#### SHRIMATI INDIRA GANDHI COLLEGE

Nationally Reaccredited at "A" Grade (3rd cycle) by NAAC

# TRANSCRIPTION

Mrs.A.Shanmugapriya
Assistant professor, Department of Biochemistry
Shrimati Indira Gandhi College
Tiruchirappalli

**Class: I M.Sc Biochemistry** 

**Code: P16BC21** 

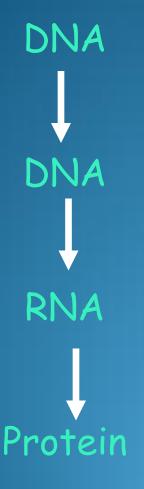
**Subject: Molecular Biology** 

**Semester: II** 

# **Synopsis**

- Introduction
- Prokaryotic transcription
- Eukaryotic transcription
- mRNA splicing
- Capping
- Poly A tailing
- Processing of rRNA in prokaryotes
- Processing of rRNA in Eukaryotes
- Processing of tRNA in prokaryotes
- Inhibitors of transcription

# DNATRANSCRIPTION



Replication

Transcription

Initiation Elongation Termination

Translation

-Initiation Elongation Processing Protein Targeting

# Transcription

Transcription is the process through which a DNA sequence is enzymatically copied by an RNA polymerase to produce a complementary DNA

Types: Prokaryotic and Eukaryotic Transcription

## **Prokaryotic Transcription**

Requirements: Template, Ribonucleotide triphosphates, RNA polymerase, metal ions and Rho protein.

#### **Steps in Prokaryotic Transcription**

I. Initiation II. Elongation III. Termination

**Initiation:** DNA polymerase recognize promoter region in DNA.

DNA is unwound and becomes single stranded called open complex.

The first nucleotide is added to the initiation complex.

**Elongation:** Proceeds by the addition of 2<sup>nd</sup> nucleotide in 5'-3' direction.

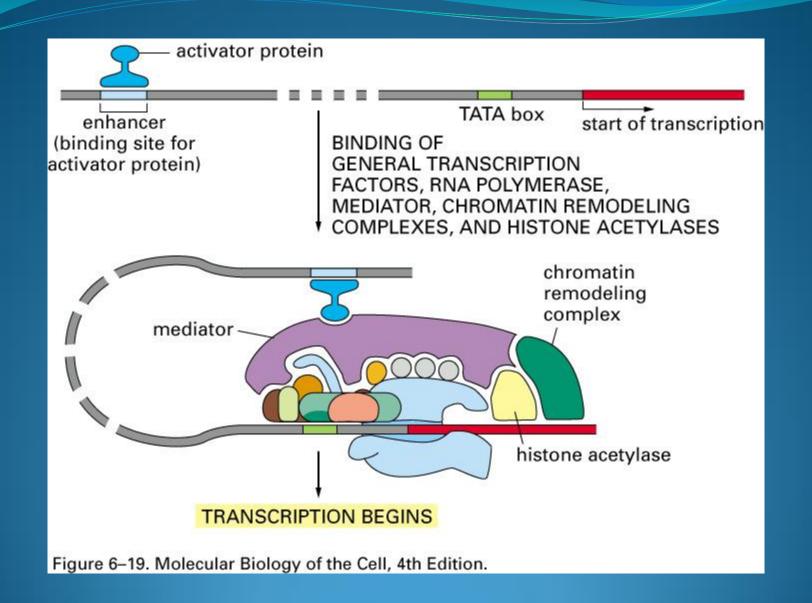
After addition of 10 bases the sigma factor dissociates and it becomes available for another initiation of RNA polymerase.

.

#### **Termination:** i. Rho dependent ii. Rho independent

Rho dependent: A specific protein, named p factor, binds to the growing RNA or weakly to DNA and in the bound state it acts as ATPase and terminates transcription and releases RNA. The p factor is also responsible for the dissociation of RNA polymerase from DNA.

Rho independent: In involves termination sequences within RNA to stop by forming stem- loop hairpin structure. This occurs due to the presence of palindromes. A palindrome is a word that reads alike forward and backward e.g. madam, rotor. The presence of palindromes in the base sequence of DNA template in the termination region is known.



