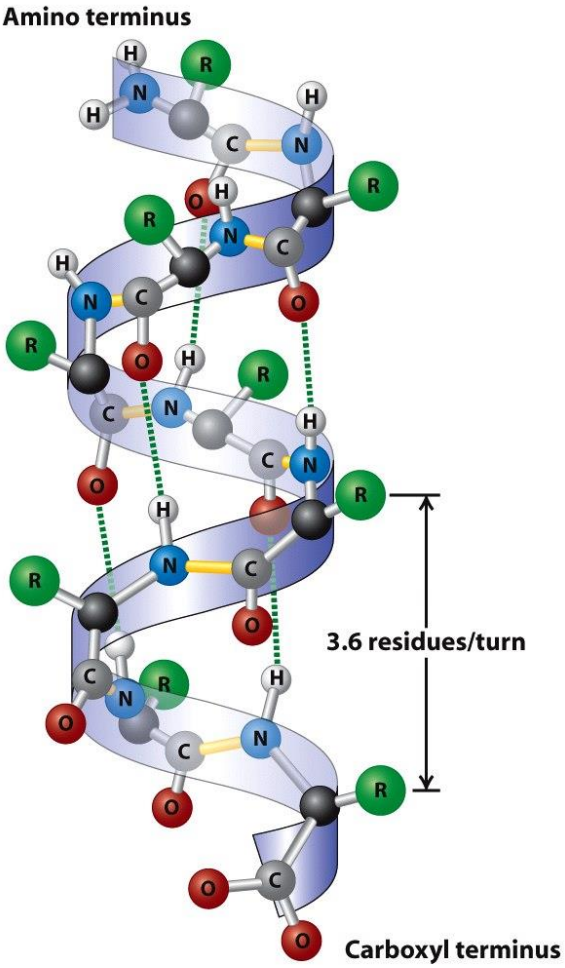

Figure 3-4
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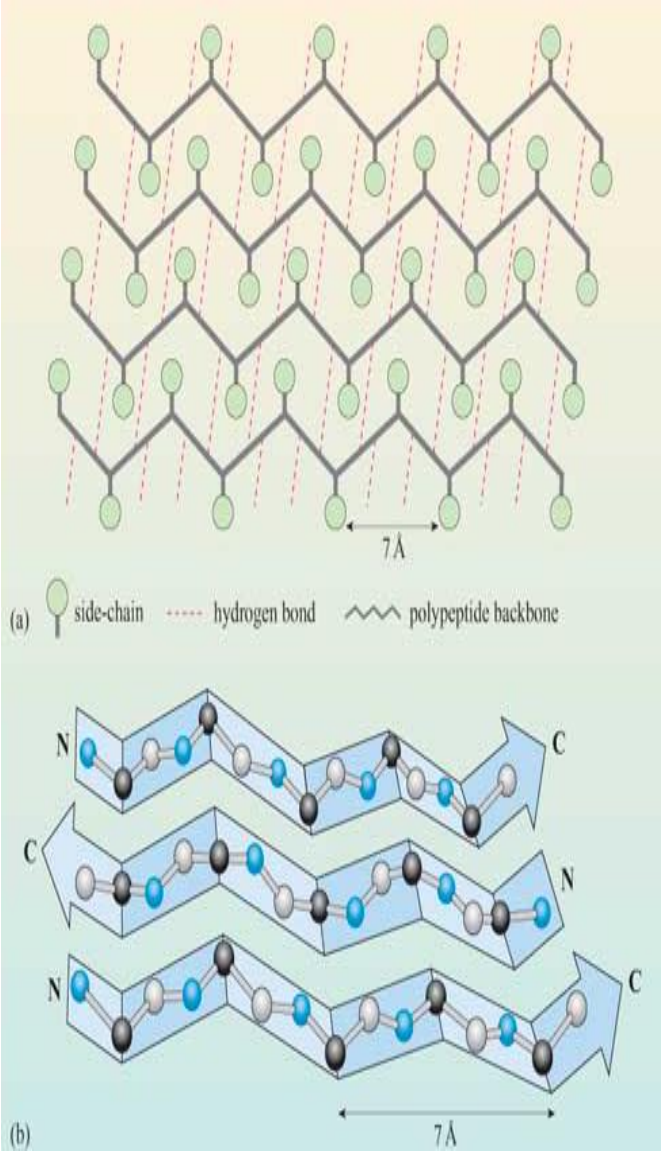
- **β-Sheets**

  A β-sheet is a fully extended configuration built up from several spatially adjacent regions of a polypeptide chain. Each region involved in forming the β-sheet is a β-strand. The β-strand conformation is pleated with main chain backbone zigzagging and side chains positioned alternately on opposite sides of the sheet. β-Strands are stabilized by hydrogen bonds between residues of adjacent strands. β-strands near the surface of the protein tend to show an alternating pattern of hydrophobic and hydrophilic regions, whereas strands buried at the core of a protein are nearly all hydrophobic. The β-strands can run in the same direction to form a parallel sheet or can run every other chain in reverse orientation to form an antiparallel sheet, or a mixture of both. The hydrogen bonding patterns are different in each configuration. The φ and ψ angles are also widely distributed in the upper left region in a Ramachandran plot. Because of the long-range nature of residues involved in this type of conformation, it is more difficult to predict β-sheets than α helices.



- **Coils and Loops**

  There are also local structures that do not belong to regular secondary structures (α-helices and β-strands). The irregular structures are coils or loops. The loops are often characterized by sharp turns or hairpin-like structures. If the connecting regions are completely irregular, they belong to random coils. Residues in the loop or coil regions tend to be charged and polar and located on the surface of the protein structure. They are often the evolutionarily variable regions where mutations, deletions, and insertions frequently occur. They can be functionally significant because these locations are often the active sites of proteins. Coiled Coils Coiled coils are a special type of supersecondary structure characterized by a bundle of two or more α-helices wrapping around each other.

The helices forming coiled coils have a unique pattern of hydrophobicity, which repeats every seven residues (five hydrophobic and two hydrophilic).

Tertiary structure

The overall packing and arrangement of secondary structures form the tertiary structure of a protein. The tertiary structure can come in various forms but is generally classified as either globular or membrane proteins. The former exists in solvents through hydrophilic interactions with solvent molecules; the latter exists in membrane lipids and is stabilized through hydrophobic interactions with the lipid molecules.

- **Globular Proteins**

  Globular proteins are usually soluble and surrounded by water molecules. They tend to have an overall compact structure of spherical shape with polar or hydrophilic residues on the surface and hydrophobic residues in the core. Such an arrangement is energetically favorable because it minimizes contacts with water by hydrophobic residues in the core and maximizes interactions with water by surface polar and charged residues. Common examples of globular proteins are enzymes, myoglobins, cytokines, and protein hormones.

- **Integral Membrane Proteins**

  Membrane proteins exist in lipid bilayers of cell membranes. Because they are surrounded by lipids, the exterior of the proteins spanning the membrane must be very hydrophobic to be stable. Most typical transmembrane segments are α-helices. Occasionally, for some bacterial periplasmic membrane proteins, they are composed of β-strands. The loops connecting these segments sometimes lie in the aqueous phase, in which they can be entirely hydrophilic. Sometimes, they lie in the interface between the lipid and aqueous phases and are amphipathic in nature (containing polar residues facing the aqueous side and hydrophobic residues towards the lipid side). The amphipathic residues can also form helices which have a periodicity of three or four residues. Common examples of membrane proteins are rhodopsins, cytochrome c oxidase, and ion channel proteins.