E-content

M.Sc. Zoology (Semester II)
CC7- Biochemistry

Unit: 3.3

Peptide conformations (Ramachandran plot, helices, turns and sheets)

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The peptide bond conformations

The **primary structure** of a protein is defined as the sequence of amino acids in a polypeptide chain.

This sequence determines the shape of the protein which depends on the spatial limitations of the atoms present in the protein, the chemical properties of the component amino acid residues, and the protein's environment.

The peptide bonds that link amino acid residues in a polypeptide are formed in a condensation reaction between the carboxyl group of one amino acid and the basic amino group of another amino acid.

In the context of a peptide, the CO–NH group is referred to as the **peptide bond**.

Linus Pauling, in the 1930s, used X-ray diffraction to examine the nature of the peptide bond formed between two amino acids. He reported that the peptide group (CO–NH) has *a rigid planar structure*.

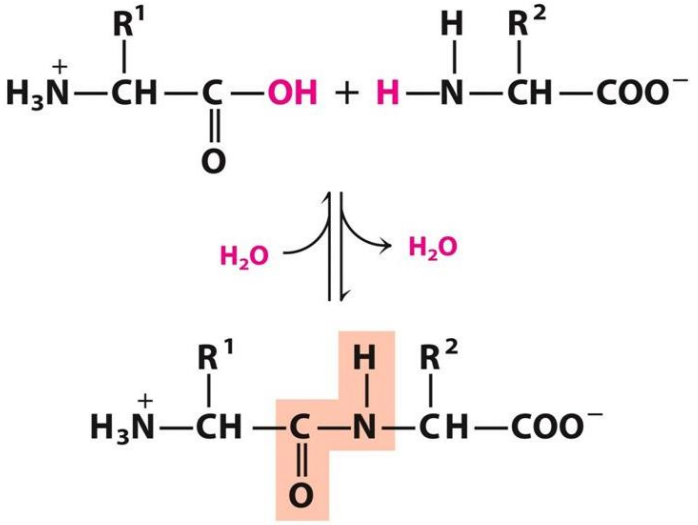


Figure 1: Peptide bond joins two amino acids

Peptide bond have partial double bond properties

The rigid planer structure of peptide bond is due to interactions between electrons of the double bond of the carbonyl group and those of the C–N bond such that the latter acquires partial (about 40%) double-bond properties.

This effect is an example of **resonance** which can be thought of as a sharing of electrons between bonds.

The partial double bond character of C–N in the peptide group means that this bond is shorter than would be predicted for a C–N single bond, whilst the C=O bond, having a partial single bond character due to resonance, is longer than would be predicted for a C=O double bond.

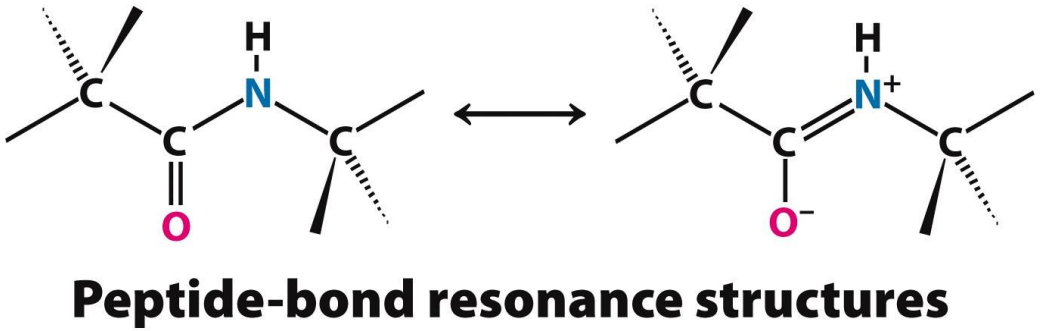


Figure 2: Resonance interactions between electrons in the C=O bond and the C–N bond of the peptide group mean that there is ‘sharing’ of electrons between these bonds. Note the charges on the N and O atoms.

Peptide bond lengths

The bond lengths in the peptide group are indicated in figure 3. Compare the C–N bond of the peptide group with that between N and C_α (the C atom to which the amino group and carboxyl group are attached).

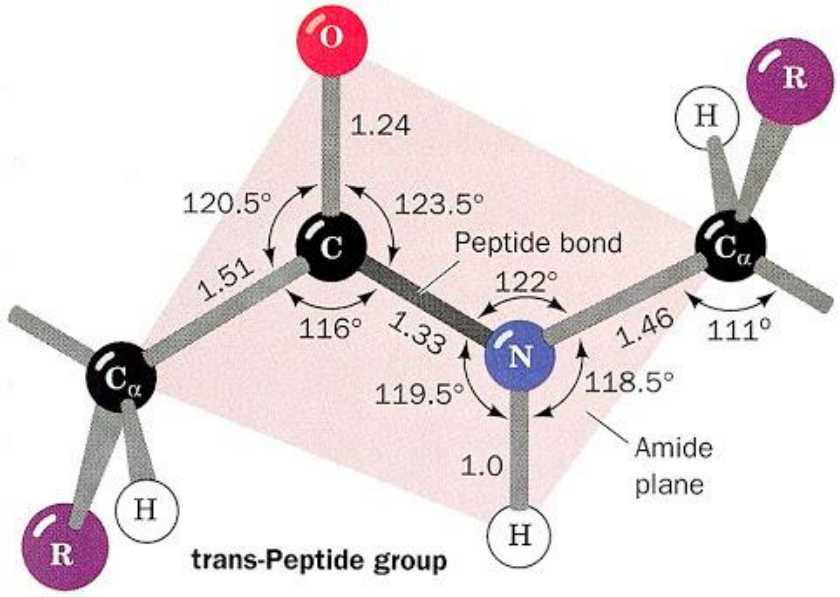


Figure 3: The average dimensions in angstroms, Å, and degrees, of the planar peptide group in the *trans* conformation

Trans and cis peptide bonds

There are two possible conformations of the planar peptide bond: in the *trans* peptide group, the C_α atoms are on opposite sides of the peptide bond (**Figure 4**) and in the *cis* peptide group, the C_α atoms are on the same side of the peptide bond (**Figure 4**).

Generally speaking, peptide bonds are in the *trans* conformation. However, c*is* forms can occur in peptide bonds that precede a proline residue. In such cases, the *cis* form is more stable than usual since the proline side-chain offers less of a hindrance.

Almost all peptides are in trans.

