Protein Folding – Secondary Structures

Lecture 6

Acknowledgements:
Alberts - Molecular Biology of the Cell
Scitable by Nature Education
Nature Resources
Internet resources

OBJECTIVE OF THE LECTURE

1. Understand the principles Protein folding
2. Phi-psi angles
3. Ramachandran plots
4. Secondary Protein structures

3-D structure of Proteins

1. the three-dimensional structure of a protein is determined by its amino acid sequence
2. the function of a protein depends on its structure
3. an isolated protein usually exists in one or a small number of stable structural forms
4. the most important forces stabilizing the specific structures maintained by a given protein are noncovalent interactions

Stability of protein structures

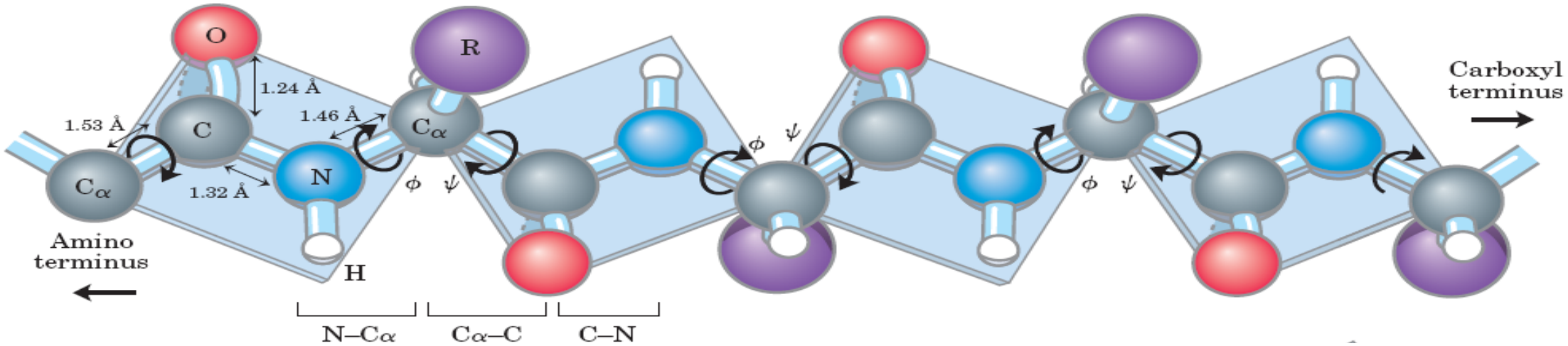
- The stability of protein structures depends on weak interactions
- It requires 200-460 kJ/mol to break a single covalent bond, compared to 4-30kJ/mol for weak interactions
  - The weak interactions predominate because they are numerous
- The free energies of the folded and un-folded states are similar

