

06.04.17

Because of this, the C-N bond length is also shorter than normal C-N single bonds. The peptide unit which is made up of the C<sub>α</sub>-NH atoms is thus relatively rigid and planar, although free rotation can take place about the C<sub>α</sub>-N and C<sub>α</sub>-C bonds i.e., the bonds either side of the peptide bond, permitting adjacent peptide units to be different angles. The hydrogen of the amino group is nearly always on the opposite side (trans) of the double bond to the oxygen of the carboxyl group, rather than on the same side (cis).

### Classification of Proteins:-

Depending on solubility properties and on known chemical and physical differences, proteins are classified into two major groups - single proteins and conjugated proteins.

1. Simple proteins:- They are compounds that on hydrolysis, yield only amino acids. On the basis of solubility properties simple proteins can be further divided into six groups as below -

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i) Albumins:- They are soluble in water and in dilute salt solutions. They can be coagulated by exposure to heat.

eg:-  $\beta$ -amylase of Barley

ii) Globulins:- They are insoluble or sparingly soluble in water. They are soluble in dilute <sup>salt</sup> solutions. They also can be coagulated by exposure to heat. Eg:- Storage proteins of seeds.

iii) Glutelins:- They are insoluble in neutral solutions but are soluble in weak acid or basic solutions.

Eg:- Glutelin of wheat, oryzenin of rice.

iv) Prolamin:- They are insoluble in water and absolute ethanol but are soluble in 70 to 80 percent ethanol. On hydrolysis they yield relatively large quantities of proline and ammonia. Eg:- zein of maize, gliadin of wheat and rye, hordein of barley.

v) Histones:- They are soluble in water and are rich in basic amino acids, such as arginine and lysine. They have been found in cell nuclei and may remain associated with nucleic acids.

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v) guanamines - They are soluble in water and are rich in basic amino acids. They also are found in nuclei and probably remain associated with nucleic acids. But this group lack amine ad as tyrosine and + arginine and certain hexaphum.

4. Conjugated proteins - The conjugated proteins are associated with a nonamino acid component which is referred to as prosthetic group. A/c to the various prosthetic groups, conjugated proteins are divided into following divisions -

i) Nucleoproteins - On hydrolysis they yield a simple protein and a nucleic acid.

ii) Glycoproteins - They are associated with carbohydrate as prosthetic groups. Eg. Some proteins of cell membrane.

iii) Lipoproteins - They are associated with lipids as prosthetic groups and are insoluble in water. Eg. Some membrane proteins.

iv) Chromoproteins - They are associated with pigments as prosthetic groups. Eg. Flavoproteins, haemoproteins, carotenoid pigments

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transglutinin etc. They have metal molecules as their prosthetic groups. E.g. some respiratory enzymes.

Biogenesis of Protein

The proteins are macromolecules consisting of a series of varying amino acids. The proteins molecules differ from each other in their length and definite sequence of amino acids. The protein molecule synthesis is directly under genetic control and the master molecule DNA dictates the exact sequence of amino acids in a synthesizing chain. There are two major steps in the mechanism of protein synthesis, which are as below —

1. Transcription — Transcription is a synthesis for RNA synthesis in molecular biology. The necessary information for specific sequence and the total number of amino acids, to be incorporated into the protein, are stored in the master molecules DNA in the form of sequence of bases. \* DNA transfers the information to the protein synthesising apparatus through the

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special messenger called messenger RNA or mRNA. The transfer of information from DNA to RNA is called transcription. The process of transcription itself involve 3 steps -  
1) Polymerase binding and initiation - First, the double stranded DNA gets uncoiled and the two strands are separated from each other by breaking of hydrogen bonds between bases. Out of the two strands, one is coding strand and the other is template strand. Later takes part in mRNA synthesis i.e. transcription. RNA polymerase is the enzyme for RNA synthesis. There is only one type of RNA polymerase in prokaryotes, while there are three different RNA polymerases in eukaryotes. RNA polymerase binds itself at a start signal or initial binding site on the template strand of the DNA, which is called as promoter site. The enzyme bears a sigma ( $\sigma$ ) factor which recognises the promoter sequence without any copies mistakes.