### **Eukaryotic Transcription**

Requirements: DNA template, Ribonucleotide triphosphates, RNA polymerase, metal ions, Transcription factors.

**RNA Polymerase:** Eukaryotes consist of three types of RNA polymerase namely I, II and III, each comprising of two large subunits and 12-15 smaller subunits. The two large subunits are homologous to the E. coli β and β' subunits. Two smaller subunits are similar to the E. coli α sub- unit. The eukaryotic RNA polymerase does not contain any subunit similar to the E. coli Sigma factor. Therefore, initiation should be mediated by other proteins

Most of the eukaryotic protein-coding genes contain segments called introns, which break up the amino acid coding sequence into segments called exons. The transcript of these genes is resulted as premRNA (precursor-mRNA).

The pre-mRNA is then processed in the nucleus where the introns are removed and the exons are spliced together into a translatable mRNA.

The mRNA subsequently released from the nucleus and are translated in to proteins in the cytoplasm by ribosomes.

#### **Mechanism of Transcription:**

- I. Preinitiation
- II. Initiation
- III. Promoter clearance
- IV. Elongation
- V. Termination

**Preinitiation:** In eukaryotes a sequence of DNA bases known as Hogness box is located on left from upstream and another site called CAAT box. These sites are identified by RNA polymerase.

- <u>Initiation</u>: A collection of proteins called transcription factors mediate the binding of RNA polymerase and initiation of transcription.
- <u>Transcription factors</u>: TFIIA, TFIIB, TFIID, TFIIF, TFIIE, TFIIH and TBP associated factors.
- Promoter clearance: After the first bond is synthesized RNA polymerase must clear the promoter. Once the transcript reaches 23 nucleotides promoter clearance occur by phosphorylation.
- **Elongation:** TFIIE and TFIIF remain associated throughout elongation. RNA polymerase II is dephosphorylated.

# **Inhibitors of Transcription**

Actinomycin D – intercalates between G-C base pairs.

Acridine – inhibit RNA synthesis

Rifampicin – binds to beta subunit and prevent promoter clearance

# Post transcriptional modification

Newly synthesized RNA

Primary Transcript

tRNA & rRNA mRNA

#### **RNA Splicing**

Eukaryotic mRNA contains non coding sequence called introns separated by Exons.

In a process called splicing introns are removed and exons are joined to form a continuous sequence.

#### **Capping of mRNA**

- 5'cap is formed by condensation of GTP at 5' end.
- Guanine is methylated at N-7 and additional methyl groups are added at 2'OH end of first and second nucleotides adjacent to the cap.

#### **Poly A tailing**

- Poly A tailing in Eukaryotes protects mRNA
- Cleavage generates free hydroxyl group to which poly A tail are added by poly adenylate polymerase.

# Processing of rRNA in prokaryotes

Pre ribosomal RNA

1

30s RNA precursor

1

Methylation of 2'OH of ribose

1

Cleavage

1

Mature RNA( 16s, 23s, 5s)

# **Processing of rRNA in Eukaryotes**

Pre ribosomal RNA 45s RNA precursor Methylation of 2'OH of ribose Cleavage Mature RNA(18s, 5.8s, 28s)