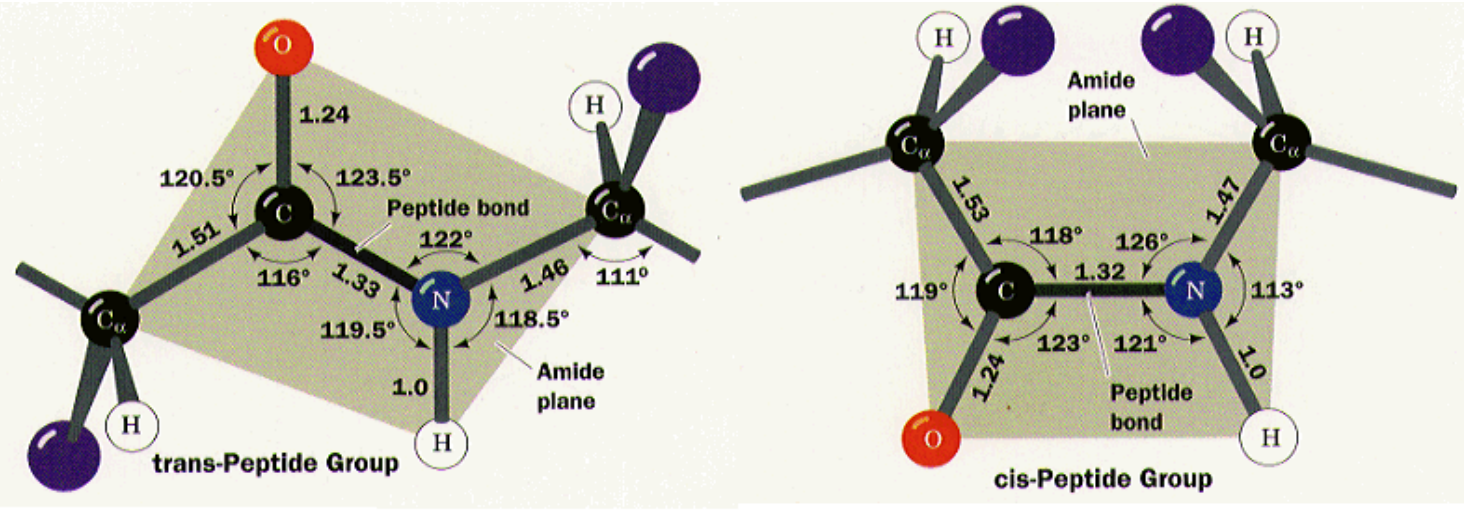


Figure 4: trans is strongly favored over cis form

Rotation of bonds in a polypeptide

Peptide conformations is defined by three dihedral angles (also known as torsion angles) called ϕ (phi), ψ (psi), and ω(omega), reflecting rotation about each of the three repeating bonds in the polypeptide backbone.

Within the peptide bond, the bond between the amino group and the α-carbon atom and between the α-carbon atom and the carbonyl group are pure single bonds.

The two adjacent rigid peptide units may rotate about these bonds, taking various orientations.

The angle of rotation between the nitrogen and the α-carbon atom is called ϕ.

The angle of rotation between the α-carbon atom and the carbonyl group is called ψ.

Ω is not often considered. It involves the carbon and nitrogen atom of peptide bond, where rotation is constrained.

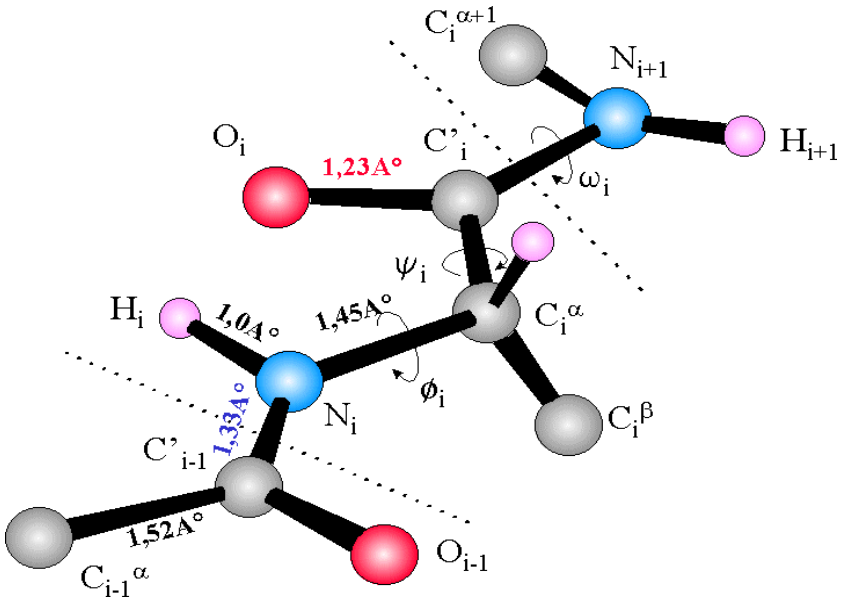


Figure 5: dihedral angles in peptide backbone

Rotation of bonds in a polypeptide: Ramachandran Plot

It has been found that steric constraints apply to ϕ and ψ. As a result of these steric constraints, only certain values of ϕ and ψ, and hence conformations of the peptide, are permitted whereas others are not.

It is possible to calculate these permitted values for a given residue in the context of a polypeptide. It is most easily done for a polypeptide containing just one kind of amino acid. A conformational plot of ϕ against ψ for a particular residue is known as a **Ramachandran plot** (after its inventor, G. N. Ramachandran).

Such a plot allows us to identify a particular value of ϕ and ψ that are sterically favourable or unfavourable, based on the following criteria:

a) Where there is no conflict between the van der Waals radii of non-bonding atoms, a conformation is 'allowed'. These conformations lie in the blue areas in **Figure 6**.

b) Conformations requiring interatomic distances at the limit of that which is permissible are defined as 'outer limit' conformations. They lie in the green areas in **Figure 6**.

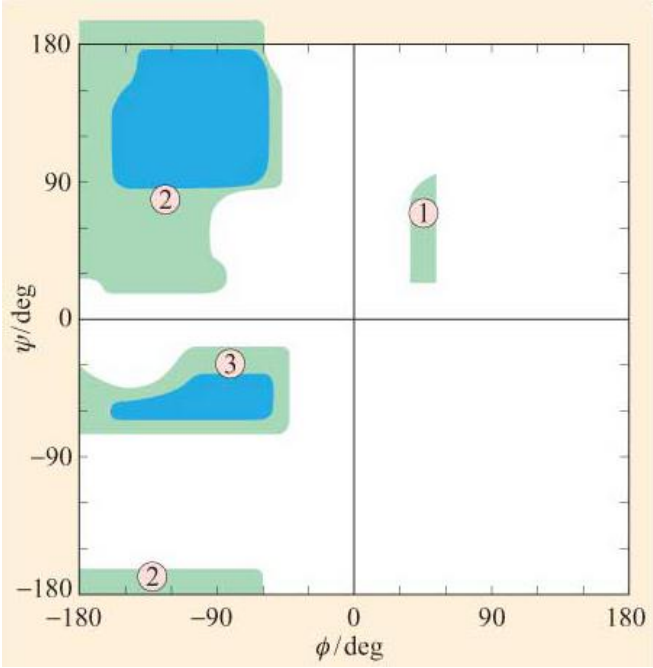


Figure 6: Ramachandran plot for a peptide chain containing only poly-L-alanine residue

Rotation of bonds in a polypeptide: Ramachandran Plot

Theoretical conformations that require any two non-bonding atoms to be closer to each other than their van der Waals radii allow are sterically 'forbidden'. These lie in the white areas in **Figure 6**.