Governing Equations

$\Delta G = \Delta H - \Delta TS$

$\Delta G = -RT\ln k$

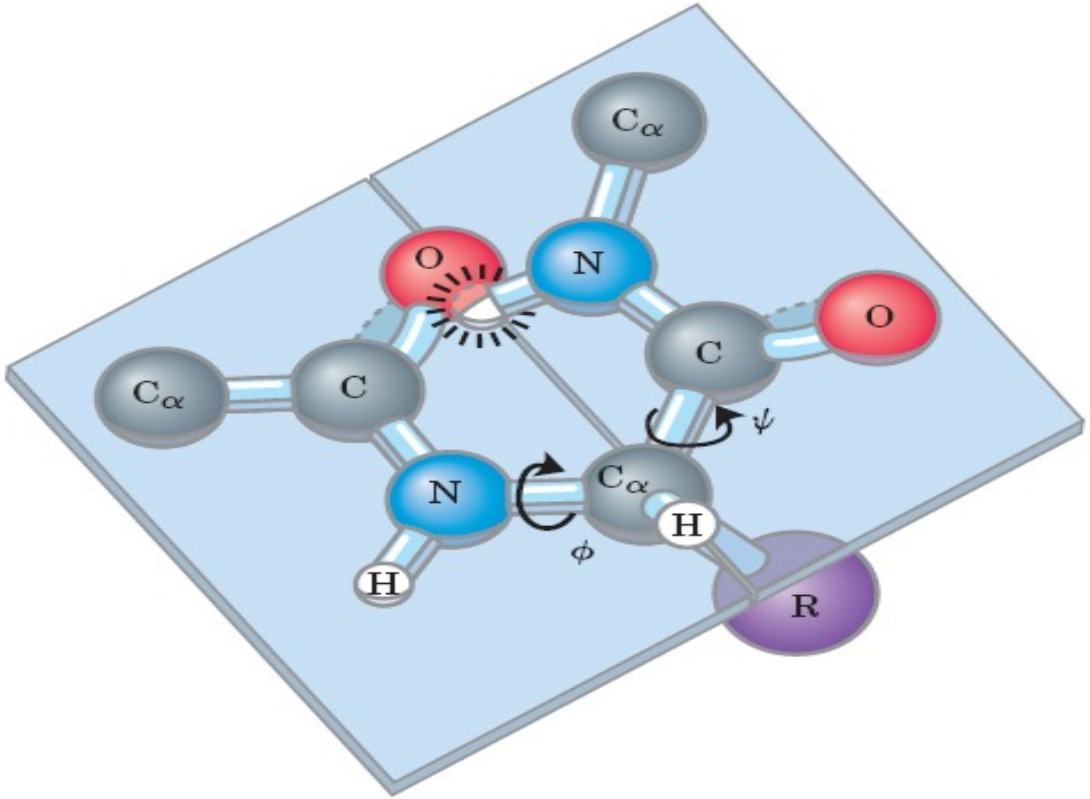
The phi and psi angles



- Three bonds separate sequential α carbons in a polypeptide chain. The N—Cα and C—C α bonds can rotate, with bond angles designated ϕ and ψ, respectively
- The peptide C—N bond is not free to rotate
- Other single bonds in the backbone may also be rotationally hindered, depending on the size and charge of the R groups. In the conformation shown, ϕ and ψ are 180 deg (or – 180 deg).
- As one looks out from the α-carbon, the ϕ and ψ angles increase as the carbonyl or amide nitrogens (respectively) rotate clockwise

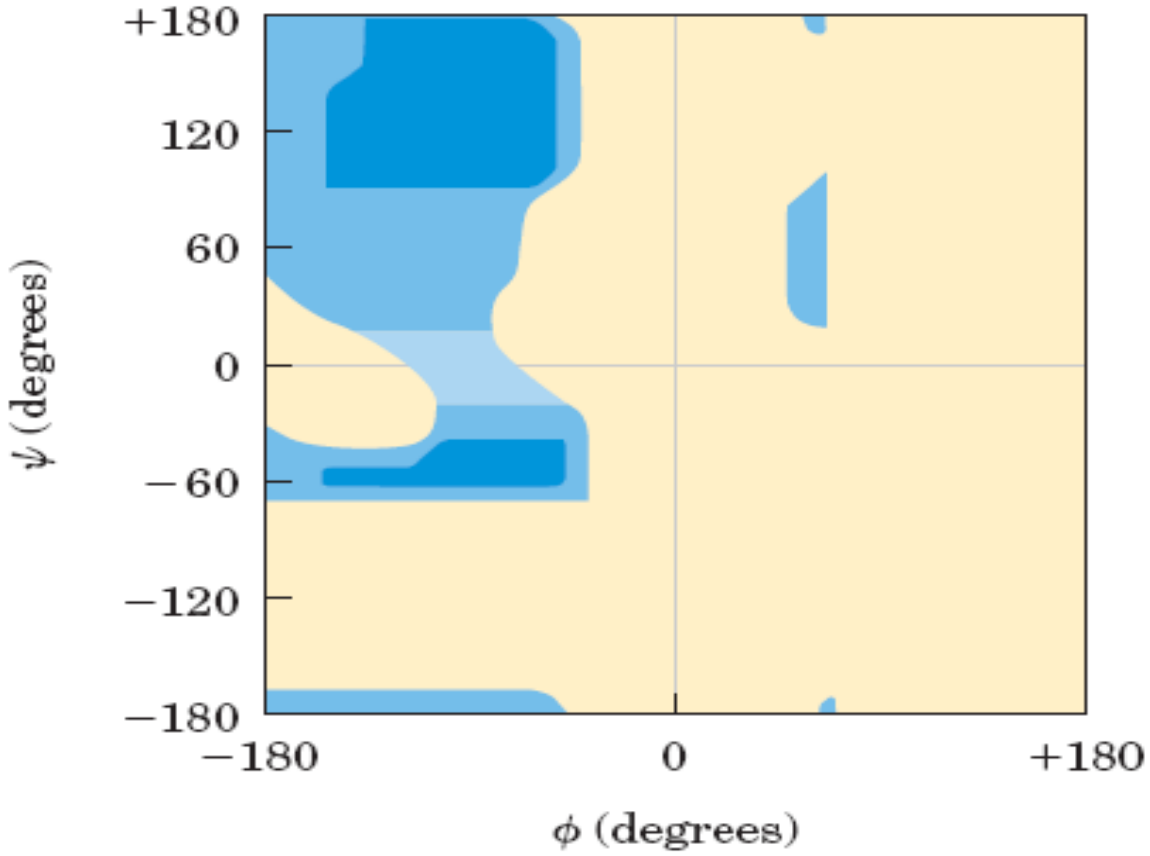
The zero phi & psi angles

- By convention, both ϕ and ψ are defined as 0 deg when the two peptide bonds flanking that carbon are in the same plane and positioned as shown.
- In a protein this conformation is prohibited by steric overlap between an -carbonyl oxygen and an –amino hydrogen atom
- To illustrate the bonds between atoms, the balls representing each atom are smaller than the van der Waals radii for this scale. 1 Å = 0.1 nm.



