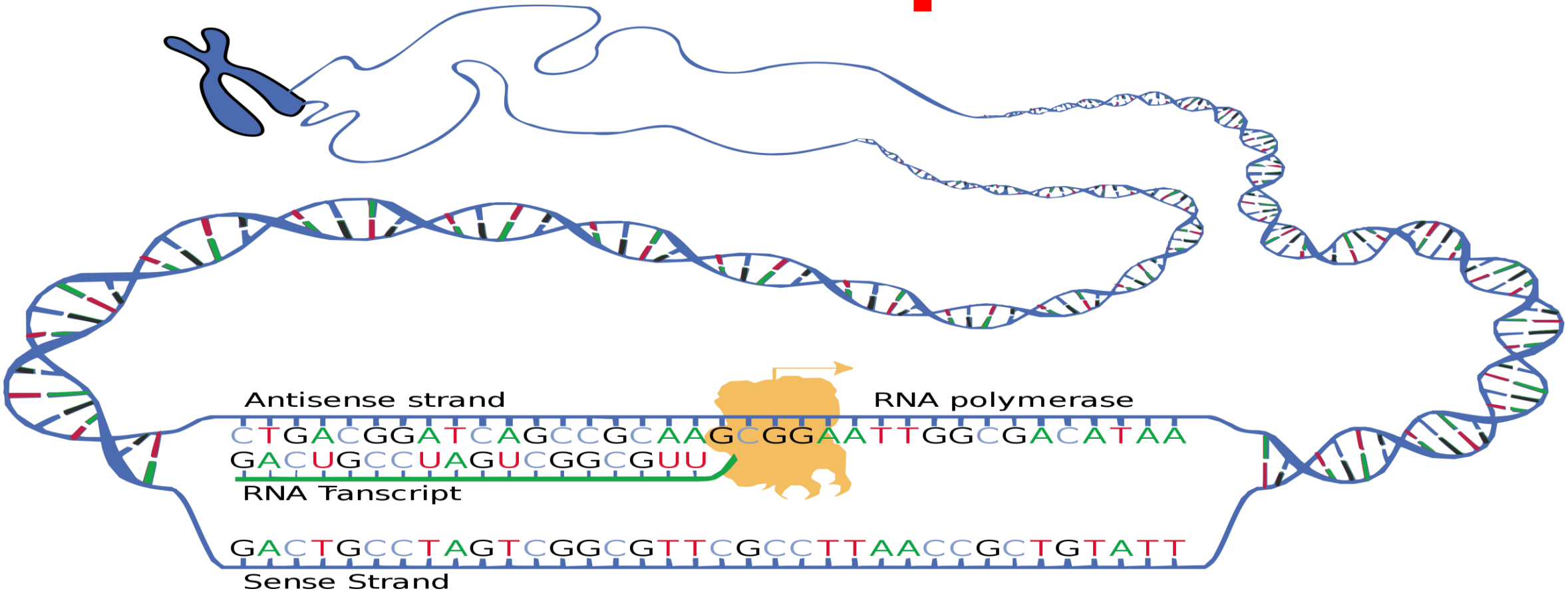
DNA Transcription



Transcription

- **Transcription, is the process of creating an equivalent RNA copy of a sequence of DNA.**

