COURSE : SC202 (CHEMISTRY)

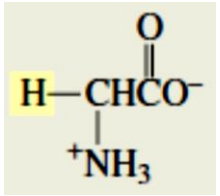
DR. SANGITA TALUKDAR

LECTURE-7

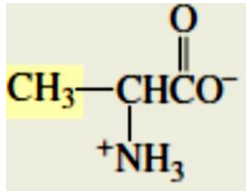
DATE: 21.1.2021

Classification of Amino Acids

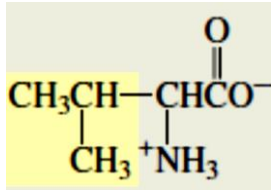
Achiral amino acid



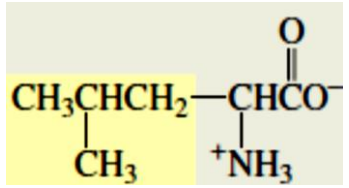
Glycine(Gly)



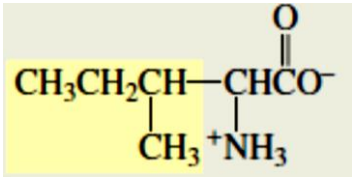
Alanine(Ala)



Valine(Val)*

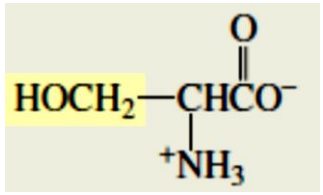


Leucine (Leu)*

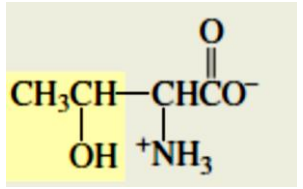


Isoleucine(Ile)*

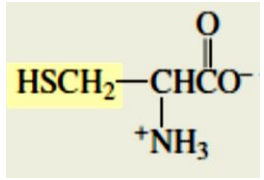
Aliphatic side chain amino acids



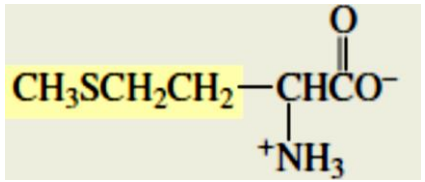
Serine (Ser)



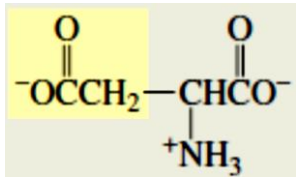
Threonine(Thr) *



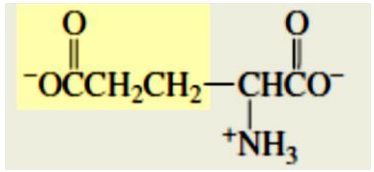
Cysteine(Cys)



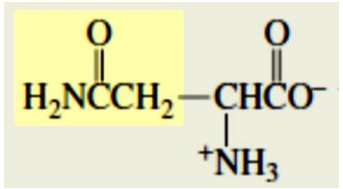
Methionine(Met)*



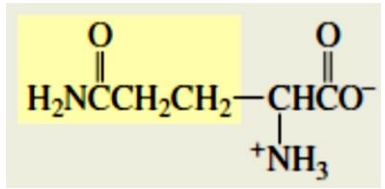
Aspartic acid(Asp)



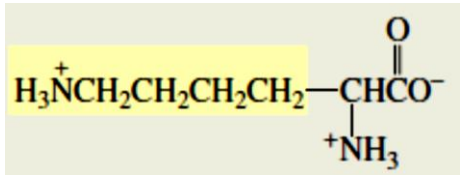
Glutamic acid (Glu)



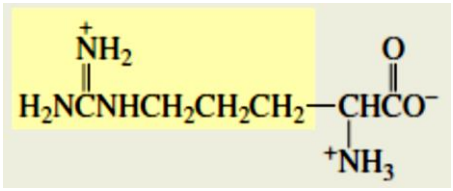
Asparagine(Asn)



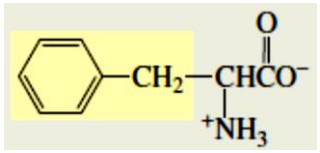
Glutamine(Gln)



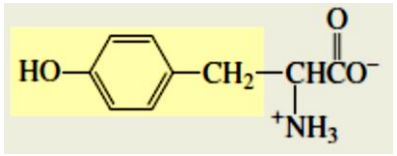
Lysine (Lys) *



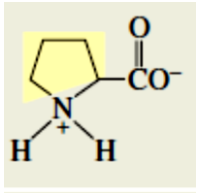
Arginine(Arg) *



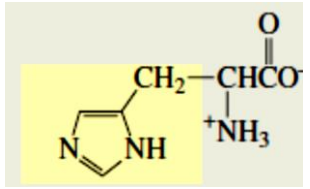
Phenylalanine(Phe) *



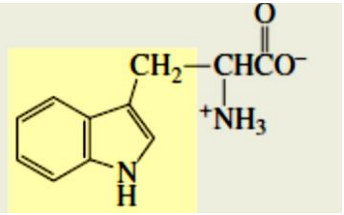
Tyrosine(Tyr)



Proline(Pro)



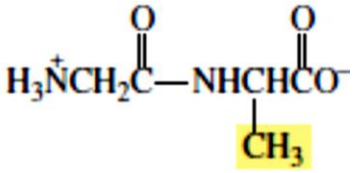
Histidine(His)*



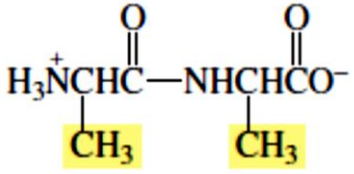
Trytophan(Trp)*

Strategy of Peptide Bond Synthesis: N-Protection and C-Activation

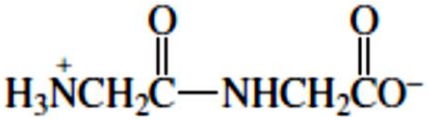
Suppose you wanted to make the dipeptide Gly-Ala. That dipeptide is only one of four possible dipeptides that could be formed from alanine and glycine.



