RNA POLYMERASE
&
TRANSCRIPTION UNIT

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The process by which RNA molecules are synthesized on DNA template is called transcription. An mRNA molecule is synthesized which is complementary to the gene sequence of one of the two strands of DNA double helix. Soon after the double-helical model of DNA was first proposed, Francis Crick proposed a principle called **Central Dogma of Molecular Biology,** which explains the directional flow of genetic information from DNA to RNA to protein (Fig. 12.5)

Like replication, transcription requires three major components:

1) A DNA template;

2) The raw materials required to build a new RNA molecule,

3) The transcription apparatus, consisting of enzymes, proteinaceous factors, cofactors, and activators necessary to catalyze the synthesis of RNA.

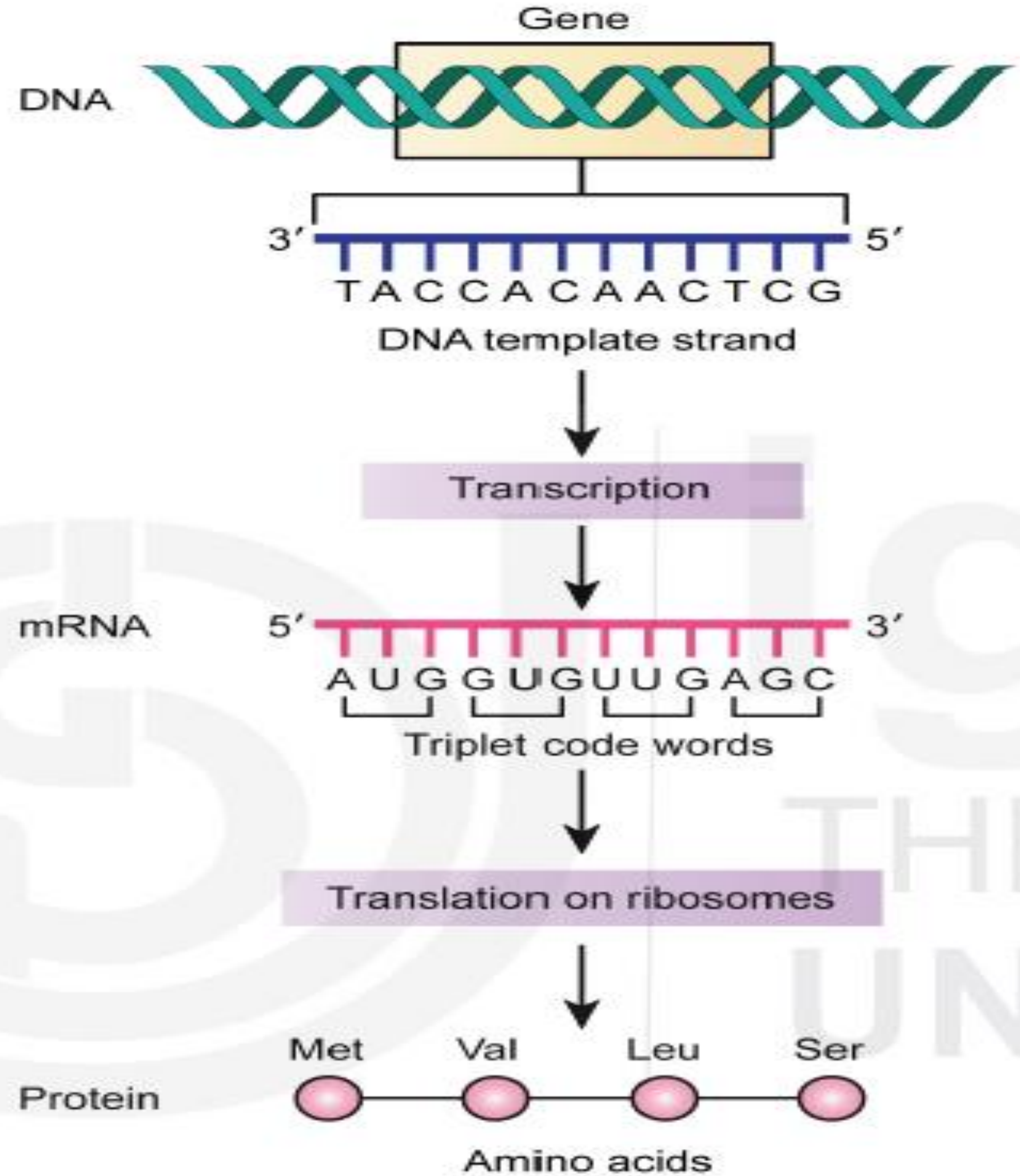

Fig. 12.5: Flow chart showing how genetic information encoded in DNA produces a protein.

