RNA METABOLISM

The expression of the information in a gene involves production of RNA molecule transcribed from the DNA template. During the transcription, the enzyme (RNA polymerase) converts the genetic information present in the segment of double stranded DNA which is called gene into an RNA strand with a base sequence complementary to one of the DNA strands. The three major kinds of RNA that are produced are mRNA that encodes the amino acid sequence of one or more polypeptides specified by the gene. The t-RNA reads the ribonucleotide sequence of mRNA and brings appropriate amino acid to ribosome for protein synthesis. The rRNAs are the constituents of ribosomes. During replication, the entire DNA (or the chromosome) is replicated but the transcription is selective process. In transcription only a part of the DNA that is called gene is transcribed in a particular condition and in a particular cell type.

As the term genome is applied to sum of all DNA present in cell required for its survival, while the sum of all the RNA molecules produced in a cell under a particular condition is called *transcriptome*.

Transcription: *the term transcription refers to synthesis of RNA strand on the DNA template of a gene by enzyme RNA polymerase*.

The basic reaction of RNA synthesis in transcription is

$$\underset{\text{RNA}}{(\text{NMP})_n} + \text{NTP} \longrightarrow \underset{\text{Lengthened RNA}}{(\text{NMP})_{n+1}} + \text{PP}_i$$

Resemblance between DNA replication and transcription

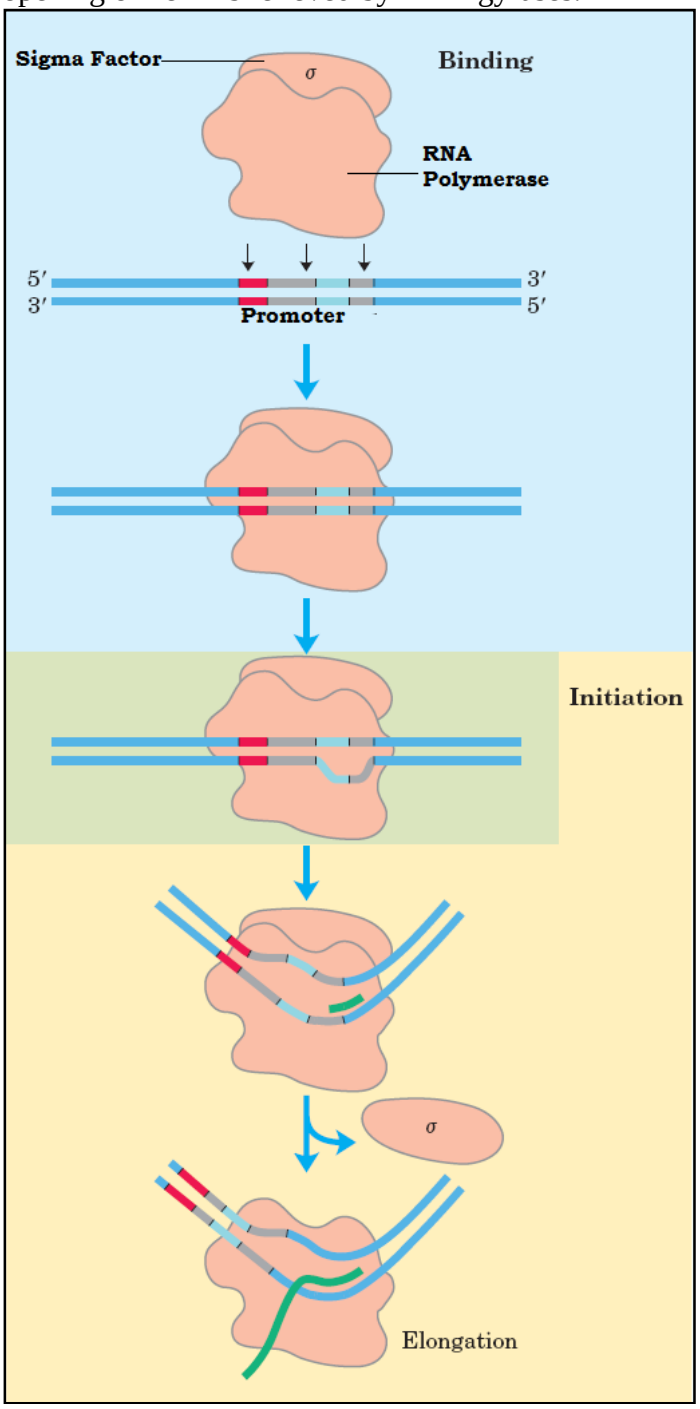
1. RNA like DNA is synthesized from 5`→3` direction.
2. DNA and RNA synthesis requires a DNA template.
3. Both the processes are divided into three phases: *Initiation*, *elongation* and *termination*.
4. The RNA strand in case of transcription is complementary to DNA template (Just like the new strand of DNA is complementary to old template strand in case of DNA replication).