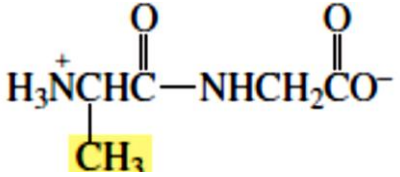
Gly-Ala



Ala-Ala



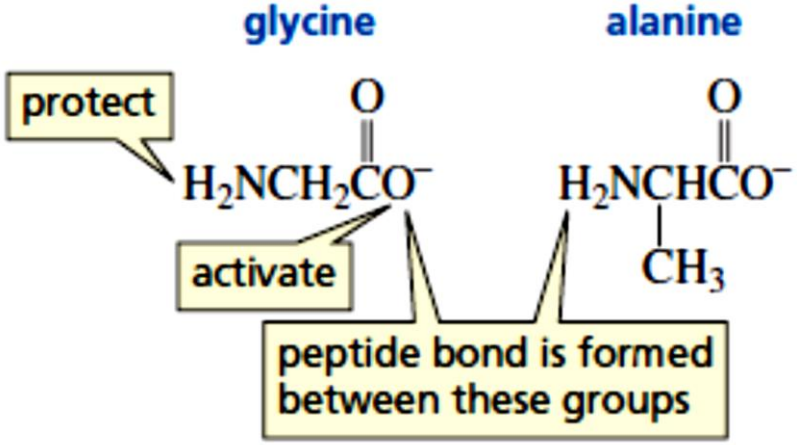
Gly-Gly



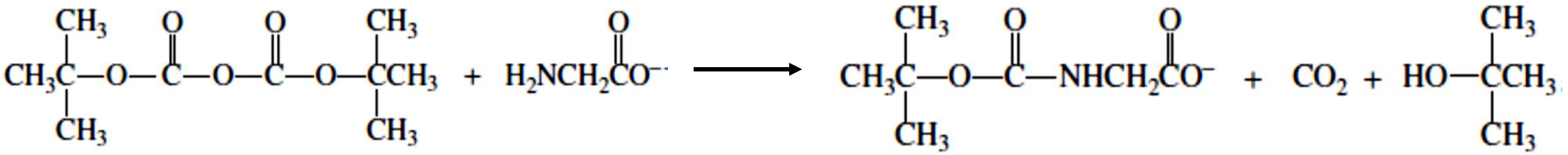
Ala-Gly

Solid-phase peptide synthesis: A method in which the C-terminal amino acid is attached to a solid support (polystyrene beads) and the peptide is synthesized in the C→N direction by successive coupling of protected amino acids. When the peptide is complete, it is cleaved from the solid support.

Solution-phase peptide synthesis (classical peptide synthesis): Any of several methods in which protected amino acids are coupled in solution in the correct sequence to give a desired peptide. Most of these methods proceed in the N→C direction

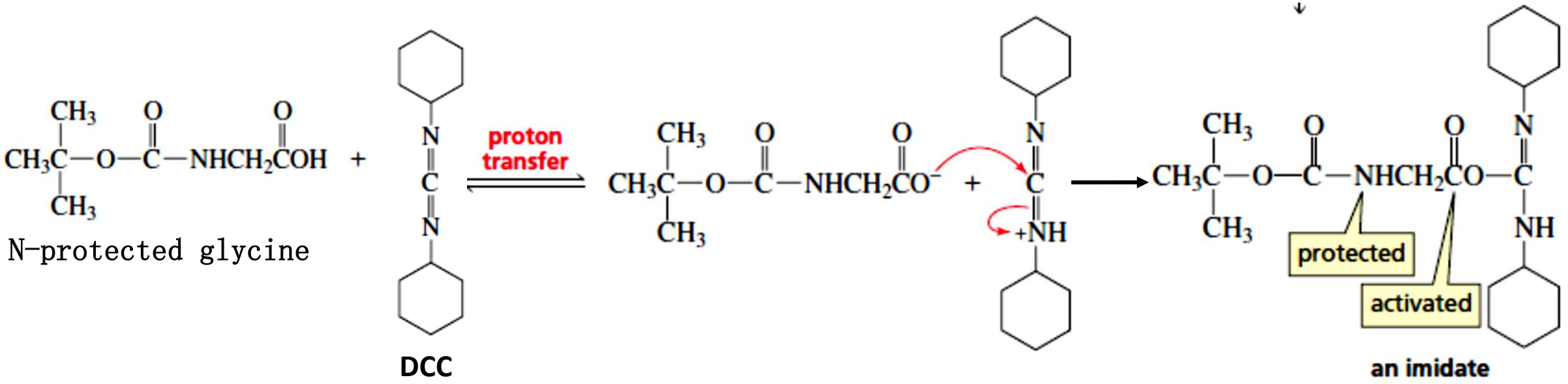


1. The reagent that is most often used to protect the amino group of an amino acid is di-*tert*-butyl dicarbonate. The protecting group is known by the acronym *t*-BOC.