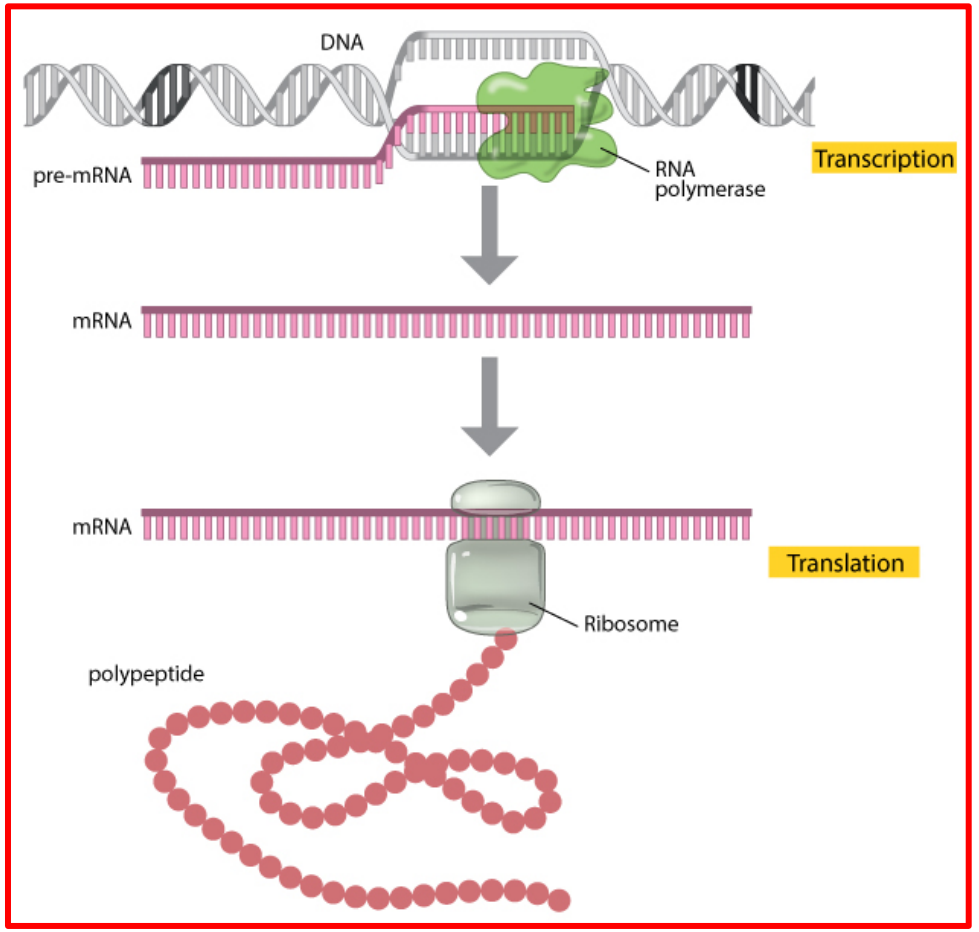
- **Transcription is the first step leading to gene expression.**

  Transcription is the first of several steps of DNA based gene expression in which a particular segment of DNA is copied into RNA by the enzyme RNA polymerase. Both DNA and RNA are nucleic acids

- Double stranded DNA must be TRANSCRIBED into Single stranded RNA
- **1. mRNA "messenger"**
- made from DNA in nucleus…travels out of nucleus and finds a ribosome.
- **2. tRNA "transfer"**
- brings amino acids to the ribosomes; found in cytoplasm
- **3. rRNA "ribosomal"**
- part of the ribosome; this is where proteins are made

How does the DNA get made into RNA and that made into Protein???