DIFFERENCE BETWEEN DNA REPLICATION AND TRANSCRIPTION

	REPLICATION	TRANSCRIPTION
1.	Primer is required	Primer is not required
2.	Both strands of DNA acts as template	Only one strand acts as template. On this strand only the synthesis of RNA occurs. The other strand is coding strand or non-template strand.
3.	Nucleotide monomers are deoxy-ribonucleotide.	Nucleotide monomers are ribonucleotide.
4.	Enzyme: DNA polymerase	Enzyme: RNA polymerase
5.	DNA polymerases have the proofreading activity	*RNA polymerases lack the proofreading activity*
6.	The DNA replication initiates at the origin of replication.	The DNA transcription initiates at the region of DNA called **Promoter**.
7.	The DNA replication involves whole of DNA molecule.	The DNA transcription involves only a part of DNA called *gene*.

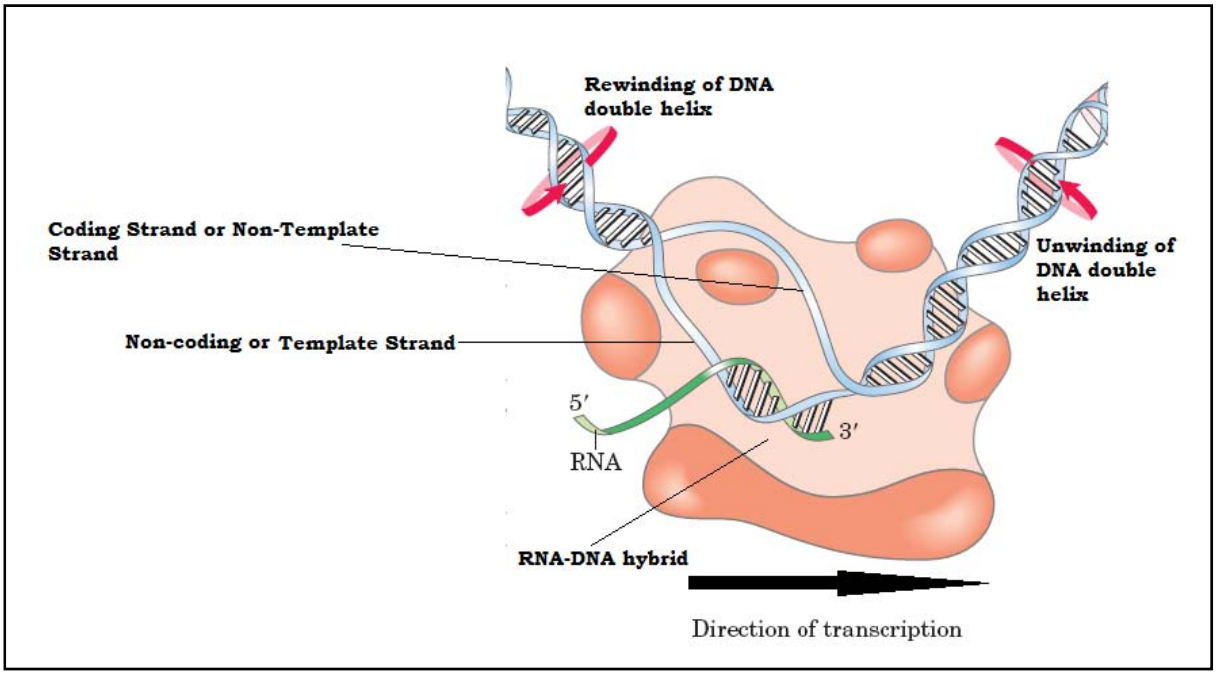
DNA TRANSCRIPTION OF PROKARYOTIC GENES