Notice that the values of ϕ and ψ in **Figure 6** range from −180º to +180º. Turning the peptide group through 360º will of course bring it back to its starting position, and −180º and +180º correspond to the same position. Thus the green strip at the bottom left corner of the plot in **Figure 6** is contiguous with the field at the top left corner.

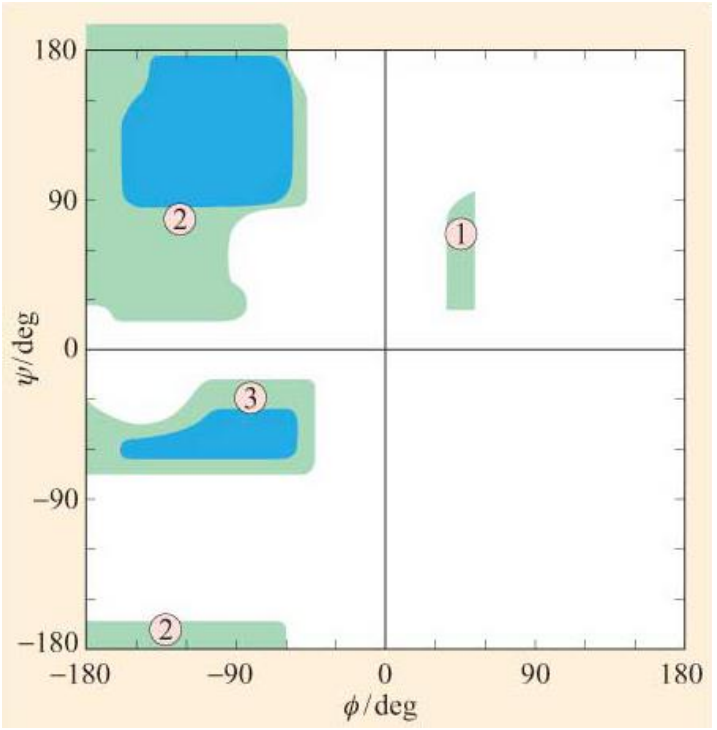


Figure 6: Ramachandran plot showing the sterically allowed ϕ and ψ angles for a peptide chain containing only L-alanine residues (poly-L-alanine). 'Allowed' conformations are within the blue or green areas, with those that are 'outer limit' conformations in the latter. The remaining conformations, in the white area, are 'forbidden'. Note that there are only three discrete regions (numbered 1–3) corresponding to allowed conformations.

Rotation of bonds in a polypeptide: Ramachandran Plot

Ramachandran plots can be constructed for polymers of each of the 20 amino acids.

It is significant to note that the Ramachandran plots for many amino acid residues are generally very similar, having only three regions with favourable or tolerated conformations (labelled 1–3 in the plot for poly-L-alanine in **Figure 6**).

Differences do occur. For instance, Proline and glycine are quite different from other amino acids in terms of allowed conformations because of their distinct R-group.

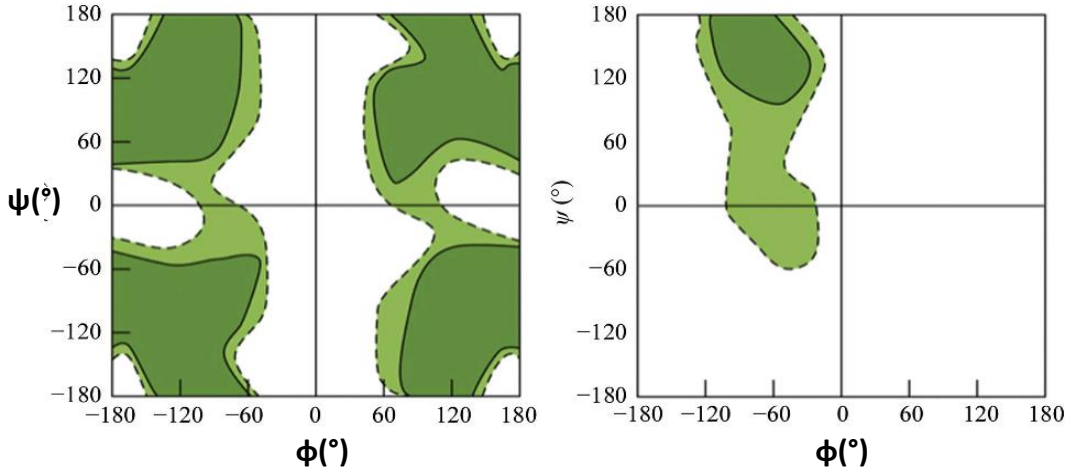


Figure 7: Ramachandran plots for glycine (left) and proline (right), showing the the allowed regions (continuous lines) and the partially allowed regions (dotted lines) (adapted from Ramakrishnan, 2001).

Similarly, if the side chain is branched, as in the case of threonine, it occupies more space close to the peptide backbone and restricts the approach of atoms in the neighbouring peptide groups. As a result, allowed conformations (ϕ and ψ angles) are more restricted for polypeptides of branched amino acids.

Polypeptide chain conformations

The various properties of peptides and proteins depend not only on their component amino acids and their bonding sequence in peptide chains, but also on the way in which the peptide chains are stretched, coiled and folded in space.

Because of their size, the orientational options for peptide and proteins seem nearly infinite. Fortunately, several factors act to narrow the structural options, and it is possible to identify some common structural themes or **secondary structures** that appear repeatedly in different protein molecules.

These conformational segments are described by the dihedral angles ϕ and ψ, defined in the previous figures. Five factors that influence the conformational equilibria of peptide chains are:

- The planarity of peptide bonds.
- Conformations are defined by dihedral angles ϕ and ψ.
- Steric crowding of neighboring groups.
- Repulsion and attraction of charged groups.
- The hydrophilic and hydrophobic character of substituent groups.

Due to the factors mentioned above the polypeptide chain can fold into following regular structures: 1. α helices, 2. β Sheets, 3. Turns and loops

Secondary structures: α helices

The α helix was proposed in 1951 by Linus Pauling and Robert Corey.

They considered the dimensions of peptide groups, possible steric constraints and opportunities for stabilization by formation of hydrogen bonds.

The structural features that define an alpha-helix are: the relative locations of the donor and acceptor atoms of the hydrogen bond, the number of amino acid units per helical turn and the distance the turn occupies along the helical axis.