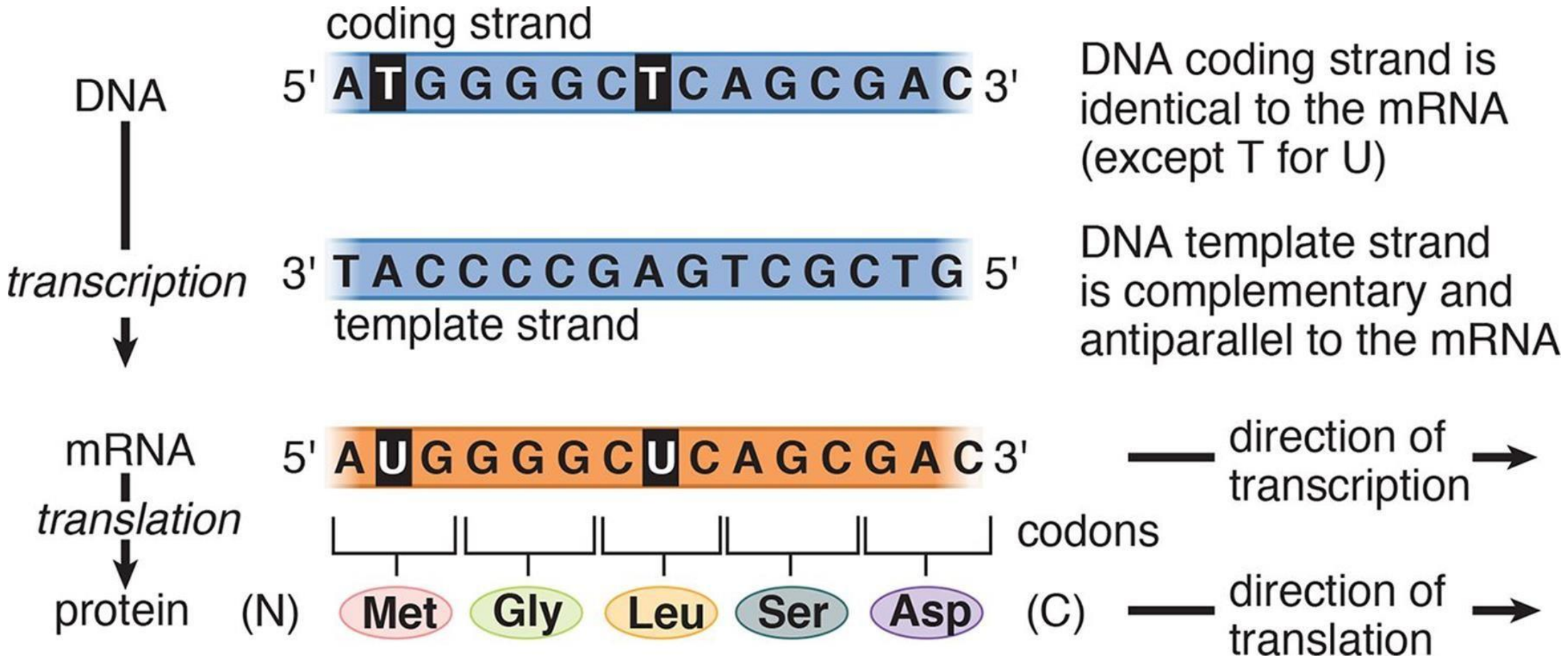
During transcription, the enzyme RNA polymerase (green) uses DNA as a template to produce a pre-mRNA transcript (pink). The pre-mRNA is processed to form a mature mRNA molecule that can be translated to build the protein molecule (polypeptide) encoded by the original gene



Transcription in prokaryote

- RNA molecules are produced by copying part of a nucleotide sequence of DNA into a complementary sequence in RNA. This Process is called **transcription**.
- Transcription requires the enzyme **RNA polymerase**.
- **During transcription, a DNA sequence is read by RNA polymerase, which produces a complementary, antiparallel RNA strand.**
- **Transcription results in an RNA complement that includes uracil (U) instead of thymine (T).**