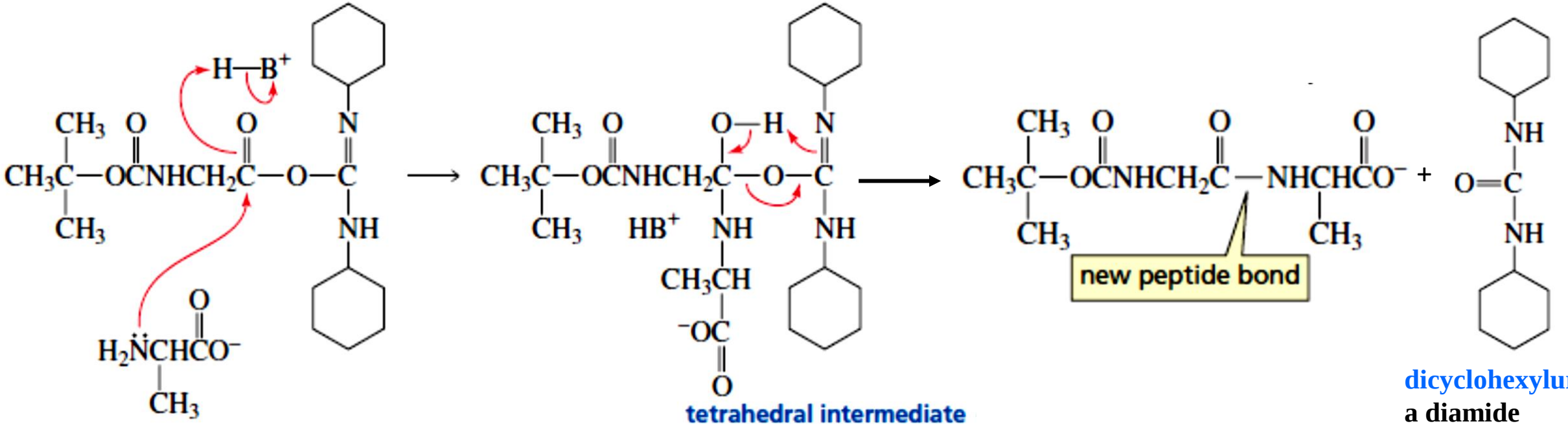
di-*tert*-butyl dicarbonate glycine N-protected glycine

2. The preferred method for activating the carboxyl group of an N-protected amino acid is to convert it into an imidate using dicyclohexylcarbodiimide (DCC).

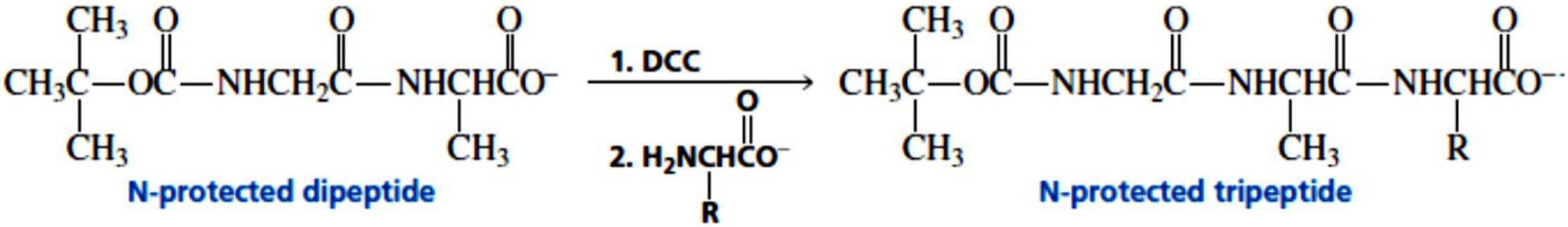


N-protected glycine DCC an imidate

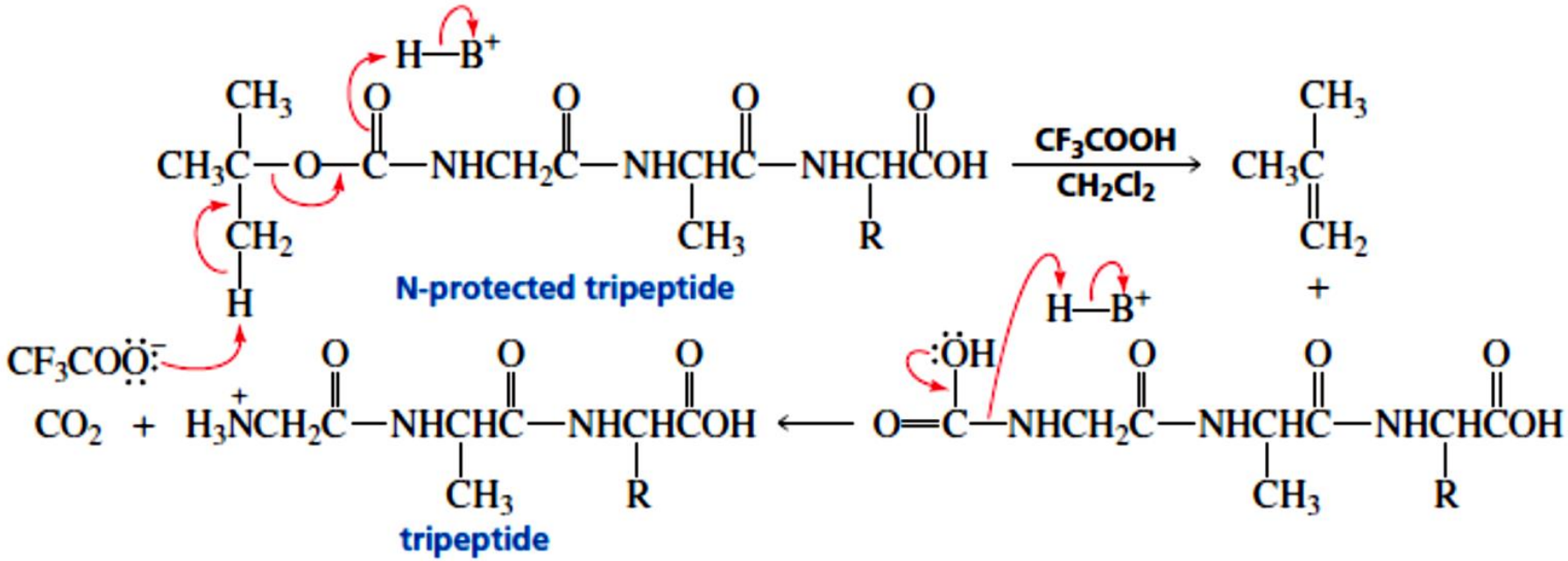
3. After the amino acid has its N-terminal group protected and its C-terminal group activated, the second amino acid is added to form the new peptide bond.



4. Amino acids can be added to the growing C-terminal end by repeating these two steps: activating the carboxyl group of the C-terminal amino acid of the peptide by treating it with DCC and then adding a new amino acid.



When the desired number of amino acids has been added to the chain, the protecting group on the N-terminal amino acid is removed. *t*-BOC is an ideal protecting group because it can be removed by washing with trifluoroacetic acid and methylene chloride. The by-products isobutylene and carbon dioxide are gases so they escape, driving the reaction to completion.