The poly peptide backbone is tightly wound around an imaginary axis longitudinally through the middle of the helix, and the R-group of amino acid residues protrude outward from the helical backbone.

The perfect α helix have conformations with ϕ= -57° and ψ= -47°, and each helical turn includes 3.6 amino acid residues. The distance covered by the turn is 5.4 Å known as pitch.

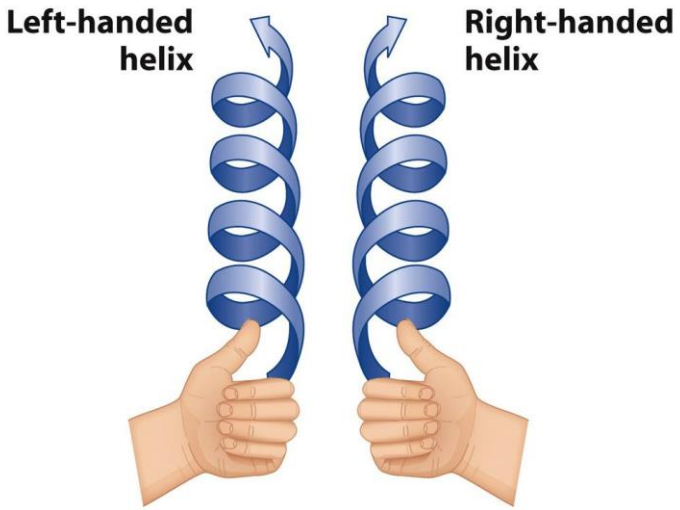
However, in natural proteins the values associated with α-helical conformations range from -57 to -70º for ϕ, and from -35 to -48º for ψ.

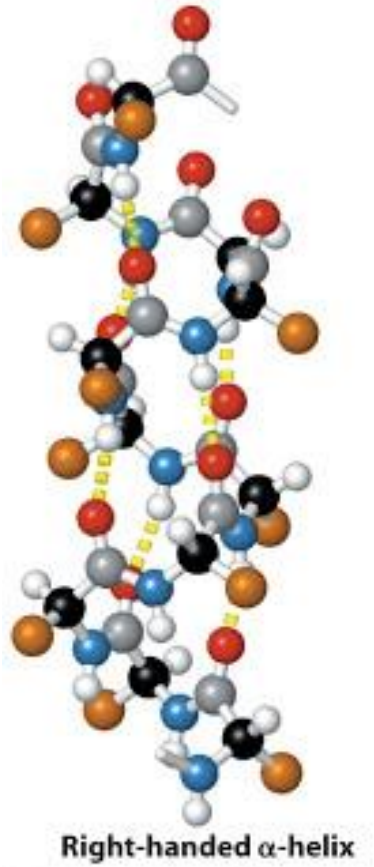
α helices

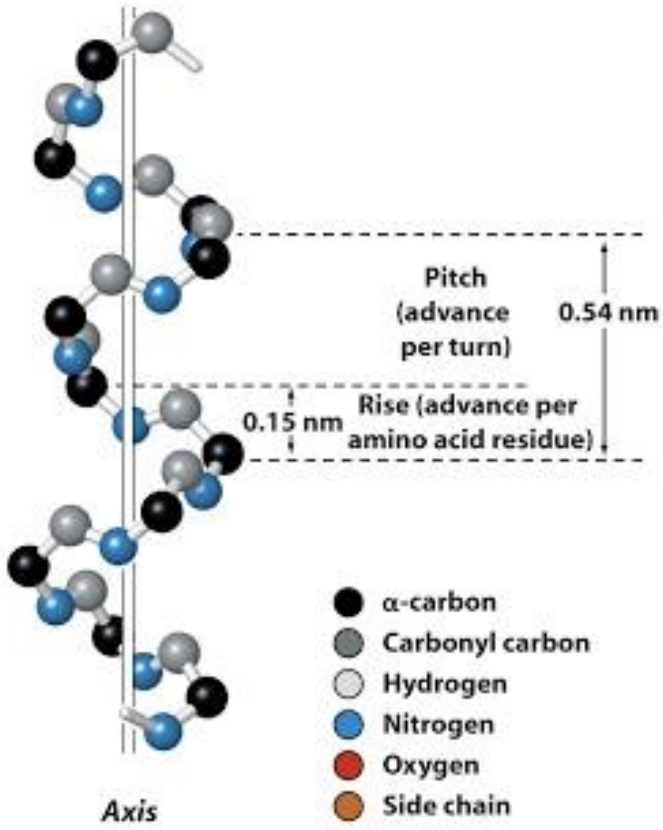
An α helix can be either a right- or a left-handed screw.

The Ramachandran diagram reveals that both the right-handed and the left- handed helices are among the allowed conformations. However, the right-handed helices are energetically more favourable because there is less steric clash between the side chains and the backbone.

Essentially all alpha helices found in proteins are right handed.