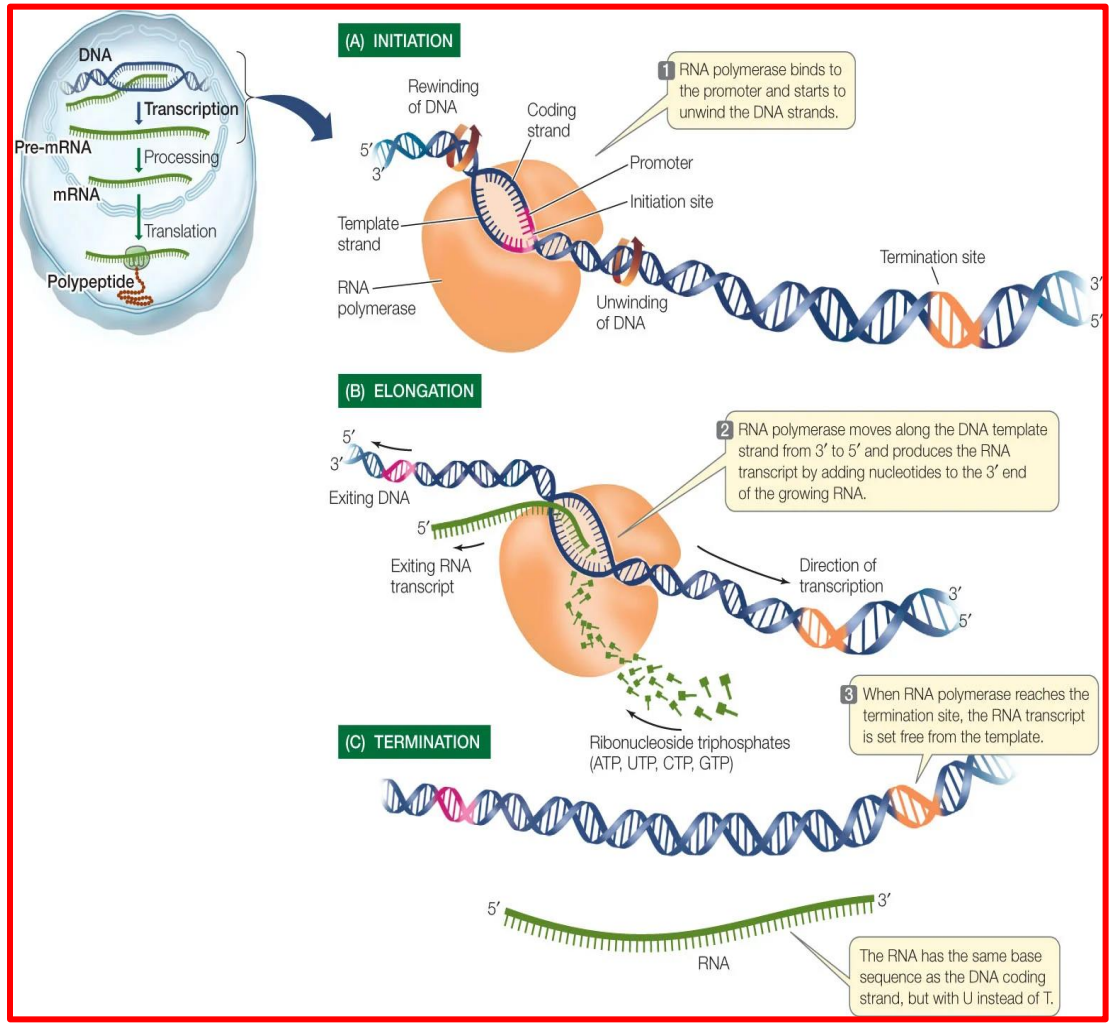
Coding and Non–Coding Strands

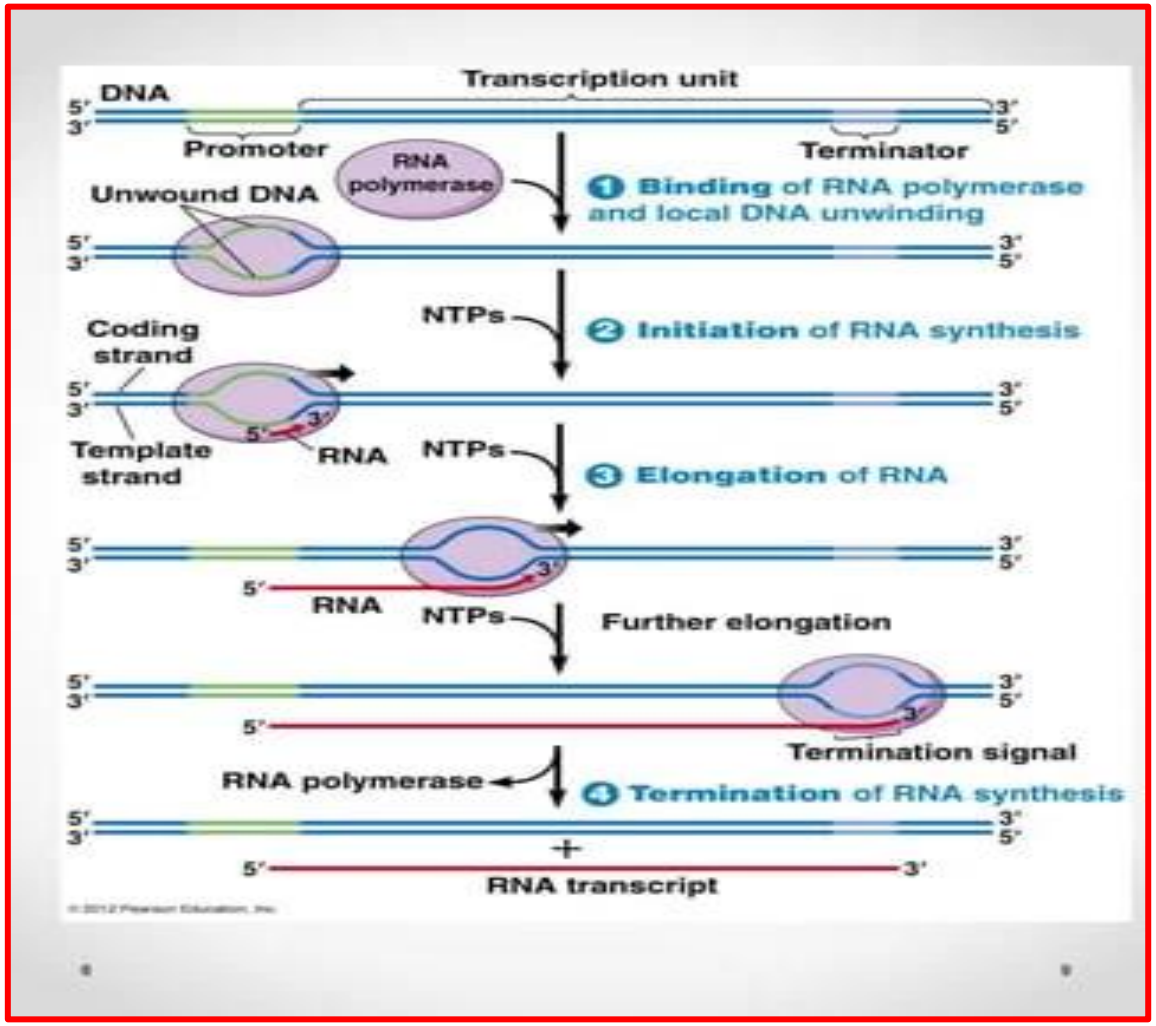


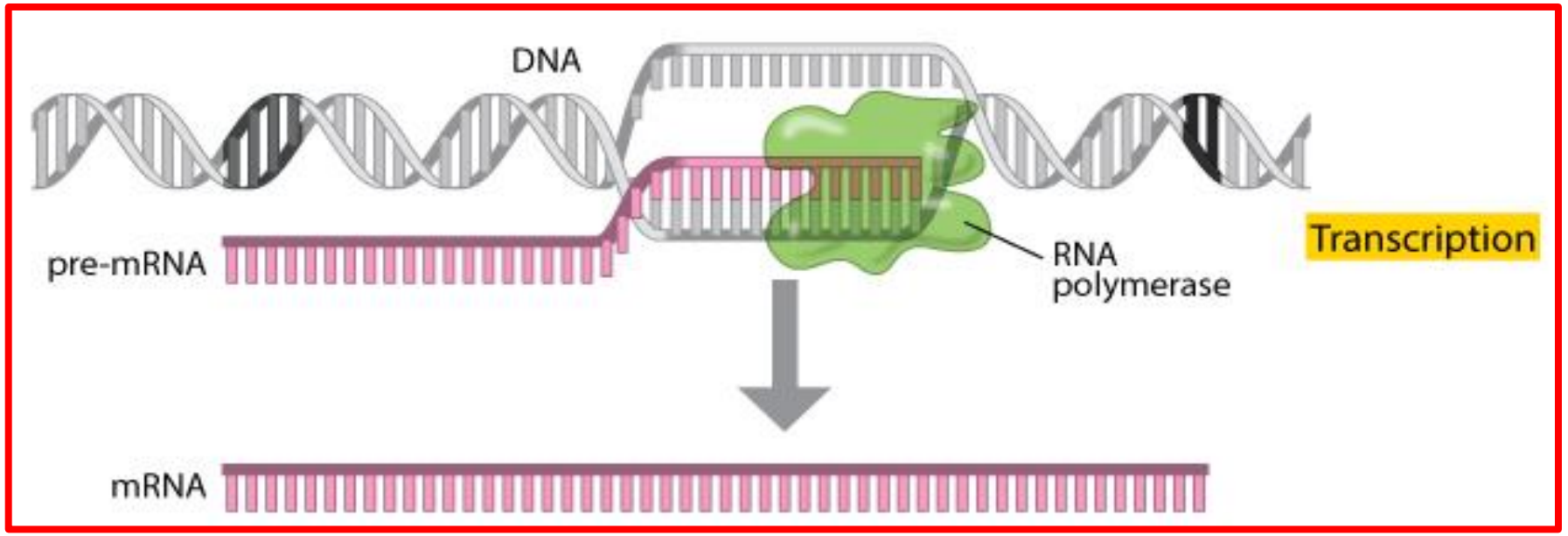
Stages of Transcription in Prokaryote

- **Initiation**
- RNA polymerase attaches to the DNA molecule and moves along the DNA strand until it recognizes a promoter sequence(**Sequence on DNA**