After each step of the synthesis, the peptide must be purified to prevent subsequent unwanted reactions with leftover reagents

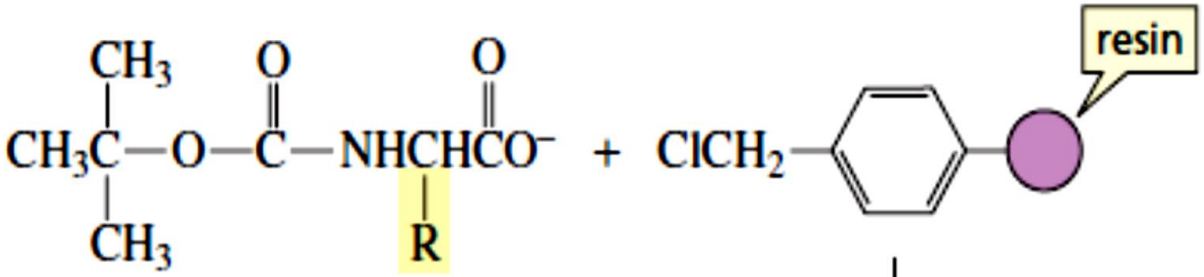
The overall yield of a nonapeptide such as bradykinin would be only 17%

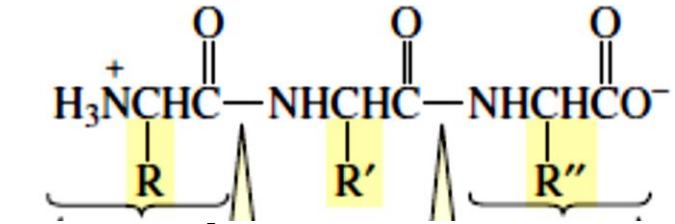
Number of amino acids	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
Overall yield	80%	64%	51%	41%	33%	26%	21%	17%