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ii) Elongation— After the initial binding, the  $5'$  end copies the base sequence of template DNA strand accurately and complementary ribonucleotides (i.e. ATP, GTP, UTP and CTP) are joined together by the enzyme at a rate of 35 nucleotides per second. Elongation of the RNA proceeds in the  $5' \rightarrow 3'$  direction.

A. The portion of DNA responsible for synthesizing mRNA leading to the production of one polypeptide chain is known as gene.

iii) Termination— Termination of transcription takes place when the polymerase arrives at a stop signal in the DNA. A transcription factor called rho ( $\rho$ ) factor, a protein with ATPase activity causes the release of the completed RNA molecules. The terminal region of the RNA transcription ends in 4-8 consecutive Uridines.

The region of DNA template (gene) coding for a single protein is also called a clonon. In prokaryotes there are gene clusters (polyclonon mRNA). Prokaryotic polyclonon mRNA contains several start (AUG) and stop (UAG, UAA, UGA) codons for several protein synthesis in the same mRNA. On the other hand in eukaryotes, the mRNA are always monoclone and have

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a single start (AUG) codon per mRNA molecule 06.02

2. Translation:— Translation involves chiefly three cell components — mRNA, tRNA and ribosomes. Out of these RNA molecule is about 74-95 nucleotides long. About half of the nucleotides of a tRNA are base paired to form double helices and all tRNA molecules <sup>common</sup> <sub>clover</sub> leaf secondary structure. The 5' terminal residues form double helices and have common primary and secondary structure. The 5' terminal residue is usually phosphorylated guanine (PGI). The base sequence at the 3' end is CCA. During synthesis of the aminoacyl-tRNA, the amino acid is covalently bound to the residue of the CCA sequence at the 3' end. Each tRNA molecule carries only single amino acid. Each tRNA molecule also bears a triplet of bases called an anticodon, which is complementary to the codon of the mRNA, corresponding to a specific amino acid.

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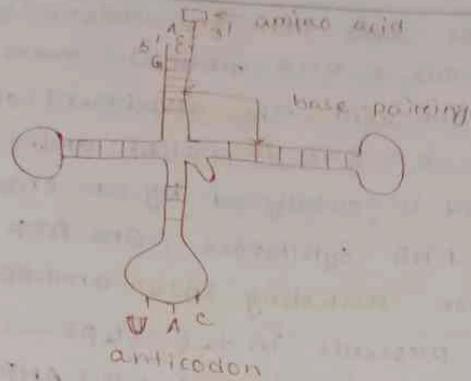


Fig - Clover leaf secondary str. of a t-RNA

Ribosomes are the cytoplasmic organelles where protein synthesis takes place. Each ribosome is made up of two sub units - one smaller and other larger. In case of prokaryotes the two subunits are 20s and 50s while in eukaryotes, they are 40s and 60s. The two subunits are not bound together when they are not involved in protein synthesis. Each a specialised RNA called ribosomal RNA (rRNA)

The process of translation proceeds in three steps - Initiation, elongation and termination

1) Initiation - The steps are as below -

a) Amino acid activation - There is at least one t-RNA for each amino acid, in the cell. However, because of the redundancy of the genetic code,  
(quality of super flow)

these may be more than one codons and hence more than one t-RNA molecules are used for the same amino acid. The attachment of an amino acid to a t-RNA is called amino acid activation and is catalysed by an enzyme called aminoacyl-t-RNA synthetase. One ATP is involved in the reaction resulting into aminoacyl-t-RNA. The reaction proceeds in two steps —

