

## 14. PROTEIN AND FAT SYNTHESIS

### Biosynthesis of protein

Protein is a complex organic nitrogenous substance found in all living tissues of plants and animals. They are polymer of amino acids in linear order. Synthesis of protein may take place from amino acids produced by direct amination of organic acids or by degradation of protein. Former is known as *primary protein synthesis* while the latter is called *secondary protein synthesis*. Protein synthesis occurs in pre – DNA synthesis phase (G<sub>1</sub> phase) of cell cycle.

Biosynthesis of protein takes place in prokaryotes as well as in eukaryotes. Kinds of protein to be synthesized depend upon the gene (DNA segment). Gene is continuous uninterrupted sequence of nucleotides which codes for a single polypeptide chain. Now it is believed that the sequence of some eukaryotic genes is found to be interrupted by nucleotides that are not represented with the amino acid sequence of protein. They are non-coding (silent). Genes control all metabolic processes by synthesizing proteins (enzymes).

Structure of an eukaryotic gene showing Exon (coding part) and Intron (non-coding part).

Mechanism of protein synthesis: Protein synthesis takes place in following two stages:

- I. Transcription
- II. Translation

**Transcription:** Transcription occurs throughout inter phase and continues up to early prophase of cell division. It is primary stage of protein synthesis. When DNA produces DNA the process is called replication but when DNA produces RNA the process is called transcription. In the former case DNA is duplicated while in the latter case protein is synthesized. During transcription RNA is synthesized on DNA template. Here, information or order contained in DNA is passed on the mRNA for synthesis of particular protein. The information is in coded form and consists of three nitrogenous bases (triplet codons). The part of DNA responsible for synthesis of mRNA is which leads to one polypeptide chain is

called cistron (functional gene). New strands of mRNA are synthesized on DNA template making use of RNA nucleotides present in surroundings in 5' 3' direction just like DNA chains.

Single DNA strand serves as template for RNA polymerase and synthesizes RNA. DNA strand which serves as template for transcription is called the sense strand. The complementary strand is antisense strand. In SV – 40 viruses both strands of DNA are transcribed and it is called symmetrical transcription, but when only one strand is transcribed it is called asymmetrical transcription. Former type of transcription also occurs in Polymerase virus DNA and in the mitochondria genome.

Mechanism of transcription

Transcription involves following events:

- I. Uncoiling of DNA molecule
- II. Synthesis and action of enzyme RNA polymerase
- III. Synthesis of hn RNA / mRNA.

Uncoiling of DNA molecule: As per “nucleosome model” of chromosome. Chromosomes are ‘matarmala’ like beaded structure. Beads are separated by string and are made up of DNA and histone protein. Histone proteins are of 5 kinds: H2A, H2B, H3, H4 and H5 or H1. First four constitute the bead core and H1 links two beads. The DNA molecules are wrapped on histone protein cores and linker protein core in beaded and linker regions respectively.

Structure and function of enzyme RNA polymerase: RNA polymerase is a holoenzyme. Core particle consists of sub units  $\alpha'$ ,  $\beta'$  and  $\psi$ . Cofactor consists of sigma factor. For functional RNA polymerase formation the two (core enzyme and sigma factor) genes unite. Sigma factor recognizes correct start signal at DNA template and core enzyme continues transcription. Sigma factor dissociates after initiation of transcription to adhere with other core enzyme of RNA polymerase. In prokaryotes RNA polymerase is only one type while in eukaryotes.

Production of mRNA / hnRNA: In prokaryotes where nucleus is not well organized mRNA is the direct product of transcription, while in eukaryotes the direct product of transcription is hnRNA (heteronucleic RNA) and mRNA is derived from hnRNA by cutting and splicing. HnRNA has coding and non-coding sequences. Coding sequences are interrupted by non-coding sequences. Non coding sequences are removed by splicing (cutting) by endonuclease enzyme and coding sequences are ligated together to form mRNA. The spliced non-coding sequences are degraded within nucleus. It never goes out of nucleus. Thus, only fraction of hnRNA is translocated to cytoplasm from nucleus via nuclear pore.

In eukaryotes migration of mRNA from nucleus to cytoplasm via nuclear pore occurs through poly a tail. According to another view, ribosome's pull the mRNA from nucleus to cytoplasm. Now mRNA gets established in cytoplasm.

**Translation:** Translation is a process in which order (message) given by DNA to mRNA for synthesis of particular protein is implemented (conveyed). Genetic information concealed in mRNA directs the synthesis of particular protein. These orders are in coded form. This coded information (expressed through codons) is recognized by tRNA having anticodons. Anticodons are opposite to codons (codons and anticodons are complementary to each other).