α helices: Ramachandran Plot

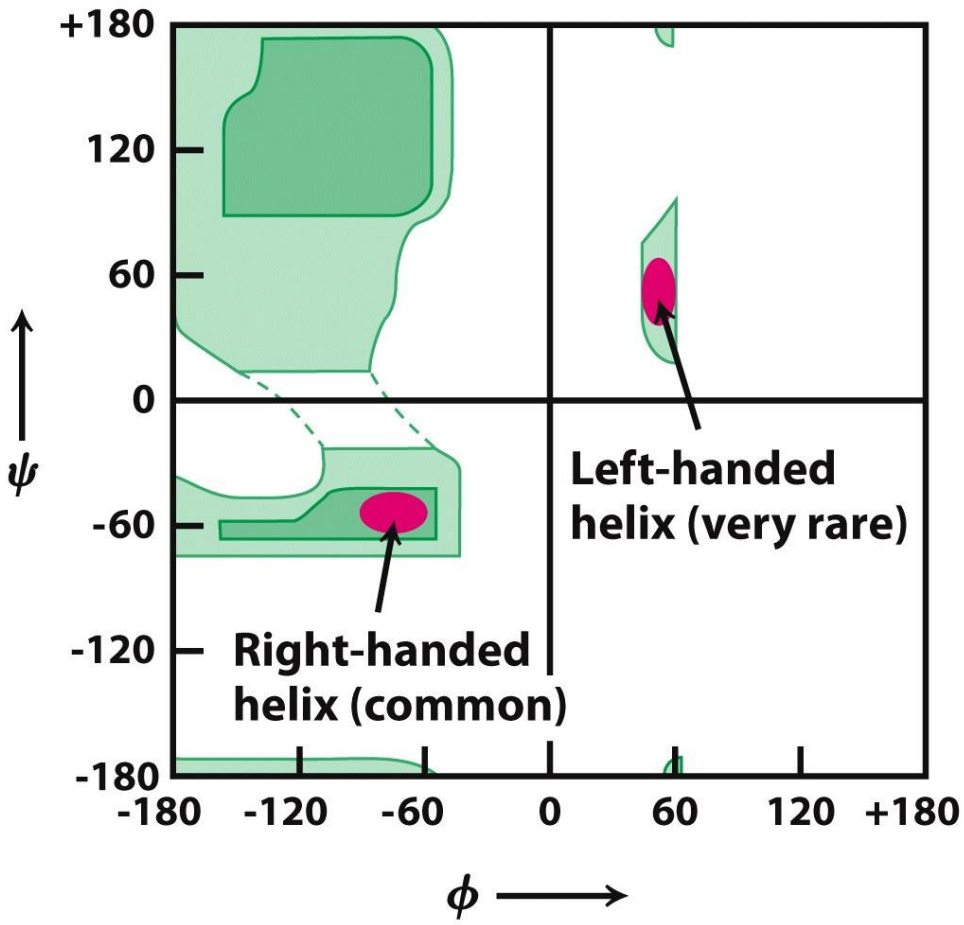


Figure 8: The Ramachandran diagram reveals that both the right-handed and the left-handed helices are among the allowed conformations.

α helices: amino acid preferences

Some amino acids are found in α helical conformations due to stability:

- Ala: small, uncharged R group and fits well into the α helical conformation.
- Tyr/Asn: bulky R group so they are less common.
- Gly: R group is a single H atom and destabilizes the structure since rotation around the Cα is unconstrained. Due to this reason, many α helices begin or end with Gly.
- Pro: least common residue in α helix because of the rigid cyclic side chain disrupts the helical strucutre by occupying space that a neighboring residue of the helix would otherwise occupy

Stability of α Helix

The α helix is stabilised by hydrogen bonds.

The hydrogen bonds are formed between hydrogen attached to electronegative nitrogen atom of the peptide linkage and the electronegative carbonyl oxygen atom of the fourth amino acid on the amino terminal side of the peptide bond.

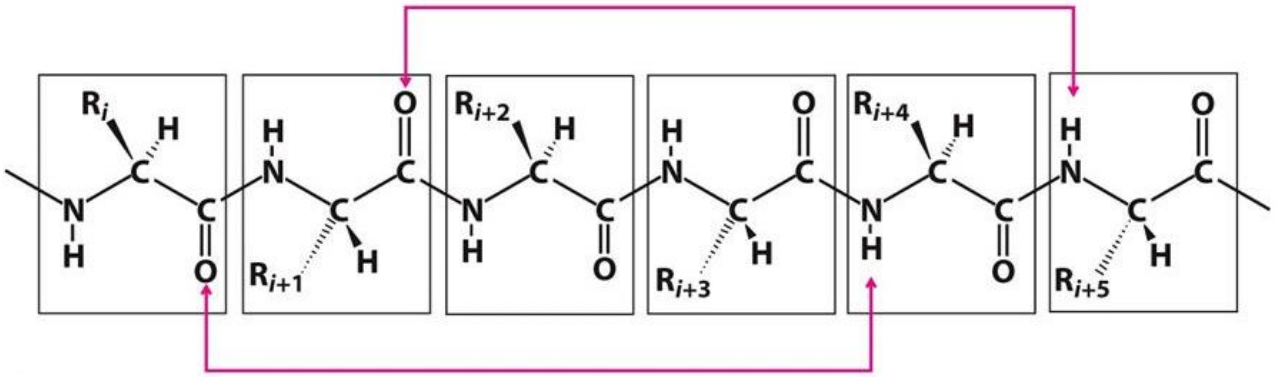


Figure 9: Hydrogen bonding pattern in alpha helix

Within the α helix every peptide bond participates in hydrogen bonding

All hydrogen bonds together provide the stability to the α helix

Distortions of α helices

The majority of alpha-helices in globular proteins are curved or distorted somewhat compared with the idealized alpha-helix model proposed by Pauling and Corey.

These distortions in the dihedral angles occurs when the helices are buried against other secondary structure elements in the core of the protein.

Proline residues also induce distortions of around 20° in the direction of the helix axis. This is because Proline cannot form a regular α-helix due to steric hindrance arising from its cyclic side chain which also blocks the main chain N atom and chemically prevents it forming a hydrogen bond.

Proline causes two H-bonds in the helix to be broken since the NH group of the following residue is also prevented from forming a good hydrogen bond.

Helices containing Proline are usually long because shorter helices would be destabilized by the presence of a Proline residue too much.

3_{10}-Helices.

3_{10}-Helices form a distinct class of helix but they are generally short and frequently occur at the termini of regular alpha-helices.

Each amino acid corresponds to a 120° turn in the helix (i.e., the helix has three residues per turn), and a translation of 2.0 Å (0.20 nm) along the helical axis, and has 10 atoms in the ring formed by making the hydrogen bond.

There are main chain hydrogen bonds between residues separated by three residues along the chain (i.e. O_i to N_{i+3}). In this nomenclature the Pauling-Corey alpha-helix is a 3.6_{13}-helix.

i + 3 → *i* hydrogen bonding **defines** a 3_{10}-helix.
i + 4 → *i* hydrogen bonding defines an α-helix.
i + 5 → *i* hydrogen bonding defines an π-helix.

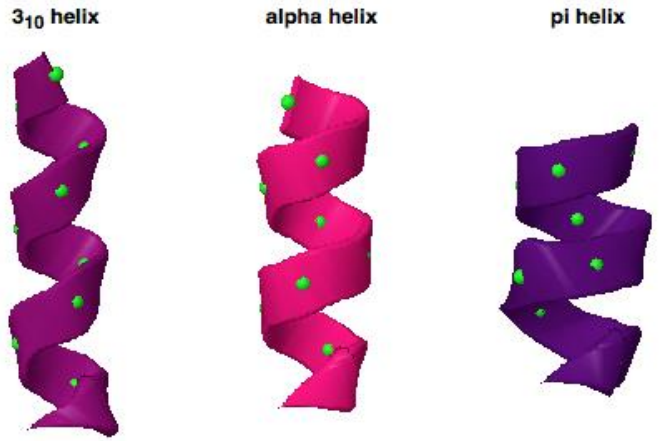


Figure 10: various types of helices

Secondary structure: β sheets

The other type of secondary structure Pauling and Corey discovered was the β sheet. The β conformations is an extended form of polypeptide. The backbone of the zigzag rather than helical structures.

The zigzag polypeptide chains can be arranged side by side to form a structure resembling a series of pleats called β sheets.

β sheets are composed of two or more polypeptide chains called β strands. Unlike the α helix, the β sheet is formed by hydrogen bonds ***between*** protein strands, rather than ***within*** a strand. The structure is stabilized by hydrogen bonds.

The R-groups of adjacent amino acids protrude from the zigzag structure in opposite directions creating the alternating patterns.