The Ramachandran Plot for *L*-Ala

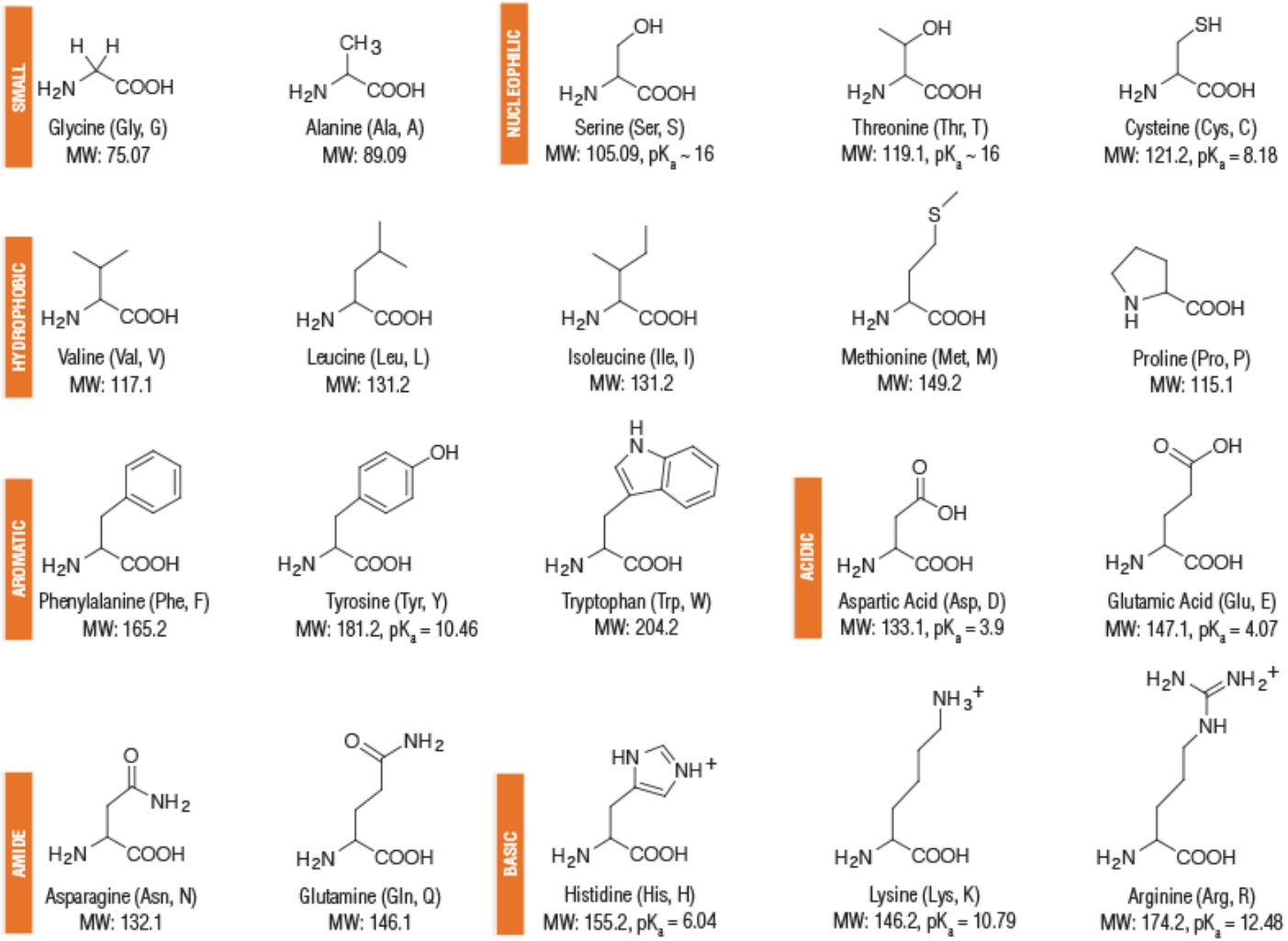
- Conformations deemed possible are those that involve little or no steric interference, based on calculations using known van der Waals radii and bond angles.
  - The areas shaded dark blue reflect conformations that involve no steric overlap and thus are fully allowed
  - medium blue indicates conformations allowed at the extreme limits for unfavorable atomic contacts
  - the lightest blue area reflects conformations that are permissible if a little flexibility is allowed in the bond angles
- The asymmetry of the plot results from the L stereochemistry of the amino acid residues



About Ramachandran Plots

1. What kind of plot do you expect for other un-branched amino-acids?
2. What kind of plot do you expect for branched amino-acids (eg Ile)?
3. What kind of plot do you expect for glycine?
4. What kind of plot do you expect for Proline?

Structure of Amino Acids



What are the possible applications of the RAMACHANDRAN PLOT?