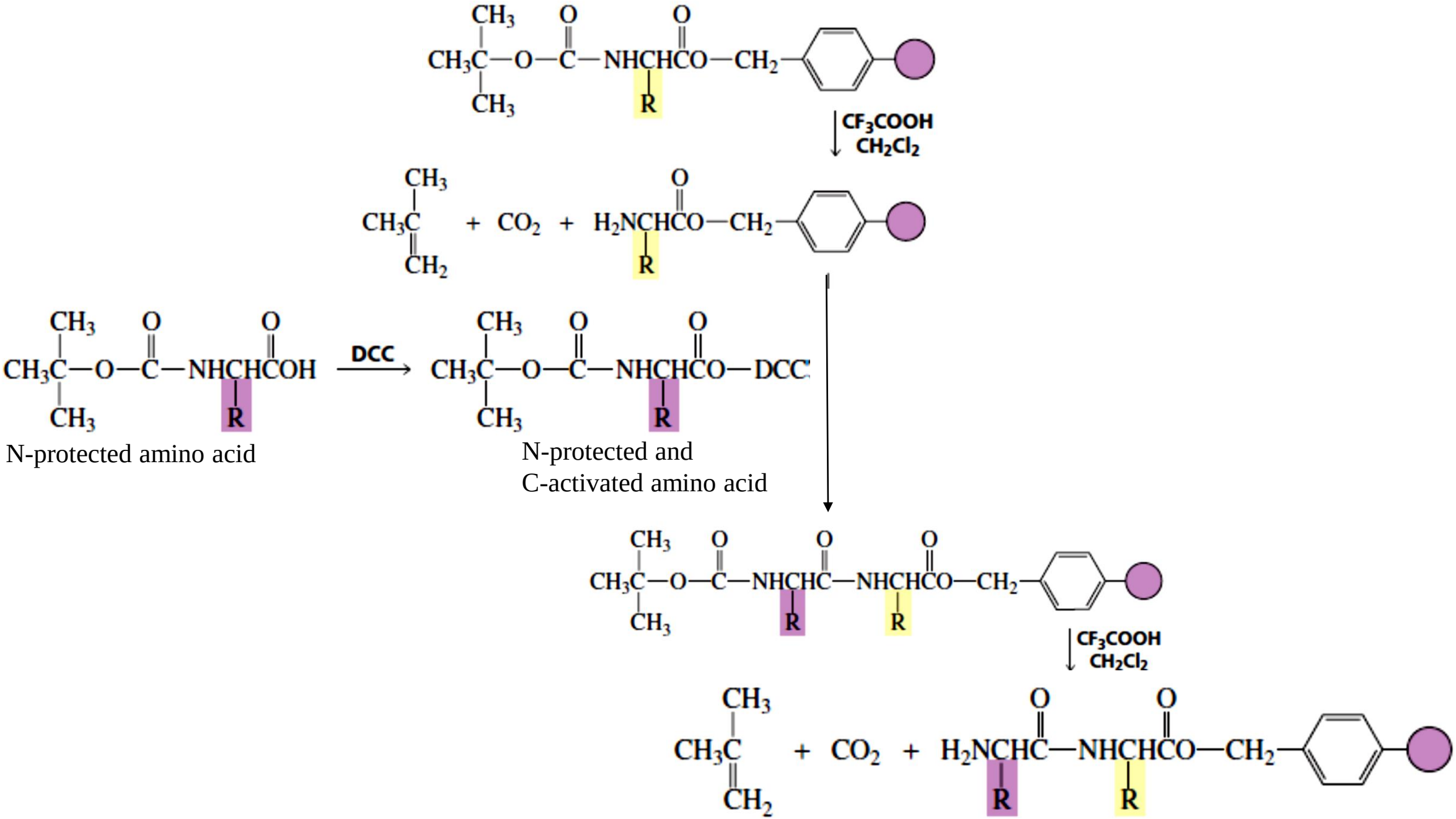
Automated Peptide Synthesis

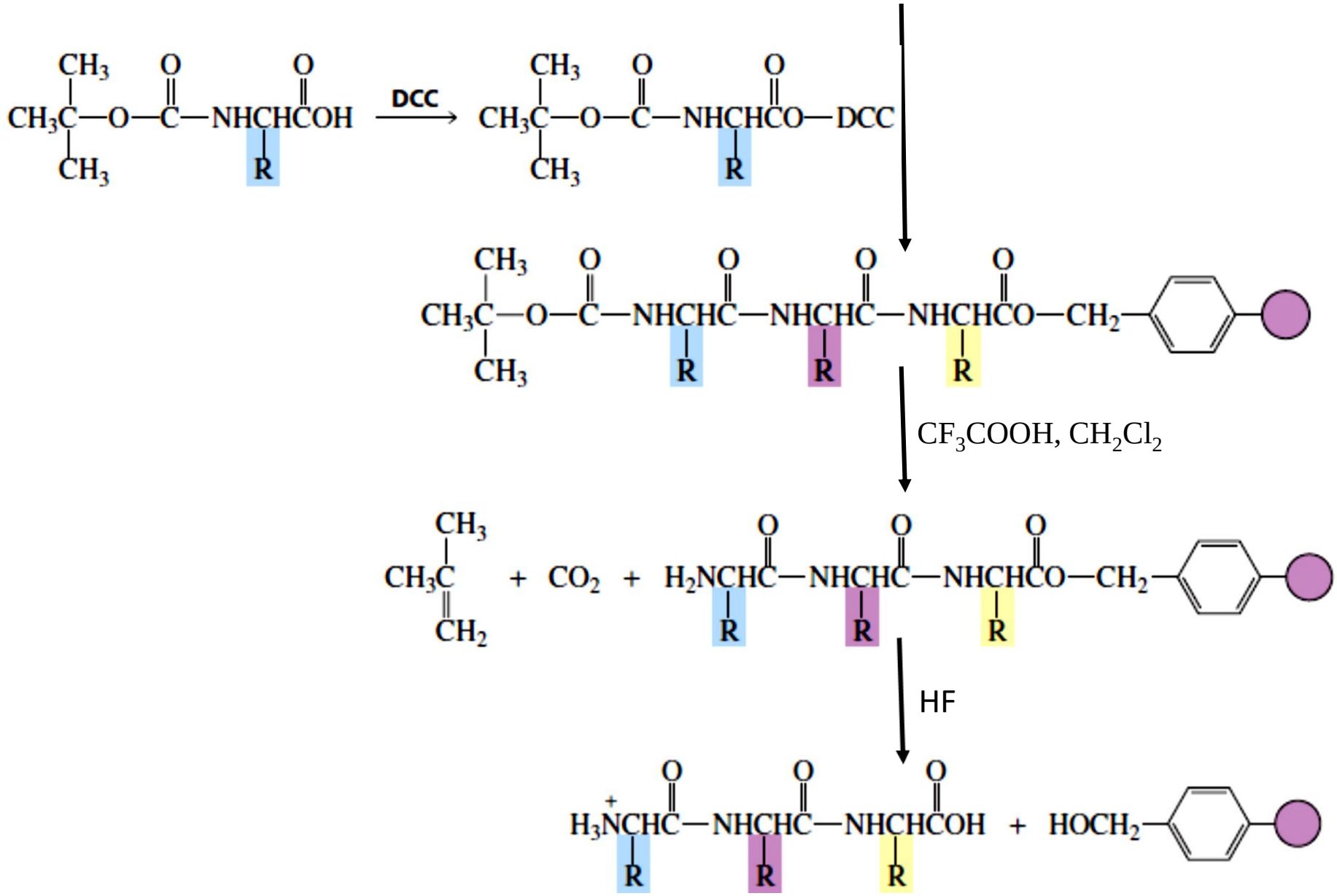
In 1969, Bruce Merrifield described a method that revolutionized the synthesis of peptides because it provided a much faster way to produce peptides in much higher yields. Using this technique, bradykinin was synthesized with an 85% yield in 27 hours.

In the Merrifield method, the C-terminal amino acid is covalently attached to a solid support contained in a column. Each N-terminal blocked amino acid is added one at a time, along with other needed reagents, so the protein is synthesized from the C-terminal end to the N-terminal end. Because it uses a solid support and is automated, Merrifield's method of protein synthesis is called **automated solid-phase peptide synthesis**.






CF3COOH
CH2Cl2
DCC
N-protected amino acid
N-protected and
C-activated amino acid
CF3COOH
CH2Cl2


DCC
CF3COOH, CH2Cl2
HF

A huge advantage of the Merrifield method of peptide synthesis is that the growing peptide can be purified by washing the column with an appropriate solvent after each step of the procedure. The impurities are washed out of the column because they are not attached to the solid support. Since the peptide is covalently attached to the resin, none of it is lost in the purification step, leading to **high yields of purified product**.

Protein Structure

Protein molecules are described by several levels of structure.
The **primary structure** of a protein is the sequence of amino acids in the chain and the location of all the disulfide bridges.
The **secondary structure** describes the regular conformation assumed by segments of the protein's backbone. In other words, the secondary structure describes how local regions of the backbone fold.
The **tertiary structure** describes the three-dimensional structure of the entire polypeptide. If a protein has more than one polypeptide chain, it has quaternary structure.
The **quaternary structure** of a protein is the way the individual protein chains are arranged with respect to each other.

Proteins can be divided roughly into two classes.

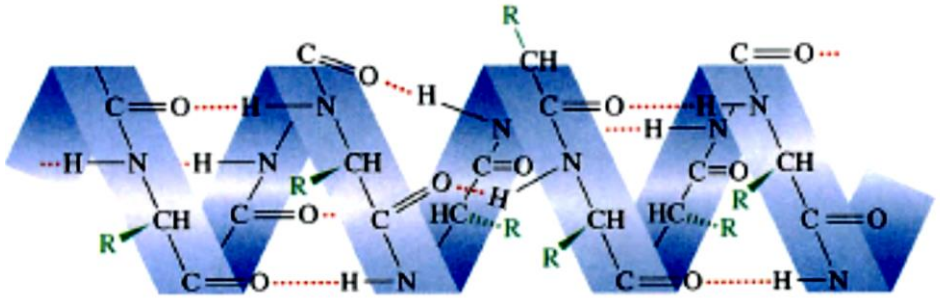
Fibrous proteins contain long chains of polypeptides that occur in bundles. These proteins are insoluble in water. Eg. keratin and collagen, are fibrous proteins.
Globular proteins are soluble in water and tend to have roughly spherical shapes. Essentially all enzymes are globular proteins.