Bulky side-chain substituents destabilize this arrangement due to steric crowding, so this beta-sheet conformation is usually limited to peptides having a large amount of glycine and alanine.

Single β-strands are not energetically favorable. However, they can form β-sheets which are characterized by a pattern of hydrogen bonds between the residues on two different β-strands.

Secondary structure: β sheets

By convention, beta-sheets are designated by broad arrows or cartoons, pointing in the direction of the C-terminus.

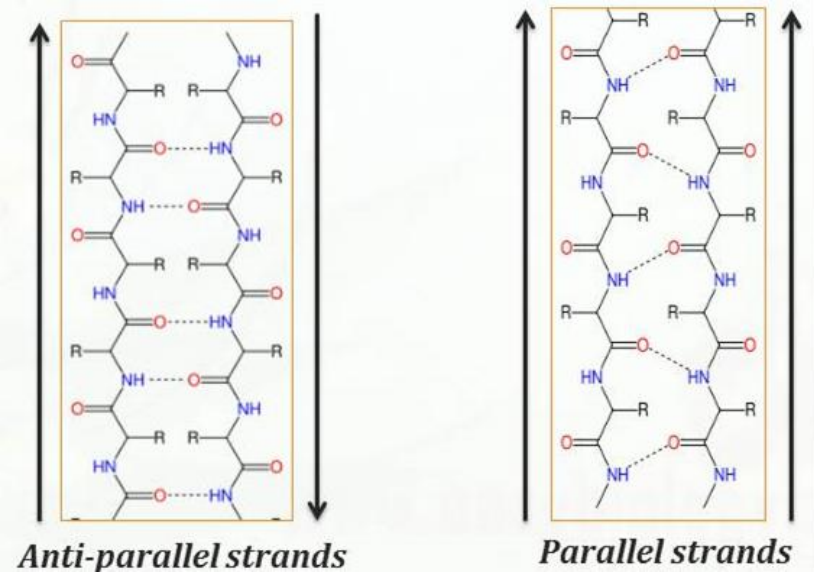
The adjacent polypeptide chain in a β sheet can be either parallel or antiparallel (having the same or opposite amino-to-carboxyl orientations, respectively).

The idealized structures corresponds to ϕ= -119° and ψ= +113 (parallel) and ϕ=-139, ψ=+135 (antiparallel); these values vary somewhat in real proteins, resulting into some structural variations.

Two types of β conformations organises polypeptide chains into sheets.

a) anti-parallel β sheets

b) Parallel β sheets



Antiparallel β sheets are slightly more stable than parallel β sheets because the hydrogen bonding pattern is more optimal.

Secondary structure: β sheets

The axial distance between adjacent residues is 3.5 Å in anti-parallel β sheet. There are two residues per repeat unit which gives the beta-strand a 7Å pitch. This compares with the alpha-helix where the axial distance between adjacent residues is only 1.5Å. Clearly, polypeptides in the beta-conformation are far more extended than those in the alpha-helical conformation.