PROTEIN STRUCTURE PREDICTION

ALPHA FOLD

https://alphafold.ebi.ac.uk/

SCFBio IIT Delhi

http://www.scfbio-iitd.res.in

PROTEIN STRUCTURE

Overview of Protein Structure

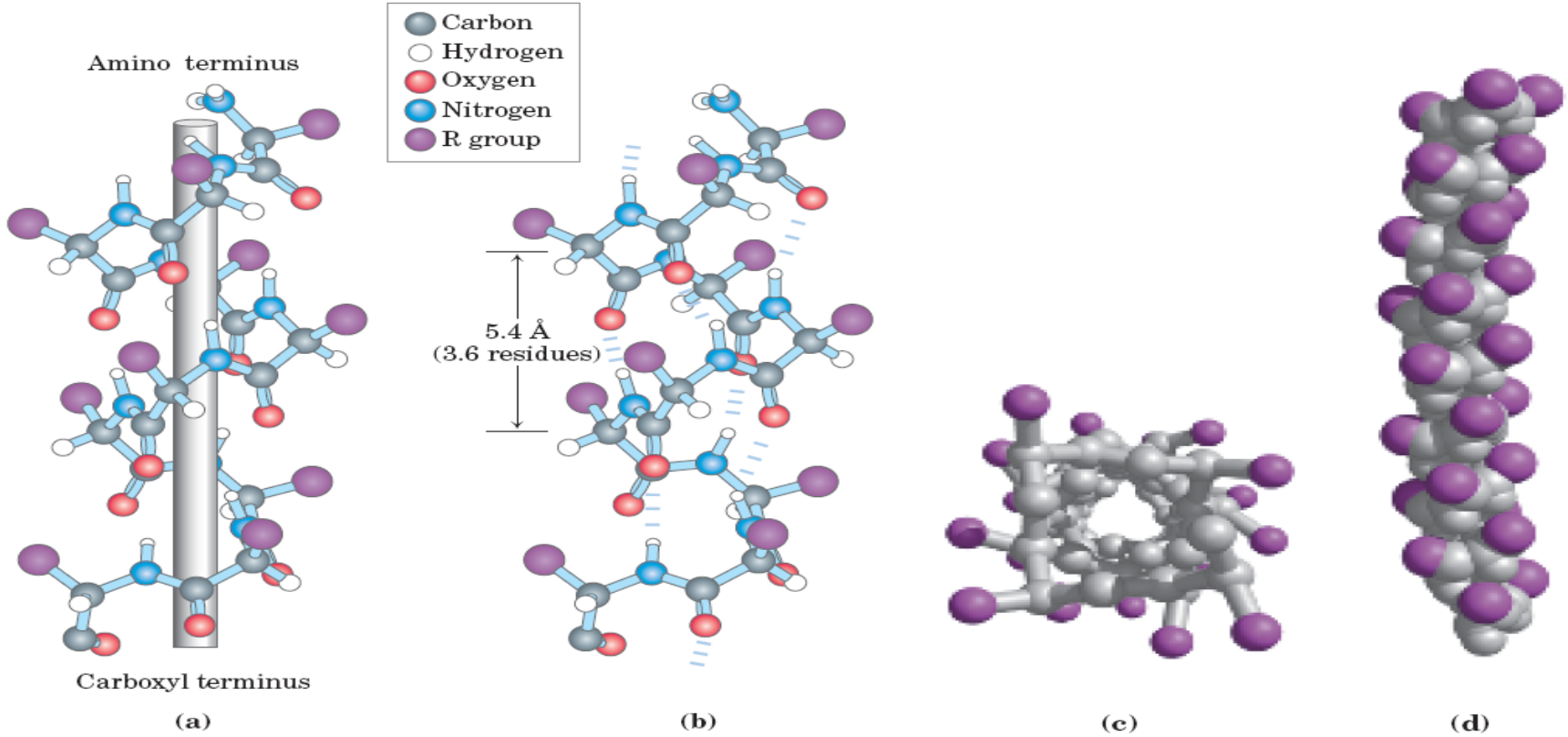
- Every protein has a three-dimensional structure that reflects its function.
- Protein structure is stabilized by multiple weak interactions. Hydrophobic interactions are the major contributors to stabilizing the globular form of most soluble proteins; hydrogen bonds and ionic interactions are optimized in the specific structures that are thermodynamically most stable.
- The nature of the covalent bonds in the polypeptide backbone places constraints on structure. The peptide bond has a partial double bond character that keeps the entire six-atom peptide group in a rigid planar configuration. The N–C_{α} and C_{α}–C bonds can rotate to assume bond angles of ϕ and ψ, respectively.

Classification of Strutures

- Secondary
  - Alpha Helix
  - Beta sheets
- 3-D conformations
  - Structure and function
- Tertiary Structures
  - Globular proteins
  - Multimerics
    - Homo
    - hetero

Protein Secondary Structure

- The α-helix architecture
  - Linus Pauling, Robert Corey
  - X-ray results of William Astbury (1930) of proteins that make up porcupine quills (α-keratin)
    - Regular structure that repeats every 5.16 – 5.2 Å
    - polypeptide backbone is tightly wound around an imaginary axis drawn longitudinally through the middle of the helix, and the R groups of the amino acid residues protrude outward from the helical backbone
    - The amino acid residues in an
    - helix have conformations with ψ= -45 to -50 deg and ϕ = - 60 deg



a) Formation of a right-handed α-helix. The planes of the rigid peptide bonds are parallel to the long axis of the helix, depicted here as a vertical rod