- **10 and 35 bases upstream of start site**). These are known as the transcription start sites. The DNA double helix then unwinds and all the bases on each of the DNA strands are exposed. This acts as a template for a new mRNA strand.
- **Elongation**
- Ribonucleotides are added to the template strand that enables the growth of mRNA growth.
- **Termination**
- RNA polymerase encounters a terminator sequence and the transcription stops. RNA polymerase then releases the DNA template.

Stages of Transcription







(5′) C G C T A T A G C G T T T (3′) DNA nontemplate (coding) strand

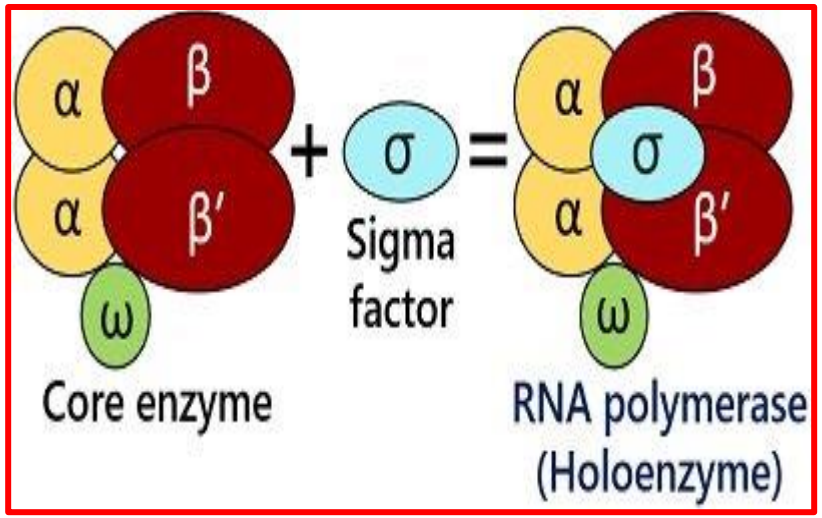
(3′) G C G A T A T C G C A A A (5′) DNA template strand