INITIATION OF TRANSCRIPTION: the RNA polymerase requires an additional protein factor called sigma factor. This factor helps in recognizing the promoter. To enable the RNA polymerase to synthesize an RNA strand on the DNA template, the double stranded DNA is required to be unwound over a short distance to form a bubble like structure called **transcription bubble**. An RNA-DNA hybrid occurs in the unwound region of DNA of 8 nucleotides length. The rate of transcription is about 50-90 nucleotides per second. The RNA polymerase enzyme itself has the helicase activity and the torsional stress in DNA due to opening of helix is relieved by DNA gyrases.



ELONGATION: The RNA polymerase moves from 5`→3` direction of RNA strand to synthesize RNA. The dissociation of sigma factor marks the beginning of transcription

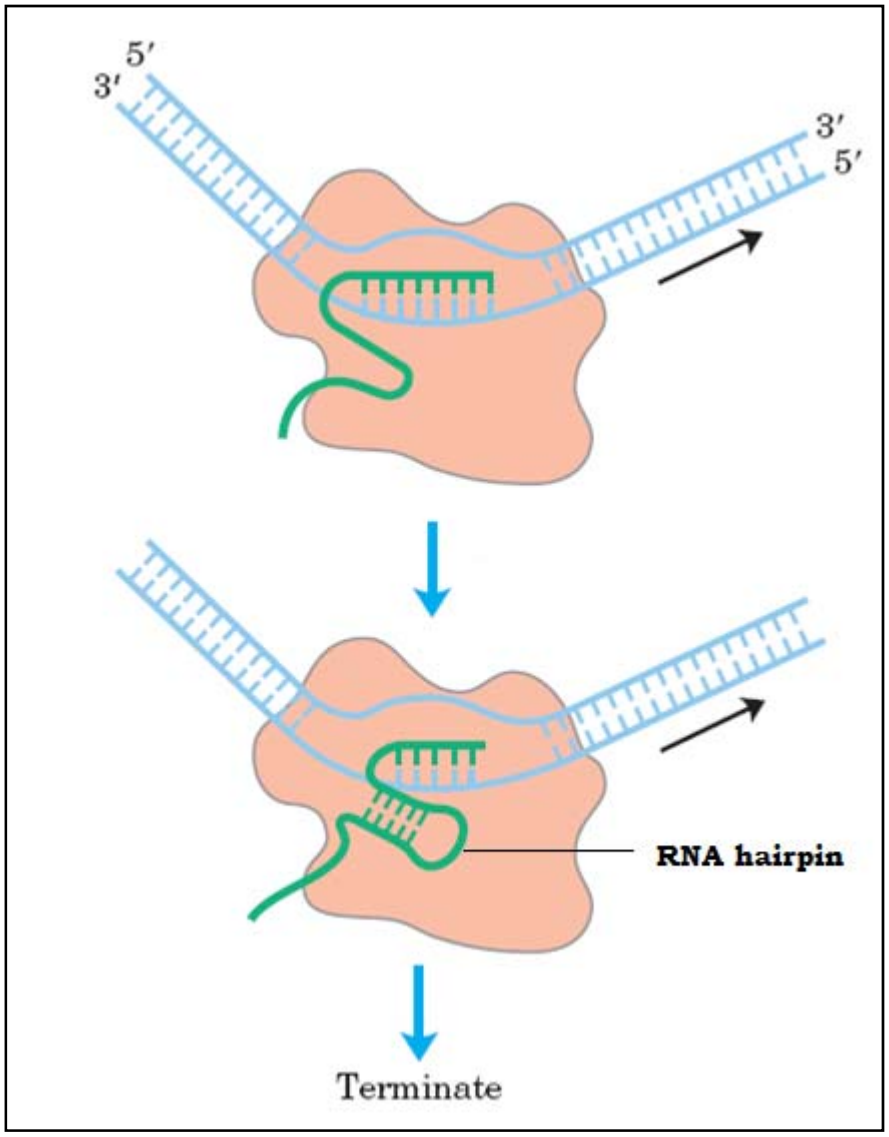
elongation. The RNA polymerase unwinds the DNA helix in front of it and then rewinds the helix at back of it.