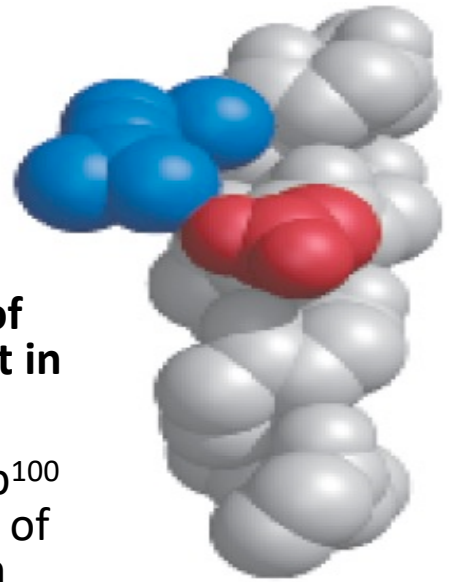
b) Ball-and-stick model of a right-handed α- helix, showing the intrachain hydrogen bonds. The repeat unit is a single turn of the helix, 3.6 residues

c) The α- helix as viewed from one end, looking down the longitudinal axis

d) Atoms in the center of the α- helix are in very close contact

Amino Acid Sequence Affects α-Helix Stability

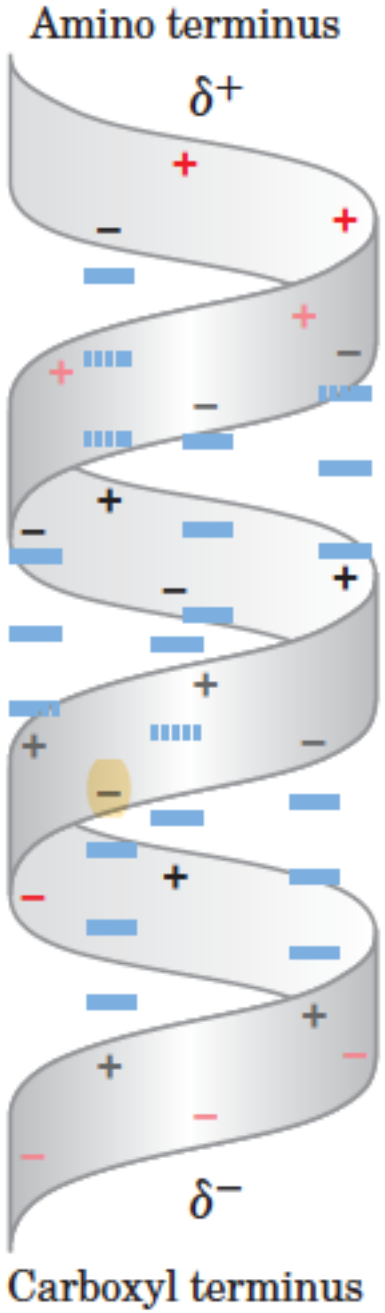
- If a polypeptide chain has a long block of Glu residues, this segment of the chain will not form an helix at pH 7.0.
  - The negatively charged carboxyl groups of adjacent Glu residues repel each other so strongly that they prevent formation of the α- helix
  - If there are many adjacent Lys and/or Arg residues, which have positively charged R groups at pH 7.0, they will also repel each other and prevent formation of the α- helix
  - The bulk and shape of Asn, Ser, Thr, and Cys residues can also destabilize an helix if they are close together in the chain
  - The twist of an helix ensures that critical interactions occur between an amino acid side chain and the side chain three (and sometimes four) residues away on either side of it. Positively charged amino acids are often found three residues away from negatively charged amino acids, permitting the formation of an ion pair



- **Interactions between R groups of amino acids three residues apart in an α-helix**
- An ionic interaction between Asp^{100} and Arg^{103} in an α- helical region of the protein troponin C, a calcium binding protein associated with muscle
- Polypeptide backbone (carbons, α-amino nitrogens, and α-carbonyl oxygens) is shown in gray for a helix segment 13 residues long
- The interacting Asp (red) and Arg (blue) side chains