(5′) C G C U A U A G C G U U U (3′) RNA transcript

Figure 26-2
Lehninger Principles of Biochemistry, Fifth Edition

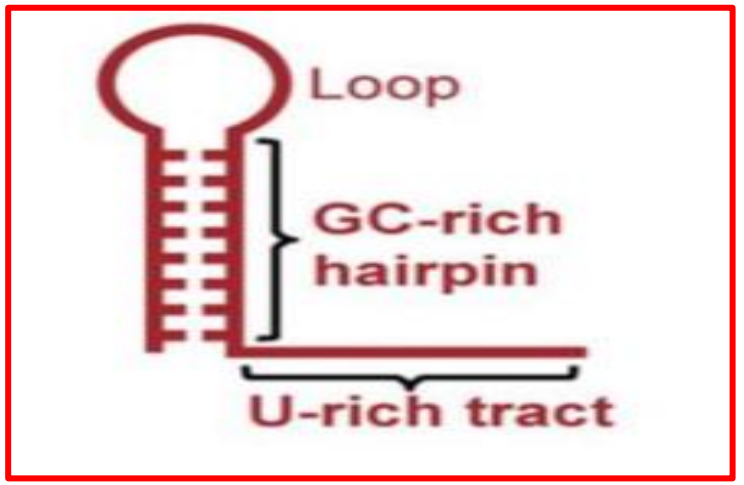
RNA polymerase in prokaryotic

- In most prokaryotes, **a single RNA polymerase species transcribes all types of RNA**. RNA polymerase "core" from E. coli consists of five subunits: two alpha (α) subunits of 36 kDa, a beta (β) subunit of 150 kDa,
- a beta prime subunit (β′) of 155 kDa,
- and a small omega (ω) subunit.
- **Sigma (σ) factor**
- *Smaller protein
- *Guides RNA polymerase to target DNA sequence
- * Sigma factor released after open complex (unwinds DNA at promoter).



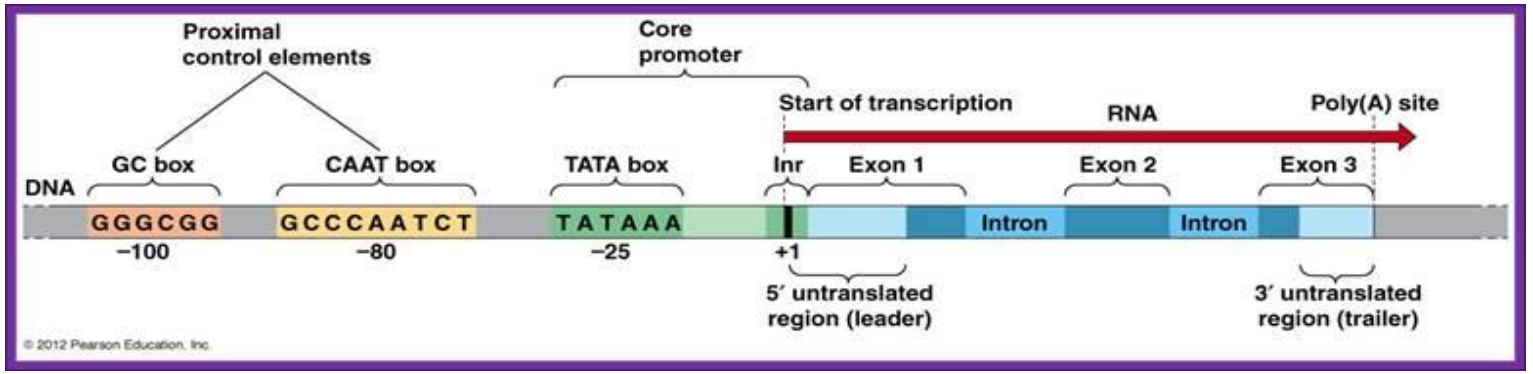
Transcription Termination

- Rho-dependent termination
- Rho (ρ) factor binds to mRNA
- Slides along mRNA to polymerase
- Breaks polymerase, mRNA off of DNA
- The ρ factor, a hexamer, is a ATPase and a Helicase
- Rho-independent termination
- The termination signal is a stretch of 30-40 nucleotides on the RNA
- transcript, consisting of many GC followed by a series of U.
- • The sequence specificity of this nascent RNA transcript will form
- particular stem-loop structures to terminate the transcription.