Transcription apparatus (*RNA polymerase* and accessory proteins)

Bacterial *RNA polymerase*: The enzyme capable of synthesizing RNA on DNA template was discovered by several investigators including Samuel Weiss (1959) who isolated it from rat liver and called it *RNA polymerase*. *RNA polymerase* from *E. coli* has been extensively investigated. Bacterial *RNA polymerase* is a multimeric enzyme (i.e., it is made up of several polypeptide chains) consisting of five subunits that make up the core enzyme: two copies of subunits called alpha (α) and single copies each of subunits beta (β), beta prime (β'), and omega (ω) (Fig.12.9).

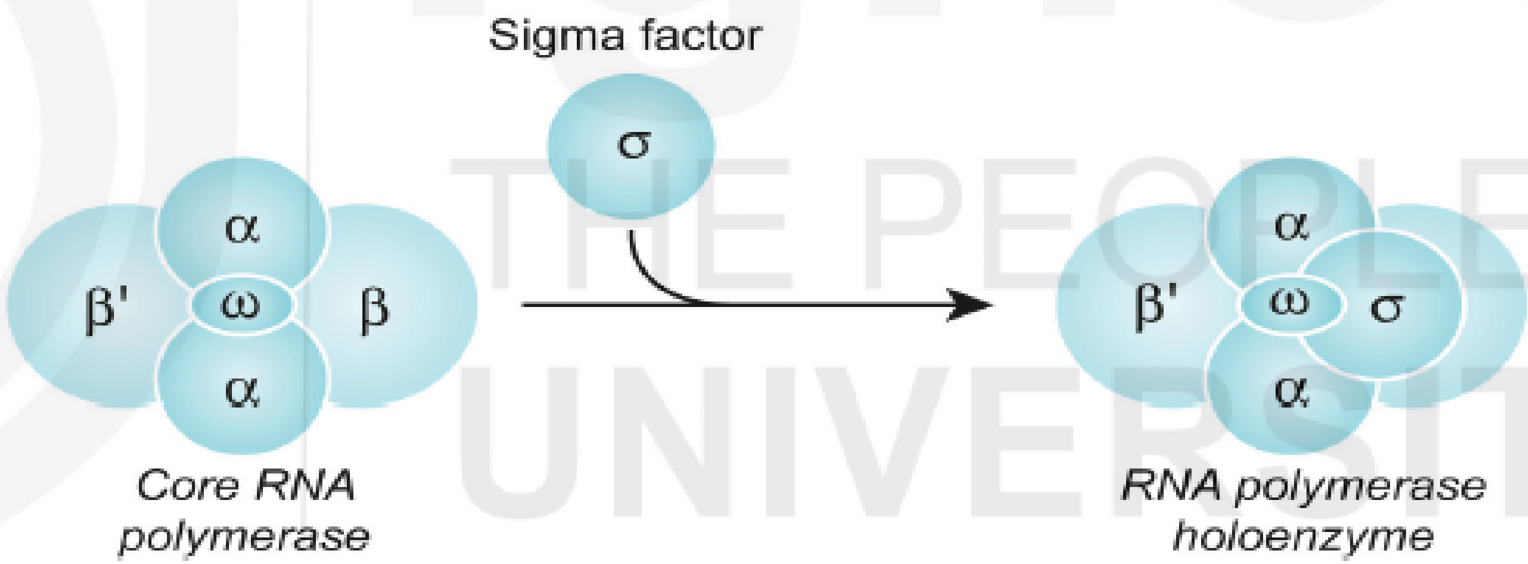

Fig 12.9: The core enzyme of bacterial *RNA polymerase* consisting of five subunits. The core enzyme catalyzes the elongation of RNA molecule and adds new RNA nucleotides.

Beta (β) and Beta prime (β') polypeptides provide the catalytic mechanism and the active site for transcription. Although the ω subunit does not directly participate in transcription, it helps in stabilizing the enzyme. Other functional subunits join and leave the core enzyme at particular stages of transcription process. The sigma (σ) factor plays a regulatory role in initiation of RNA transcription. It controls the binding of *RNA polymerase* to the promoter thereby ensuring that initiation takes place at the precise site. Without sigma factor, *RNA polymerase* will initiate transcription at random point along the DNA. The complex active form of the enzyme, i.e., the holoenzyme is formed only when sigma factor binds with the core enzyme. It is only after this binding that the *RNA polymerase* acquires the capability to stably bind to the promoter region, and not anywhere else, and initiate transcription at proper start site. After a few RNA nucleotides have joined together, the sigma factor usually detaches from the core enzyme.