Helix Dipole

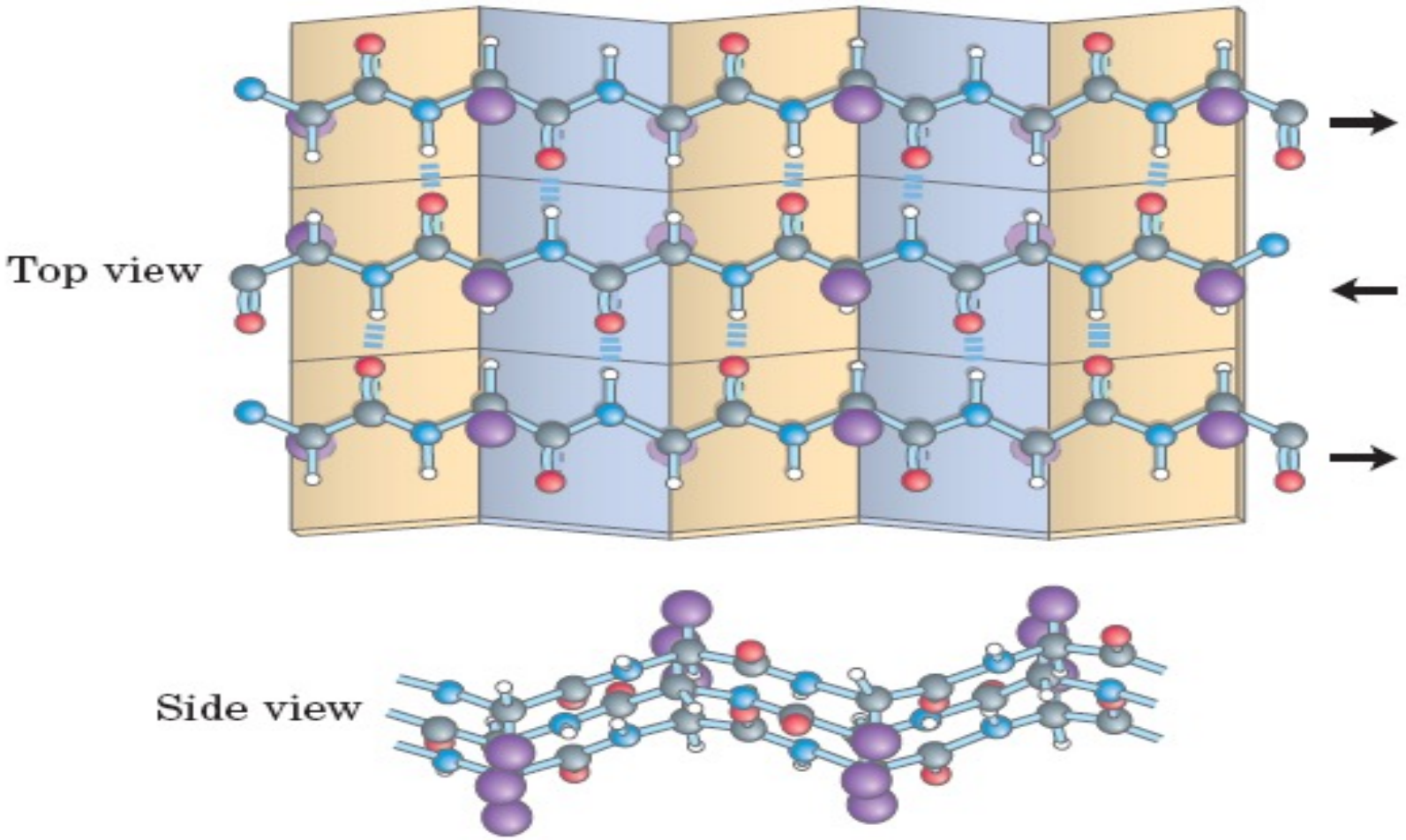
- The electric dipole of a peptide bond is transmitted along an α-helical segment through the intra-chain hydrogen bonds, resulting in an overall helix dipole
- The amino and carbonyl constituents of each peptide bond are indicated by + and - symbols
- Non-hydrogen bonded amino and carbonyl constituents in the peptide bonds near each end of the α-helical region are shown in red.