1) Amino acid + ATP  $\rightleftharpoons$  Aminoacyl-AMP + PP<sub>i</sub>  
2) Aminoacyl-AMP + t-RNA  $\rightarrow$  Aminoacyl-t-RNA + AMP

b) Initiation of protein synthesis is catalysed by 3 proteins called initiation factors (IF<sub>1</sub>, IF<sub>2</sub>, IF<sub>3</sub>) in prokaryotes. Initiation begins with the binding of IF<sub>1</sub> and IF<sub>3</sub> to the small (30S) ribosomal subunit. The small subunit then binds to the mRNA via the Shine-Dalgarno sequence (AUG AUG) and moves along the mRNA until it locates the AUG initiation codon. In prokaryotes the first amino acid initiation codon in prokaryotes the first amino acid of a few proteins is N-formylmethionine (fMet). The initiator t-RNA charged with N-formyl

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methionine and in a complex with IF<sub>2</sub> and GTP (Met-tRNA<sup>Met</sup>/IF<sub>2</sub>/GTP) now binds. IF<sub>2</sub> is released subsequently. The complex of mRNA, Met-tRNA + Met, IF<sub>2</sub>, IF<sub>3</sub>, and the 30S subunit is called the 30S initiation complex. The large (50S) ribosomal subunit now binds, with the release of IF<sub>1</sub> and IF<sub>3</sub> and hydrolysis of GTP, to form a complete 70S initiation complex. Besides the binding site for mRNA, each complex 70S ribosome has three binding sites for t-RNAs. They are aminoacyl-tRNA site or A site, peptidyl-tRNA site or P site and exit site or E site. In initiation stage, the initiator aminoacyl-tRNA binds directly at the P site.

(i) Elongation:—Elongation of the polypeptide chain occurs in three steps namely aminoacyl-tRNA binding, peptide bond formation and translocation.

(ii) Aminoacyl-tRNA binding:—In this first step, the corresponding aminoacyl-tRNA for the second codon binds to the A-site via codon-anticodon interaction. This binding of the aminoacyl-tRNA requires elongation factor EF-TU and GTP, which bind as an

aminoacyl-tRNA/EF-TU/GTP complex following  
knowing the GTP molecule. In the EF-TU following  
peptide bond formation? The second step is  
the peptide bond formation is catalyzed by  
peptidyl transferase, part of the large riboso-  
somal subunit. In this step, the carboxyl end  
of the amino acid bound to the tRNA in  
the P-site is uncoordinated from the amino  
terminus joined by a peptide bond to the amino  
group of the amino acid linked to the tRNA in  
the A-site.

Translocation: In the third step, a complex  
of elongation factor EF-G (also called translo-  
cator GTPase, EF-G/GTP) binds to the ribosome.  
Three concerted movements now occur, collectively  
called ratcheted movement. t-RNA moves from the A-site  
moves to the P-site, and the ribosome moves  
along the m-RNA from 5' to 3' direction by  
three nucleotides to place the next codon in  
the A-site. During translocation events GTP  
is hydrolysed to GDP and inorganic phos-  
phate and EF-G is released.

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After translocation, the A-site is empty and ready to receive the next aminoacyl-tRNA. The A-site and the E-site cannot be occupied simultaneously. Thus the deacylated tRNA is released from the E-site before next amino acyl tRNA binds to the A-site to start a new round of elongation. Elongation adding of amino acid to the c-terminal end of the growing polypeptide for each codon that is read by the ribosome.

iii) Termination: UAG, UAA and UGA are said to be termination codons or stop codons, which unlike codons, do not contain aminoacyl t-RNAs. Complementary to them. So when one of these three termination codons are present, they release factor RF<sub>3</sub> is also needed to assist RF<sub>1</sub> or RF<sub>2</sub>. These release factors cause the peptidyl transferase to transfer the polypeptide to a water molecule instead of aminoacyl tRNA. Thus the bond between the polypeptide and t-RNA in the P-site is cleaved effectively. The released polypeptide now leaves ribosome, followed by the mRNA and free t-RNA and the ribosome dissociates into 30S and 50S subunits.