Eukaryotic *RNA polymerases*: Eukaryotic cells contain three distinct nuclear *RNA polymerases* viz., *RNA polymerase* I, II and III. These enzymes transcribe different classes of genes forming different RNAs (see Table 12.2). Protein-coding genes are transcribed by *RNA polymerase II* to yield mRNAs. *RNA polymerase II* also transcribes microRNAs (miRNAs), snRNAs, snoRNAs and long noncoding RNAs (lncRNAs), which are critical regulators of gene expression in eukaryotic cells. Ribosomal RNAs (rRNAs) and transfer RNAs (tRNAs) are transcribed by *RNA polymerase I* and III. *RNA polymerase I* is specifically devoted to transcription of three largest species of rRNAs, which are designated as 28S, 18S, and 5.8S. *RNA polymerase III* transcribes the genes for tRNAs and for the smallest species of ribosomal RNAs (5.8S rRNAs). Some of the small RNAs involved in splicing and protein transport (snRNAs and scRNAs) are also transcribed by *RNA polymerase III*. Interestingly, chloroplasts and mitochondria also possess separate *RNA polymerases* which are similar to bacterial *RNA polymerases*. These enzymes specifically transcribe the DNAs for the organelles.

All the three *RNA polymerases* are complex enzymes consisting of 12 to 17 different subunits each. Although they recognize and transcribe distinct classes of genes, they share several features common with each other as well as with bacterial *RNA polymerase*. In particular, all three eukaryotic *RNA polymerases* contain nine conserved subunits, five of which are related to the, β, β', and ω subunits of bacterial *RNA polymerase*.

Table: 12.2: Eukaryotic *RNA polymerases*.

Type	Present in	Transcribes
RNA polymerase I	All eukaryotes	large RNAs
RNA polymerase II snRNAs, snoRNAs, some miRNAs	All eukaryotes	pre-mRNAs
RNA polymerase III, some snRNAs and miRNAs	All eukaryotes	tRNAs, some rRNAs,
RNA polymerase IV	Plants and some siRNAs	
RNA polymerase V	Plants RNA molecules taking part in heterochromatin formation	

The Transcription Unit

A transcription unit is a stretch of DNA that encodes an RNA molecule and the sequences necessary for its transcription. It includes three critical regions: a promoter, an RNA-coding sequence and a terminator (Fig. 12.7). The transcription apparatus recognizes and binds to the promoter DNA sequence. The binding decides which of the two DNA strands is to be read as template and what will be the direction of transcription. In addition, the promoter also determines the transcription start site, the first nucleotide that will be transcribed into RNA. Although in most transcription units, the promoter is located next to transcription start site, it does not itself get transcribed.

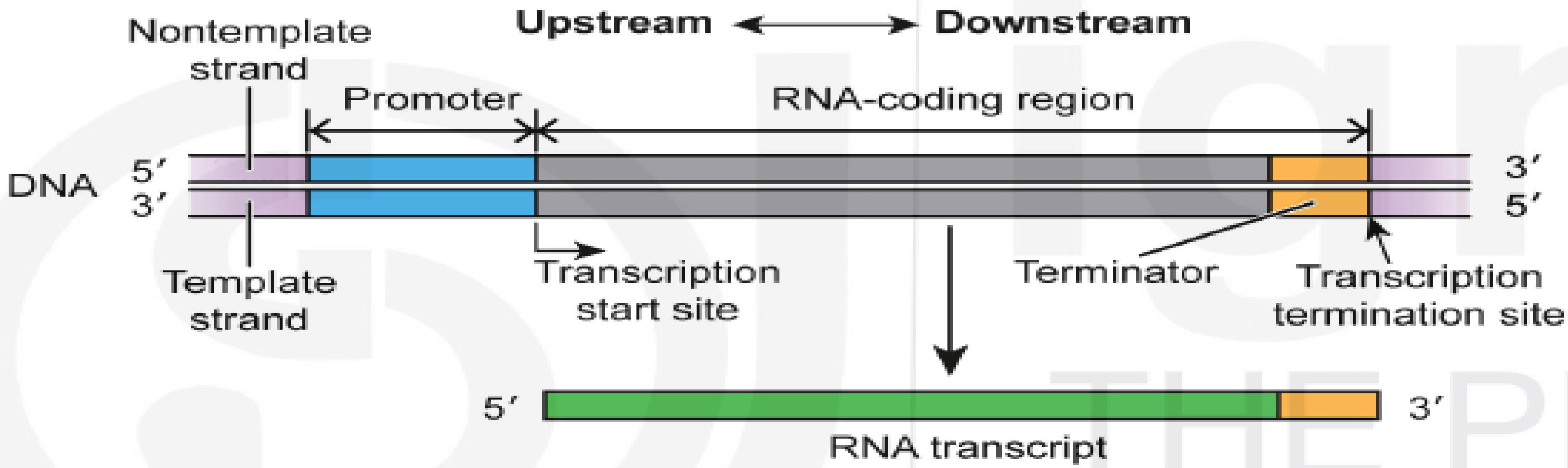

Fig. 12.7: A transcription unit showing a promoter, a RNA-coding region, and a terminator.

The second critical region of the transcription unit is a sequence of DNA nucleotides that is copied into a RNA molecule. This is called as the RNA- coding region. The third component of a transcription unit is the terminator, a sequence of nucleotides that signals where transcription is to end. The terminator portion is usually the part of the RNA-coding sequence. It is only after the terminator has been copied into RNA that the process of transcription stops.