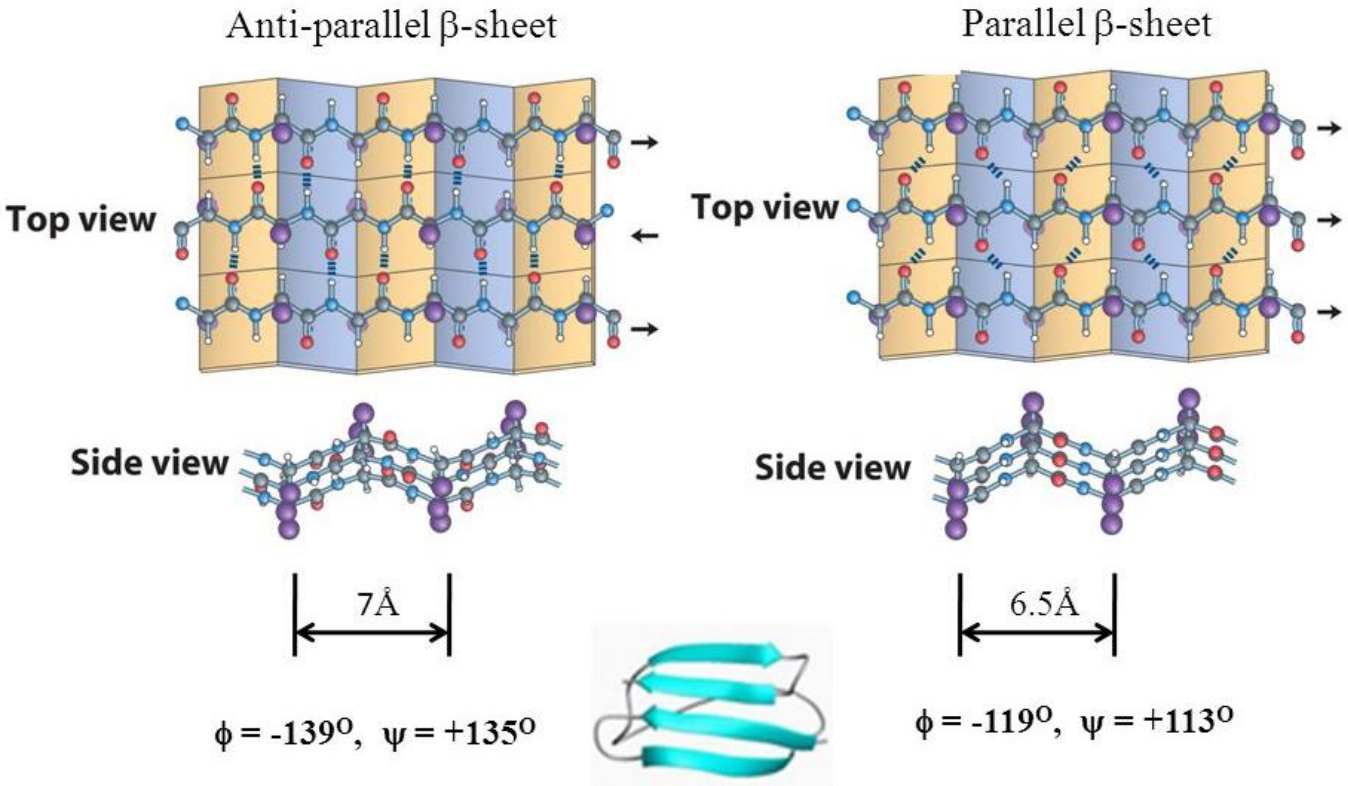


Figure 11: Properties of β sheets

β strand: Ramachandran Plot

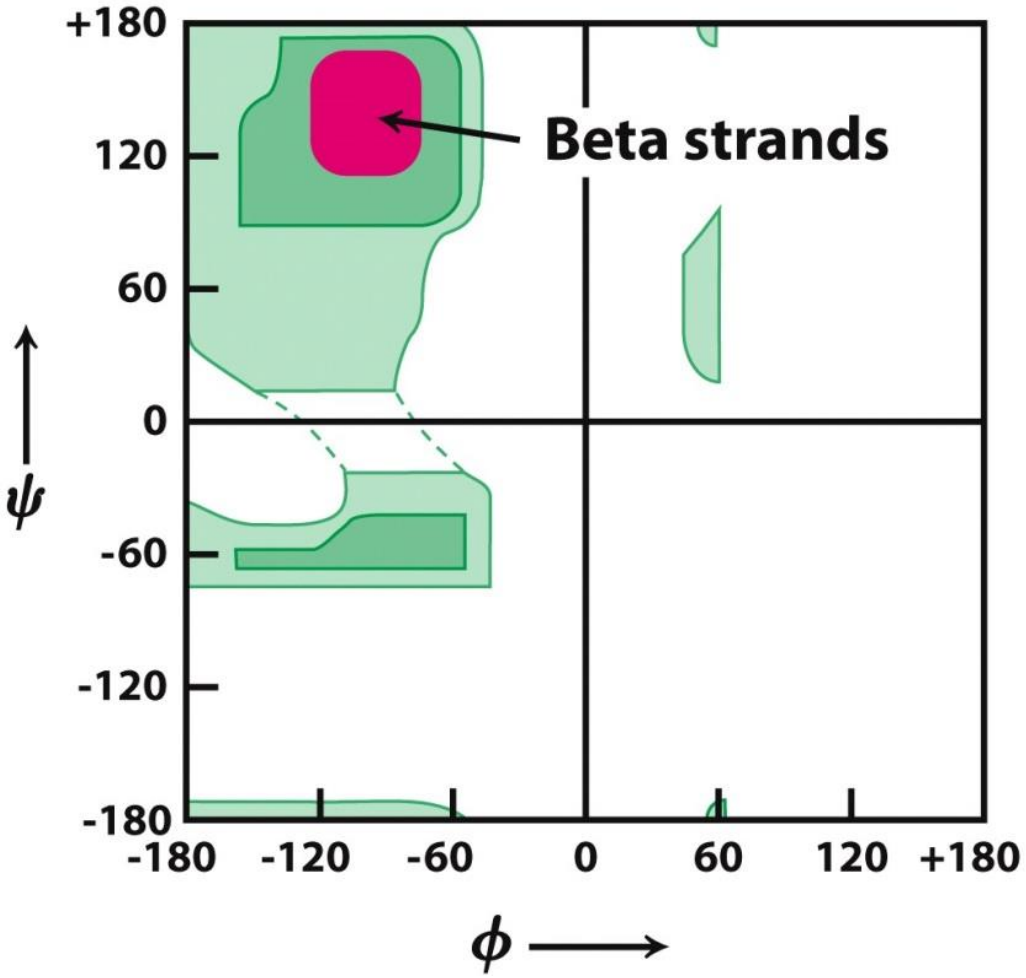


Figure 12: Ramachandran diagram for β strands. The red areas show the sterically allowed conformations of extended, β strand-like structures

Secondary structure: β turns and Ω loops

Most proteins have compact folded structures and nearly one-third of the amino acid residues are in turns or loops where polypeptide chain reverse directions.

These are connecting elements that links successive runs of α helix and β stands.

The most common structural element is called β turn or hairpin turn or reverse turn. In many reverse turn the CO group of first residue of a polypeptide is hydrogen bonded to NH group of fourth. This interaction stabilises abrupt changes in the direction of polypeptide chain.

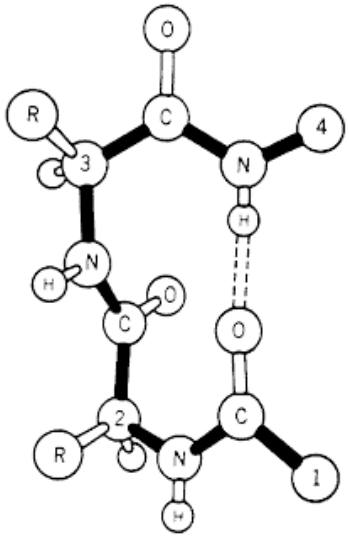


Figure 13: Turn structure

The more elaborate structures comprises the Ω loops. The loops do not have regular, periodic structures. However, loop structures are often rigid and well defines.

Turns and loops are mostly present on the surface of the proteins and thus often participates in interactions between proteins and other molecules.

Secondary structure: β turns

A β turn is a region of the protein involving four consecutive residues where the polypeptide chain folds back on itself by nearly 180 degrees.

β turns helps in protein compaction, since the hydrophobic amino acids tends to be in the interior of the protein, while the hydrophilic residues interacts with the aqueous environment. Therefore, β turn reversals give a protein its globularity rather than linearity.

There are two most common subtype (Types I and II) of β turns which is defined by the ϕ and ψ angles of the middle two residues. The essential difference between them being the orientation of the peptide bond between residues at 2nd and 3rd positions in the loop.