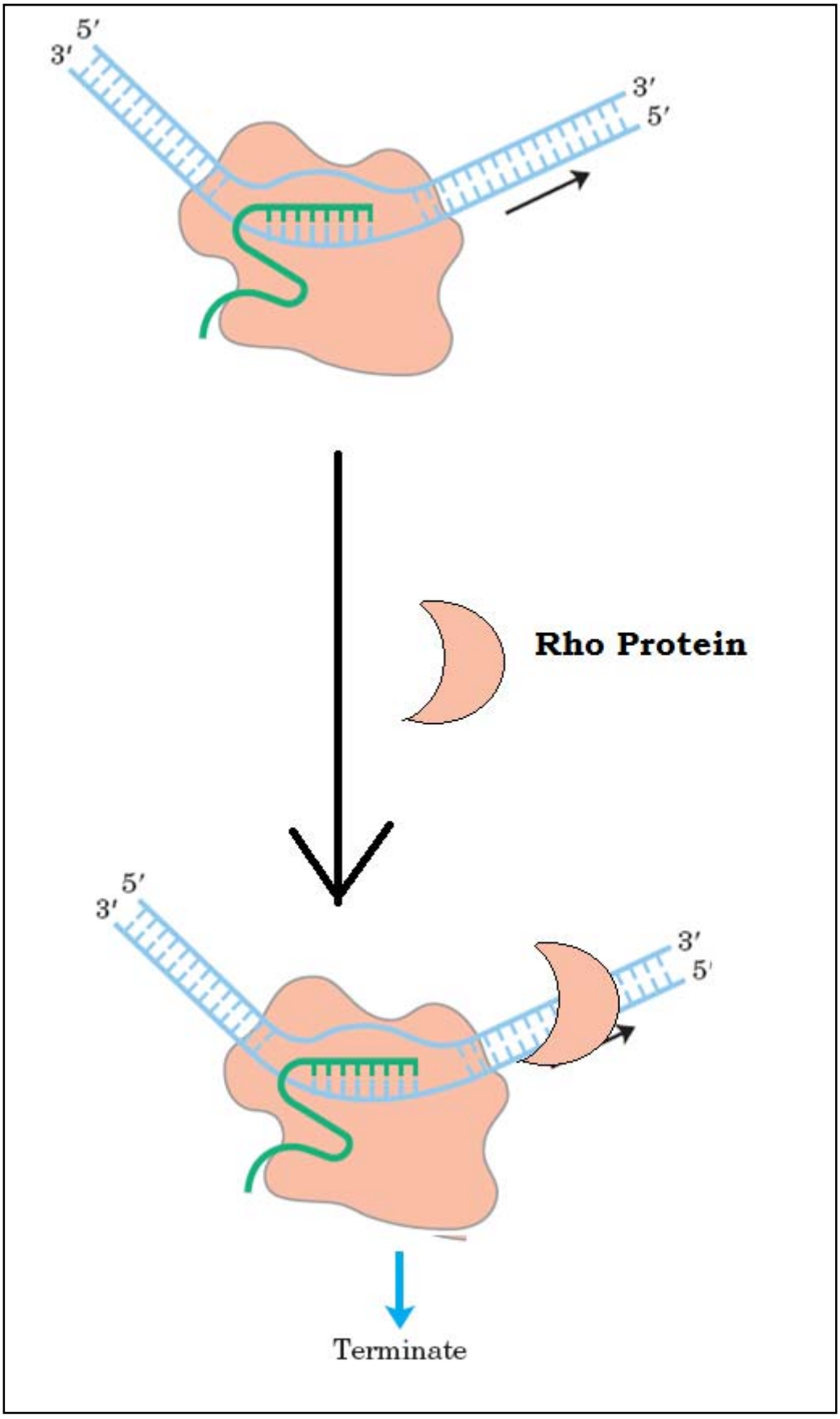
TERMINATION OF DNA TRANSCRIPTION: The termination of transcription occurs in either of two different manners:

a. Rho independent termination.
b. Rho dependent termination.

a. Rho independent termination: When polymerase reaches a sequence at 3`end of the gene, it encounters self complementary DNA sequence that causes the RNA to form hairpin. This hairpin in RNA does not allow the RNA polymerase to move ahead causing dissociation of enzyme from the DNA causing the termination (as shown figure below).



b. Rho dependent termination: the Rho protein binds a region of DNA present at the end of a gene called RUT sequence (Rho Utilization sequence). This protein dissociates the RNA from DNA template and RNA polymerase (as shown figure below).


5′
3′
3′
5′
Rho Protein
5′
3′
3′
5′
Terminate

TRANSCRIPTION OF EUKARYOTIC GENES

The transcriptional machinery in the nucleus of a eukaryotic cell is much more complex than that in bacteria. Eukaryotes have three RNA polymerases, designated I, II, and III, which are distinct complexes but have certain subunits in common. The transcription in eukaryotes is catalyzed by three different types of RNA polymerases namely RNA polymerase I, RNA polymerase II and RNA polymerase III. RNA polymerase I (Pol I) is responsible for the synthesis of only one type of RNA, a transcript called ribosomal-RNA. The principal function of RNA polymerase II (Pol II) is synthesis of mRNAs and some specialized RNAs (such as catalytic RNAs). RNA polymerase III (Pol III) makes tRNAs.

RNA Polymerase II Requires Many Other Protein Factors for Its Activity called *transcription factors*. Some proteins are required during the phase of elongation phase of transcription called *elongation factors*.

Functions of the transcription factors:

1. Recognition of promoter.
2. Provides the helicase activity.

RNA PROCESSING

The RNA synthesized out of DNA transcription undergoes the post-transcriptional RNA processing. Many of the RNA molecules in bacteria and virtually all RNA molecules in eukaryotes are processed to some degree after synthesis. This process occurs primarily in eukaryotic and Archeobacteria genes. It is also seen in some bacterial genes.

The newly synthesized RNA in nucleus of eukaryotes is called primary transcript. This primary transcript (in case of mRNA it is called pre-mRNA) is subjected to RNA processing making it matured transcript (in case of mRNA it is matured mRNA).

Following are the processes that occur in case of eukaryotic mRNA processing.

1. 5` mRNA capping.
2. Splicing.
3. 3` polyadenylation.

1. 5` mRNA capping: the unusual nucleotide 7-methyl-guanosine is added to 5` end via tri-phosphate linkage (an unusual linkage). This cap helps mRNA in locating the ribosomes for protein synthesis.
2. Splicing: in this process, the portions of mRNA that are called introns are removed. The remaining portions of mRNA other than introns are called exons. These exons are joined together to form matured mRNA.
3. 3` Polyadenylation: the poly-A tail added at the 3` end of mRNA by the enzyme polyA-polymerase. This poly-A tail is bound by a protein called poly-A tail binding protein. This protein protects the mRNA from degradation by ribonucleases (*the ribonucleases degrades the mRNA from 3`end*).