# Processing of tRNA in prokaryotes and Eukaryotes

Removal of 5' leader sequence by RNase P



Removal of 3' trailer sequence by endo and exonuclease



Addition of CCA to 3' end



Splicing of introns in eukaryotes



Modification of multiple residues



Separation of multiple tRNA

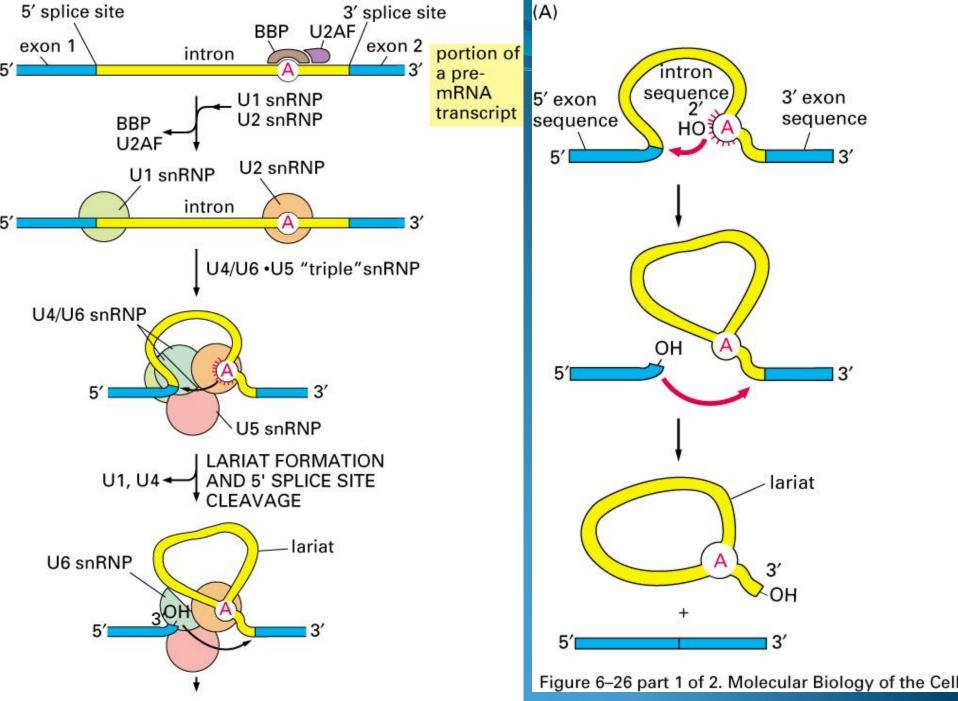


Figure 6-29 part 1 of 2. Molecular Biology of the Cell, 4th Edition.

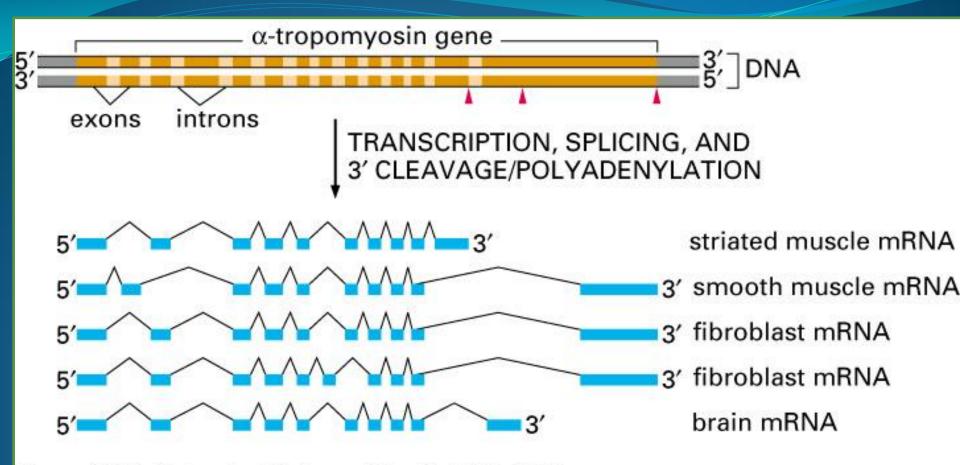


Figure 6–27. Molecular Biology of the Cell, 4th Edition.

## **Transcript export**

Proteins associated with mRNA mark it for export

Only mature mRNA is exported from nucleus

Exit via nuclear pore complexes

### **Possible Questions**

- 1. Write in brief about Prokaryotic RNA polymerase
- 2.Explain about Prokaryotic and Eukaryotic Transcription
- 3. Describe Eukaryotic RNA polymerase
- 4. Define promoters and Enhancers
- 5.Explain Rho dependent and Rho independent termination
- 6. Write a note on Transcriptional inhibitors
- 7. Describe Poly "A" tailing and Capping
- 8. Enumerate on RNA splicing
- 9. Write an essay on post transcriptional modification of tRNA and rRNA in prokaryotes and Eukaryotes.