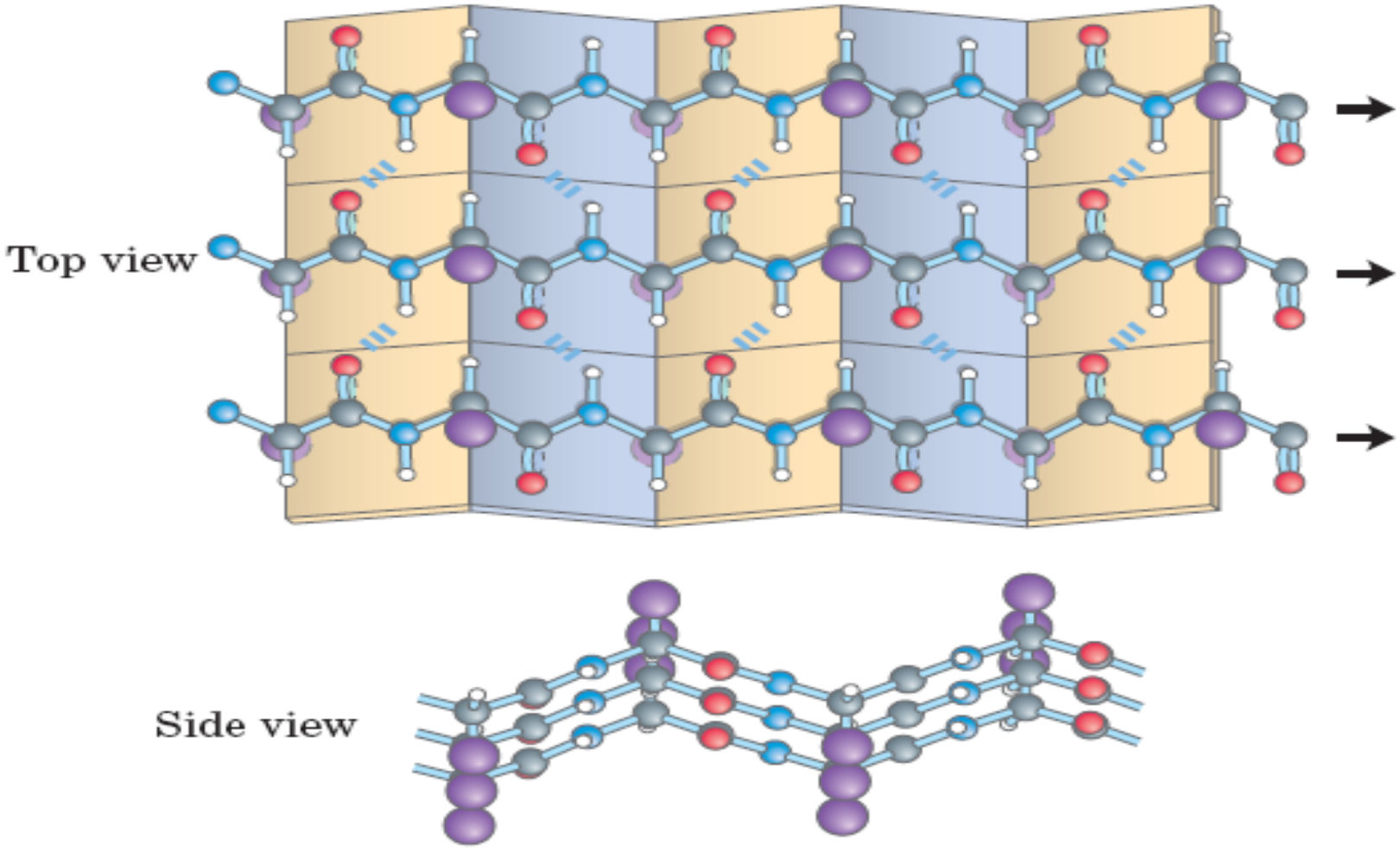
Constraints affecting stability of α-helix

1. The electrostatic repulsion (or attraction) between successive amino acid residues with charged R groups
2. The bulkiness of adjacent R groups
3. The interactions between R groups spaced three (or four) residues apart
4. The occurrence of Pro and Gly residues
5. The interaction between amino acid residues at the ends of the helical segment and the electric dipole inherent to the helix