Secondary Structure of Proteins

Secondary structure describes the conformation of segments of the backbone chain of a peptide or protein. To minimize energy, a polypeptide chain tends to fold in a repeating geometric structure such as an α –helix or a β pleated sheet.

α-Helix

In an α-helix the backbone of the polypeptide coils around the long axis of the protein molecule. The helix is stabilized by hydrogen bonds. When a peptide chain winds into a helical coil, each carbonyl oxygen can hydrogen-bond with an N -H hydrogen on the next turn of the coil.

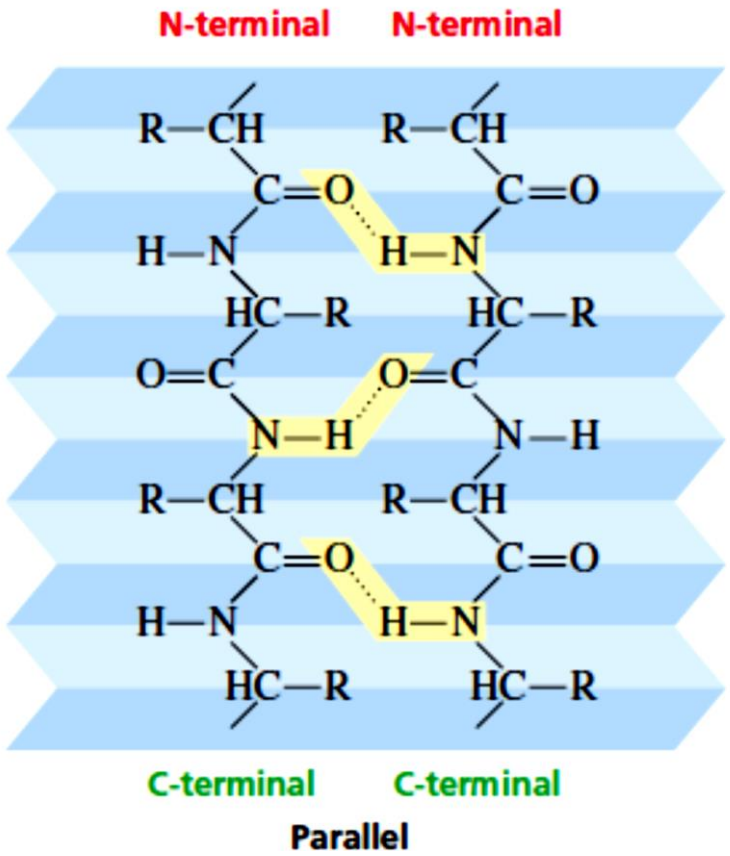


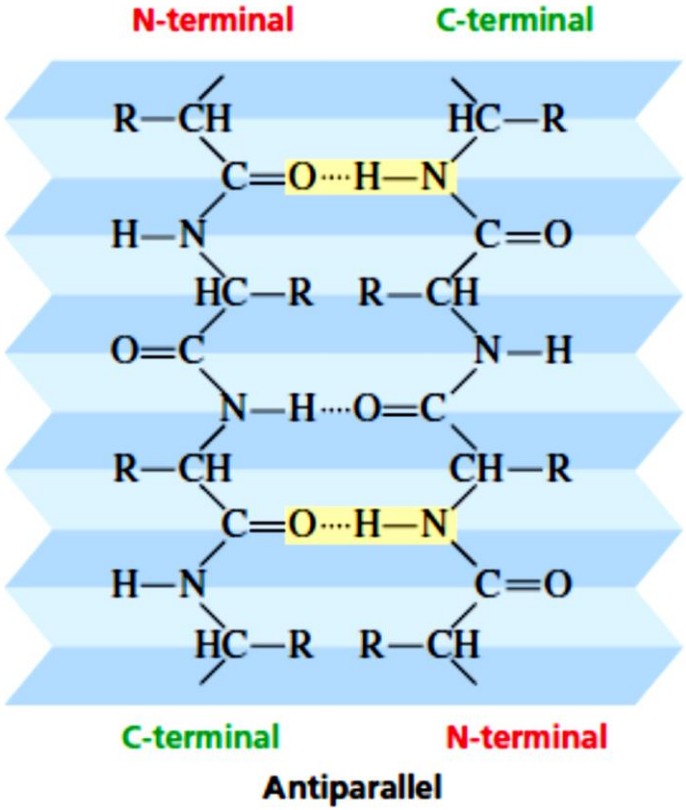
Because the amino acids have the L-configuration, the α-helix is a right-handed helix; Each turn of the helix contains 3.6 amino acid residues, and the repeat distance of the helix is 5.4 Å.

- Wool and the fibrous protein of muscle are examples of proteins with secondary structures that are almost all α-helices Consequently, these proteins can be stretched.
- In contrast, the secondary structures of silk and spider webs are predominantly β-pleated sheets. Because the β-pleated sheet is a fully extended structure, these proteins cannot be stretched.

β-Pleated Sheet

The second type of secondary structure is the β-pleated sheet. In a β-pleated sheet, the polypeptide backbone is extended in a zigzag structure resembling a series of pleats. A sheet is almost fully extended—the average two-residue repeat distance is 7.0 Å. The hydrogen bonding in a sheet occurs between neighboring peptide chains. The adjacent hydrogen-bonded peptide chains can run in the same direction or in opposite directions. In a **parallel sheet**, the adjacent chains run in the same direction. In an **antiparallel sheet**, the adjacent chains run in opposite directions.