**Mechanism of translation:** It involves following events:

- I. Activation and selection of amino acids
- II. Transfer of amino acids to tRNA molecules
- III. Formation of protein synthesizing apparatus and chain initiation
- IV. Chain elongation
- V. Chain termination

## **FAT SYNTHESIS**

Fat synthesis can be studied under the following heads: -

### **1. Fat synthesis of Glycerol**

There may be different methods of the formation of glycerol in plants, but one of the very common methods is from dihydroxy acetone phosphate which is an intermediate of glycolysis. Dihydroxyacetone phosphate is first reduced to  $\alpha$ -glycerophosphate by the enzyme glycerol – 3 – phosphate dehydrogenase. Co-enzyme  $\text{NADH}_2$  is oxidized in this reaction.  $\alpha$ -glycerophosphate is then hydrolysed by *glycerol phosphatase* to liberate phosphoric acid and forming glycerol.

## 2. Synthesis of Fatty acids

Long chain saturated fatty acids\* are synthesized in plants from active two carbon units, the acetyl – CoA ( $\text{CH}_3\text{CO}\cdot\text{CoA}$ ). Although the reactions of  $\beta$  – oxidation of fatty acids are reversible, the fatty acids are not formed simply by the reverse reactions of  $\beta$  – oxidation. Synthesis of fatty acids from  $\text{CH}_3\text{CO}\cdot\text{CoA}$  takes place step by step. In each step the fatty acid chain is increased by two carbon atoms. Each step involves two reactions –

(i) In the first reaction which takes place in the presence of acetyl – CoA carboxylase, acetyl – CoA combines with  $\text{CO}_2$  to form malonyl – CoA (malonic acid is 3 – C compound). ATP provides energy while  $\text{Mn}^{++}$  and biotin are required as co-factors.

(ii) Malonyl CoA reacts with another molecule of  $\text{CH}_3\text{CO}\cdot\text{CoA}$  in the presence of fatty acid synthetase and Coenzyme  $\text{NADPH}_2$  to form Coenzyme – A derivative butyric acid (butyric acid contains 4 – atoms). One mol. Of  $\text{CO}_2$ ,  $\text{H}_2\text{O}$  and CoA are released while  $\text{NADPH}_2$  oxidised in the reaction.

Butyryl CoA, in the next step will combine with malonyl CoA to form CoA derivative of fatty acid containing 6-C atoms. This process is repeated till Coenzymes-A derivative of long chain fatty acid (which may contain up to 16-18C atoms) is produced.

(As a matter of fact the enzyme fatty acid synthetase is not simple but a complex of many enzymes (multienzyme complex) and an acyl carrier protein called as ACP\*\*. And actually the reaction (ii) described above only summarises a number of reactions involved in the synthesis of fatty acid from acetyl – CoA which can be grouped under 3 categories

**Initiation reaction:** In this reaction acetyl CoA transfers its acetyl group to one of the – SH groups of multienzyme complex i.e. fatty acid synthetase.

Unsaturated fatty acids are synthesized by denaturation of saturated fatty acid. ACP is similar to CoA in having phosphopantetheine as the functional unit in their structures. In CoA, it is esterified to Adenosine 3, 5 – bisphosphate but in ACP, it is esterified to serine of a protein chain consisting of 81 amino acids.

### **Chain elongation reactions**

Six different reactions involved here are (i) malonyl transfer, (ii) condensation, (iii) reduction, (iv) dehydration, (v) reduction and (vi) acyl transfer. Chain elongation starts with the transfer of malonyl group from malonyl – CoA to second – SH group of the multienzyme complex. Then, there is a condensation of the latter so that a 4 – C unit is produced. This unit by next three reactions i.e. reduction, dehydration and reduction is converted into saturated 4 – C unit (i.e. butyryl – CoA). In acyl transfer reaction the fatty acid residue is transferred back to the – SH group to which the acetyl group was transferred in initiation reaction. The cycle is repeated again and again with malonyl transfer, condensation etc. till the fatty acid residue consists of up to 16 - 18 C atoms. Each such turn elongates fatty acid chain by 2 – C atoms. Details of chain elongation reactions are given below

### **Termination reaction**

When the fatty acid residue has attained a desired length the chain elongation stops at reaction (v) and the cycle is not repeated. The acyl group instead of being transferred to the – SH of the enzyme is transferred to – SH group of Co-enzyme A (CoASH) molecule. Thus, CoA derivate of the fatty acid is produced which can then be utilized in fat synthesis. The enzyme becomes free.

It is believed that during this process of fatty acid synthesis, the acyl group of fatty acid bound to the – SH group of ACP. The latter then passes it from one enzyme of the complex to the other.

### **(3) Condensation of fatty acids and Glycerol**

The fats or triglycerides are synthesized not from glycerol and free fatty acids but from  $\alpha$  – glycerophosphate and CoA derivatives of fatty acid, i.e. fatty acyl CoA residues. First, there is acylation of  $\alpha$  – glycerophosphate by two fatty acyl – CoA molecule to form phosphatidic acid. Now dephosphorylation occurs in the presence of phosphatase and a diglyceride is formed. The acylation of the free – OH groups of diglyceride completes the biosynthesis of triglyceride or fat.