Antiparallel β-Sheet Conformation



Parallet β-Sheet Conformation

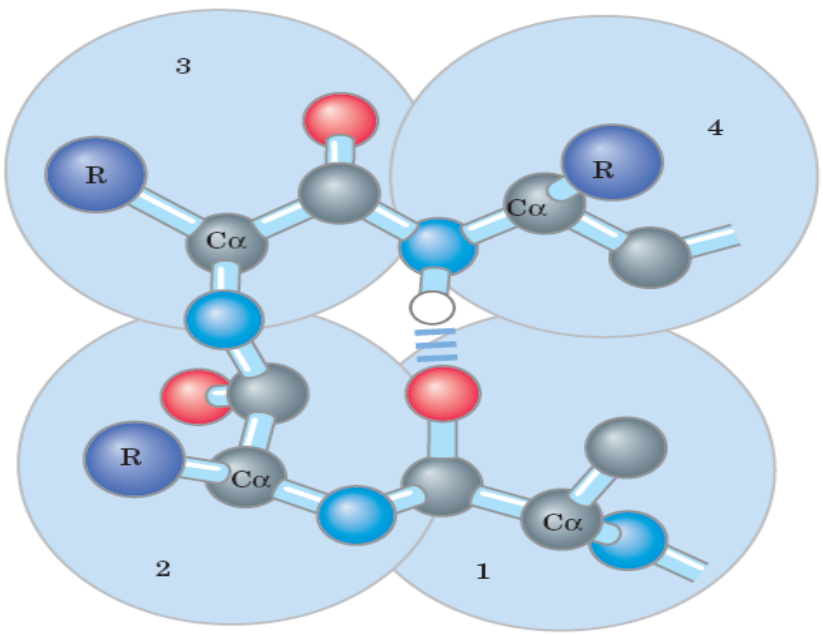


β-Sheet Conformation

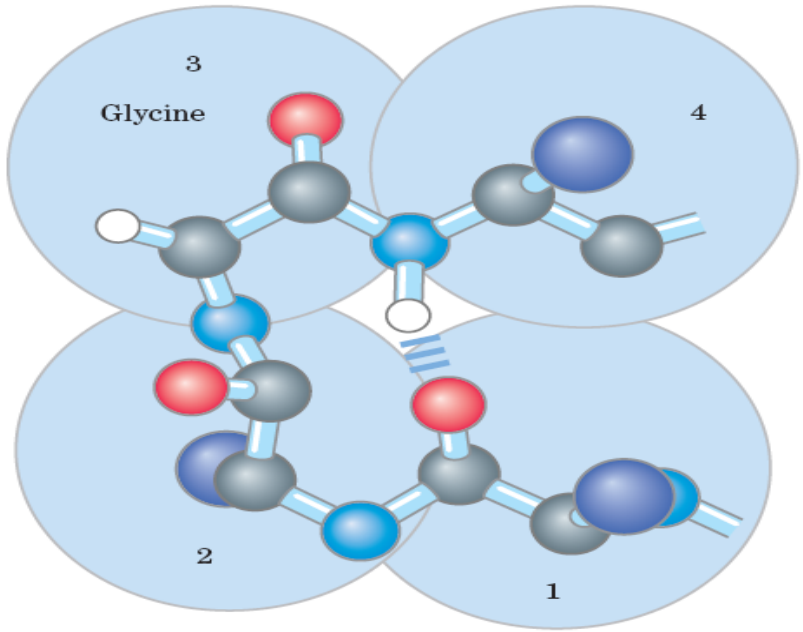
- The zigzag polypeptide chains can be arranged side by side to form a structure resembling a series of pleats
- Hydrogen bonds are formed between adjacent segments of polypeptide chain
- The adjacent polypeptide chains in a sheet can be either parallel or antiparallel (having the same or opposite amino-to-carboxyl orientations)
- The repeat period is shorter for the parallel conformation 6.5 Å, versus 7 Å for antiparallel

β-turns in protein architecture

- **Turns** that connect the ends of two adjacent segments of an antiparallel sheet. The structure is a 180 deg turn involving four amino acid residues, with the carbonyl oxygen of the first residue forming a hydrogen bond with the amino-group hydrogen of the fourth

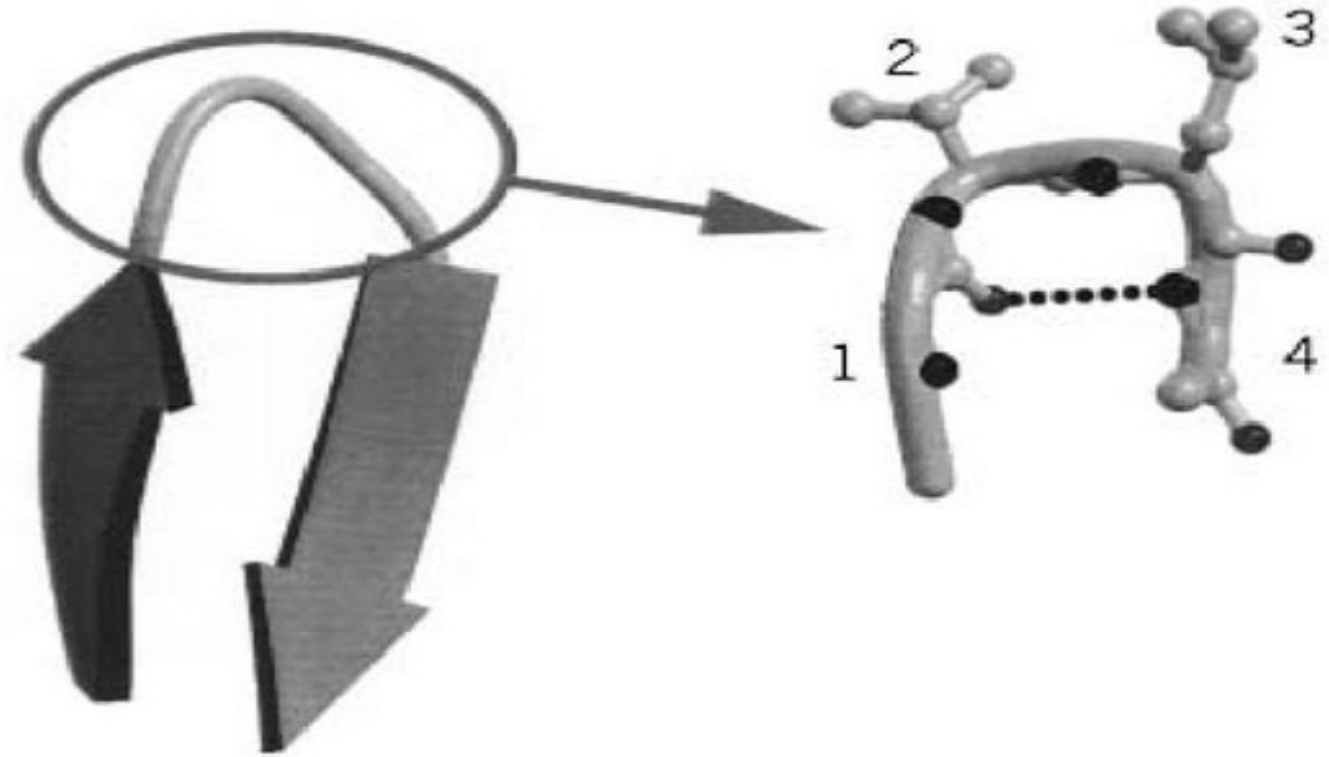


Type I



Type II

Beta Turn



Bonding between the 1st and 3rd amino acid. The 3rd position is usually occupied by glycine or proline. In such positions, proline takes on a cis-orientation (which has only about 6% occurrence in proteins)