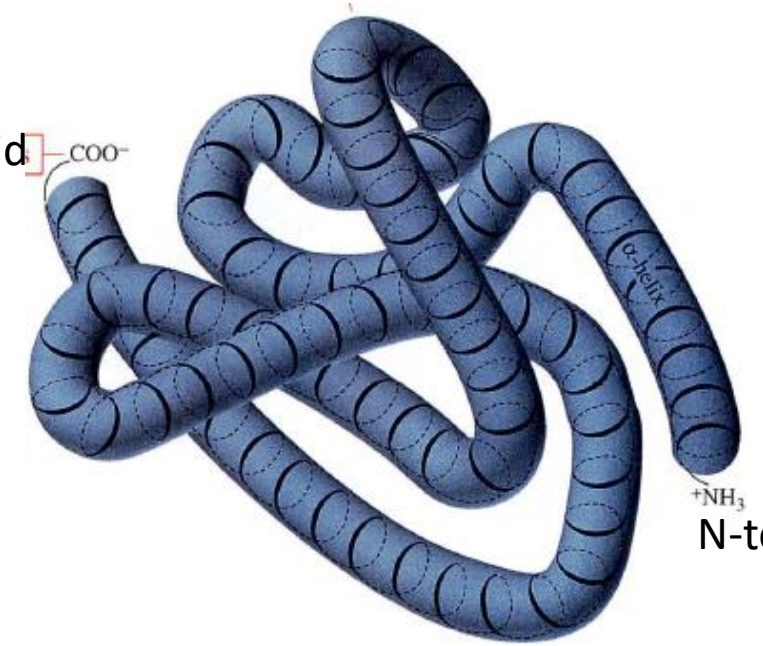




Silk, for example, a protein with a large number of relatively small amino acids (glycine and alanine), has large segments of β-pleated sheets.

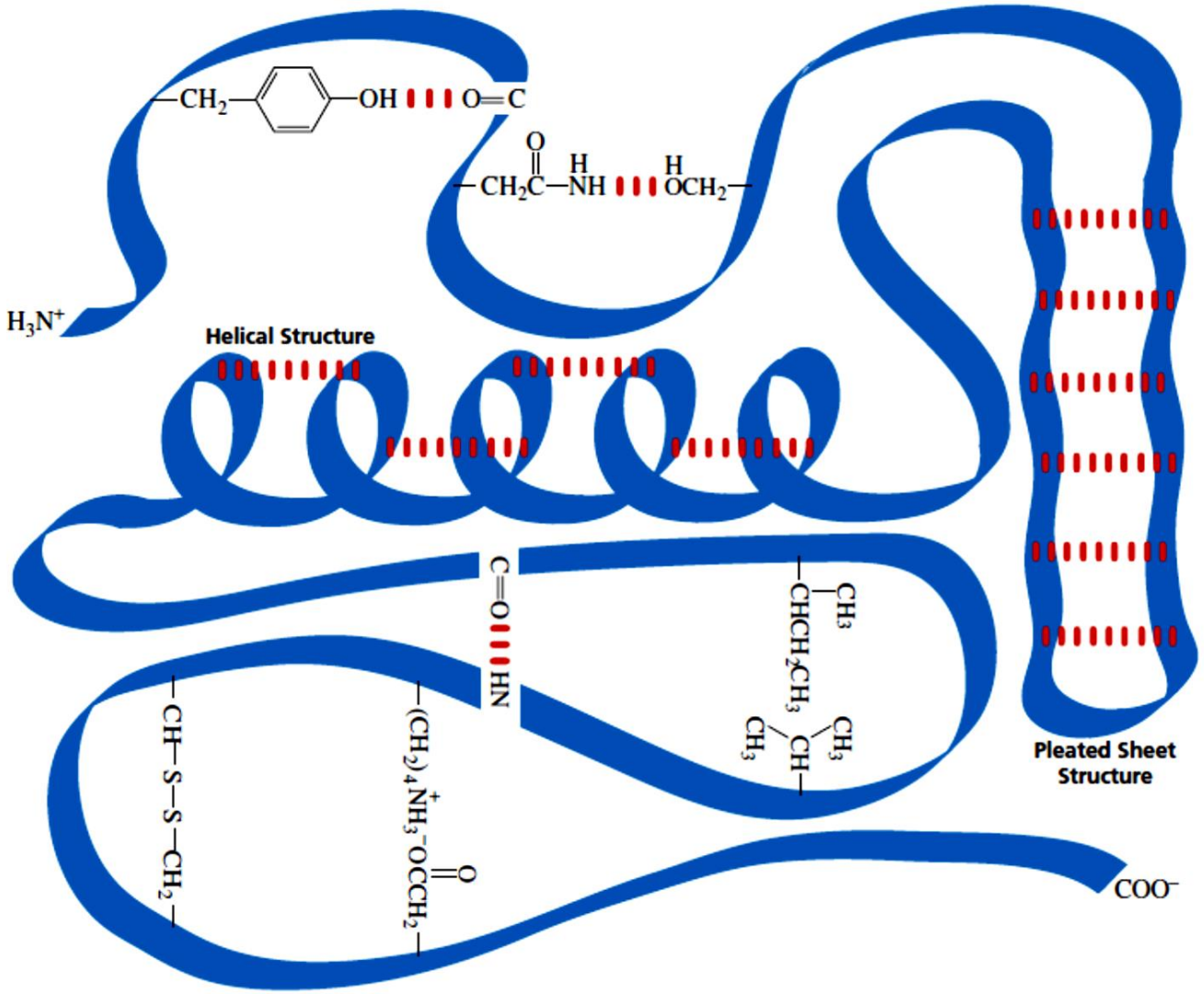
Tertiary Structure of Proteins

C-terminal amino acid



N-terminal amino acid

- The *tertiary structure* of a protein is the three-dimensional arrangement of all the atoms in the protein. Proteins fold spontaneously in solution in order to maximize their stability.
- The stabilizing interactions include covalent bonds, hydrogen bonds, electrostatic attractions (attractions between opposite charges), and hydrophobic (van der Waals) interactions. Stabilizing interactions can occur between peptide groups (atoms in the backbone of the protein), between side-chain groups and between peptide and side-chain groups.
- Disulfide bonds are the only covalent bonds that can form when a protein folds.
- The interactions between nonpolar groups are known as **hydrophobic interactions**