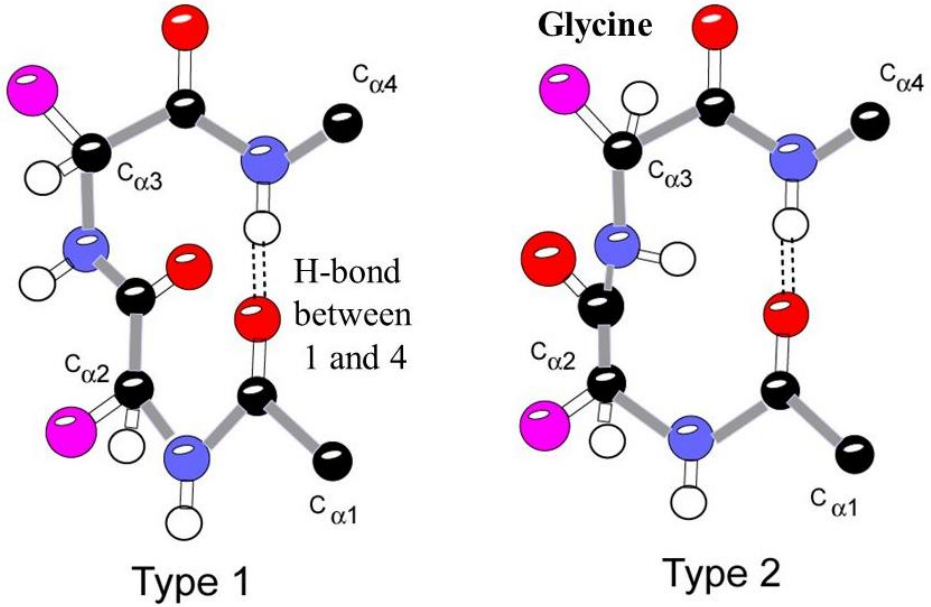
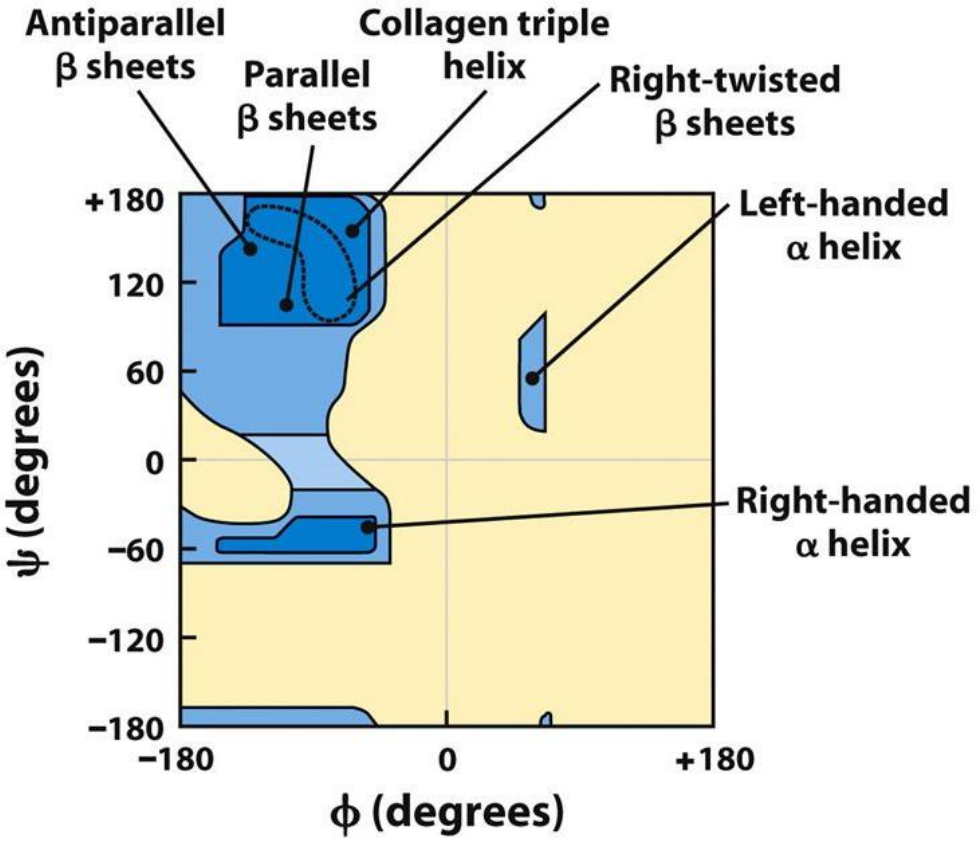
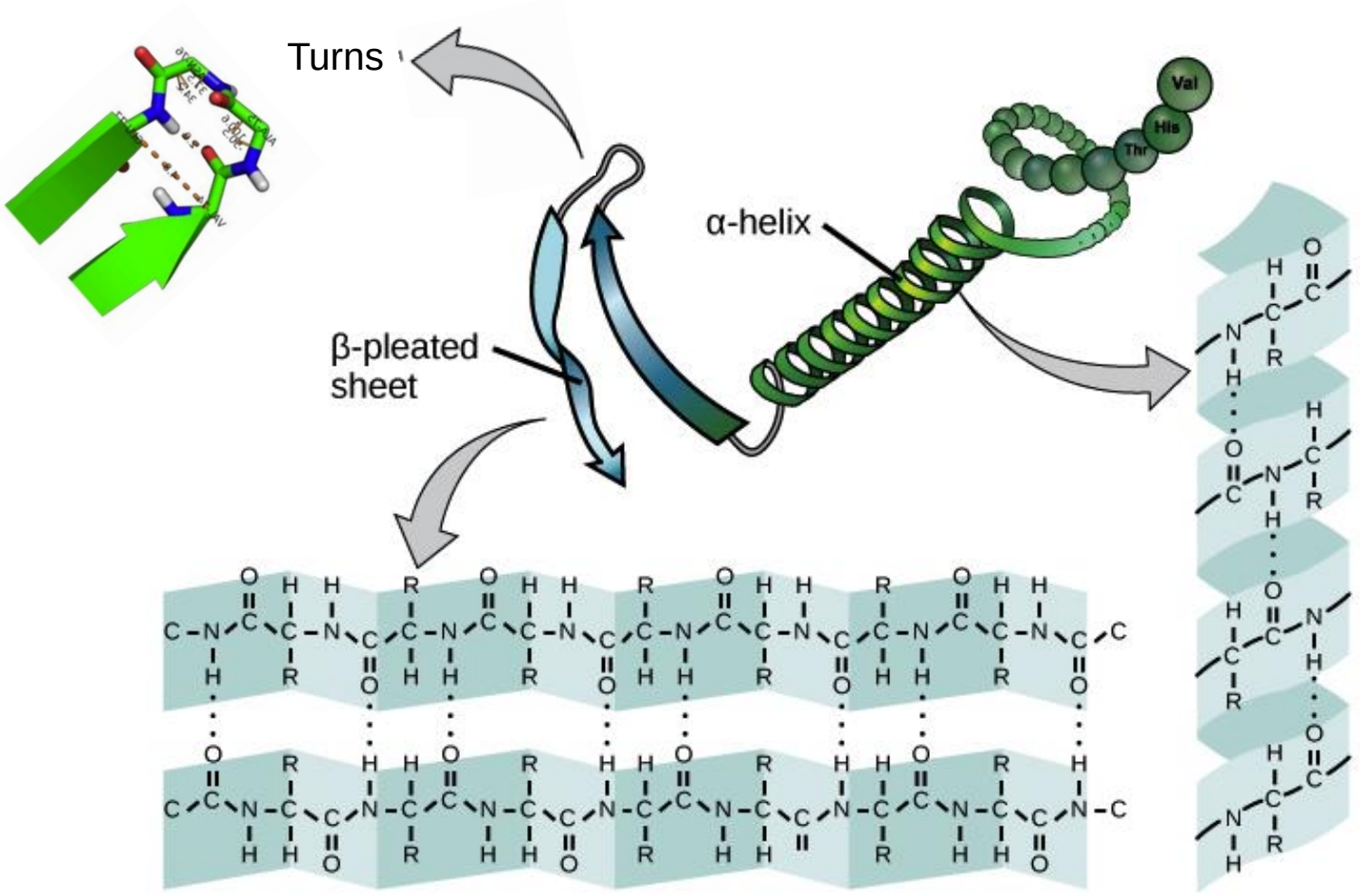


Figure 14: types of β turns

Ramachandran Plot
Showing the position of various secondary structures



Summary



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