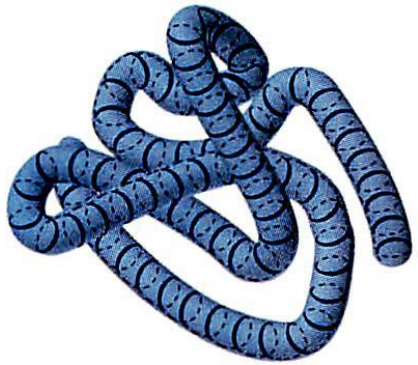


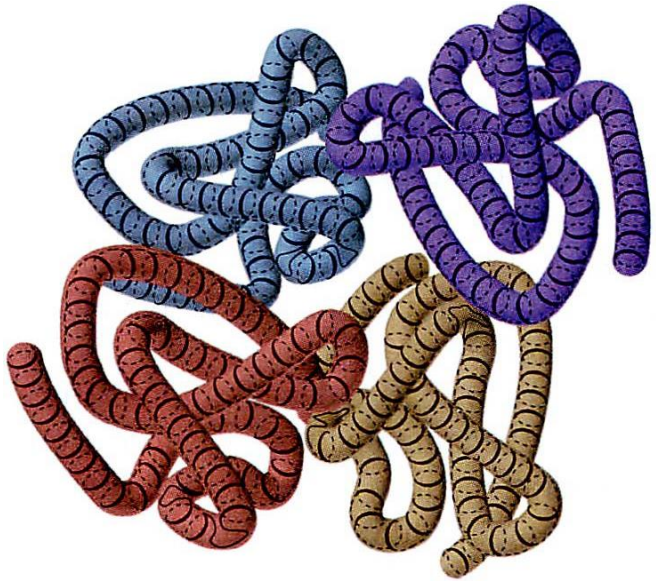
Coiling of an enzyme can give three-dimensional shapes that produce important catalytic effects. Polar, hydrophilic (water-loving) side chains are oriented toward the outside of the globule. Nonpolar, hydrophobic (water-hating) groups are arranged toward the interior. Coiling in the proper conformation creates an enzyme's active site, the region that binds the substrate and catalyzes the reaction.

Quaternary Structure of Proteins

The quaternary structure of a protein describes the way the subunits are arranged in space. The subunits are held together by hydrophobic interactions, hydrogen bonding, and electrostatic attractions.

Proteins that have more than one peptide chain are called **oligomers**. The individual chains are called **subunits**. A protein with a single subunit is called a *monomer*, one with two subunits is called a *dimer*; one with three subunits is called a *trimer*, and one with four subunits is called a *tetramer*. Hemoglobin is an example of a tetramer.





Protein Denaturation

Destroying the highly organized tertiary structure of a protein is called **denaturation**. Anything that breaks the bonds responsible for maintaining the three-dimensional shape of the protein will cause the protein to denature (unfold). The totally random conformation of a denatured protein is called a **random coil**.

The following are some of the ways that proteins can be denatured:

• Changing the pH denatures proteins because it disrupts electrostatic attractions and hydrogen bonds.

• Certain reagents such as urea and guanidine hydrochloride denature proteins.

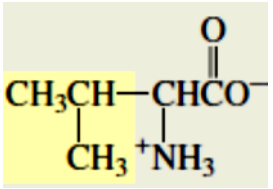
• Detergents such as sodium dodecyl sulfate denature proteins by associating with the nonpolar groups of the protein, thus interfering with the normal hydrophobic interactions.

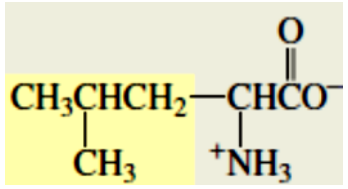
• Organic solvents denature proteins by disrupting hydrophobic interactions.

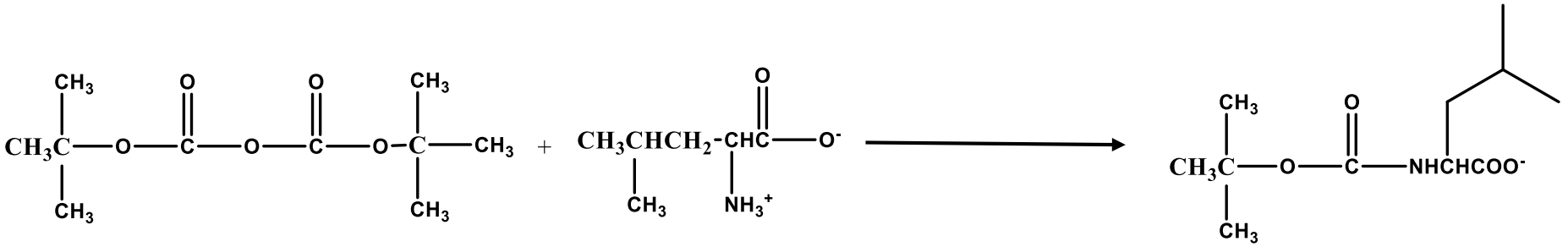
• Proteins can also be denatured by heat or by agitation. Example is the change that occurs to the white of an egg when it is heated or whipped.

1.What dipeptides would be formed by heating a mixture of valine and N-protected leucine?

Ans:







2. Show the steps in the synthesis of the tetrapeptide Leu-Phe-Lys-Val.

3. What would be the overall yield of a peptide containing 15 amino acid residues if the yield for the incorporation of each is 80%?

4. Suppose you are trying to synthesize the dipeptide Val-Ser. Compare the product that would be obtained if the carboxyl group of N-protected valine were activated with thionyl chloride with the product that would be obtained if the carboxyl group were activated with DCC.

5. Which of the following water-soluble proteins would have the greatest percentage of polar amino acids—a spherical protein, a cigar-shaped protein, or a subunit of a hexamer?

b. Which of these would have the smallest percentage of polar amino acids?

Ans : (Hint)Most proteins exist in aqueous environments. Therefore, they tend to fold in away that exposes the maximum number of polar groups to the aqueous environment and that buries the nonpolar groups in the interior of the protein, away from water.