Eukaryotic Transcription

- Promoters
- Much more complex than those found in bacteria.
- These are consensus sequences located at the upstream regions of Coding strand.
- Mutation of this region usually significantly lowers the rate of transcription.

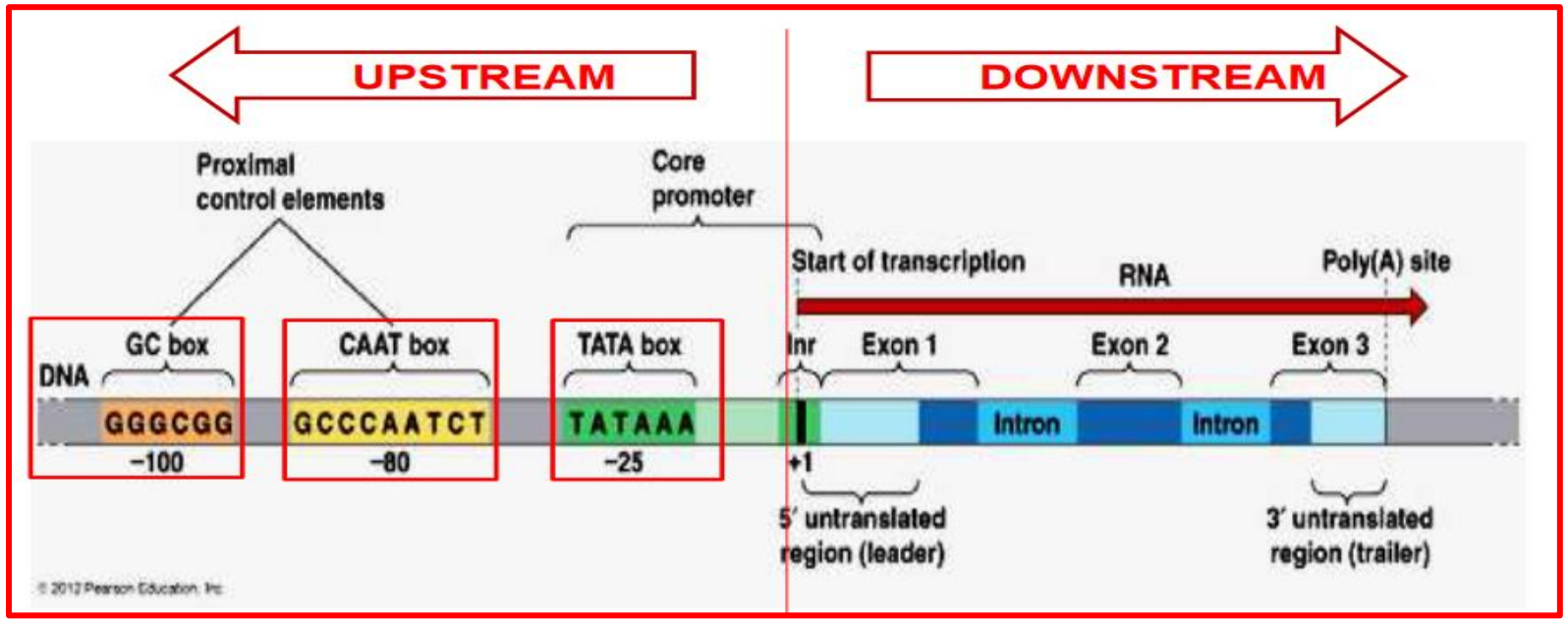


Promoters

- **1) TATA box (Hogness Box)**
- Very similar to the prokaryotic TATA box,except the sequence is slightly different
- (TATAAA) and it is located in between25 to -30.
- **2) CAAT box**
- Located in between -70 to -80.
- Always contains CCAAT.
- 3) GC box
- Usually has the sequence GGGCGG and is typically found at -110.

ENHANCERS

- Enhancers elements are the sequences located in a variety of regions of a gene both upstream and downstream of the transcription start site and even within the transcribed portions of some genes.
- Enhancers increases the transcription rate by several folds.

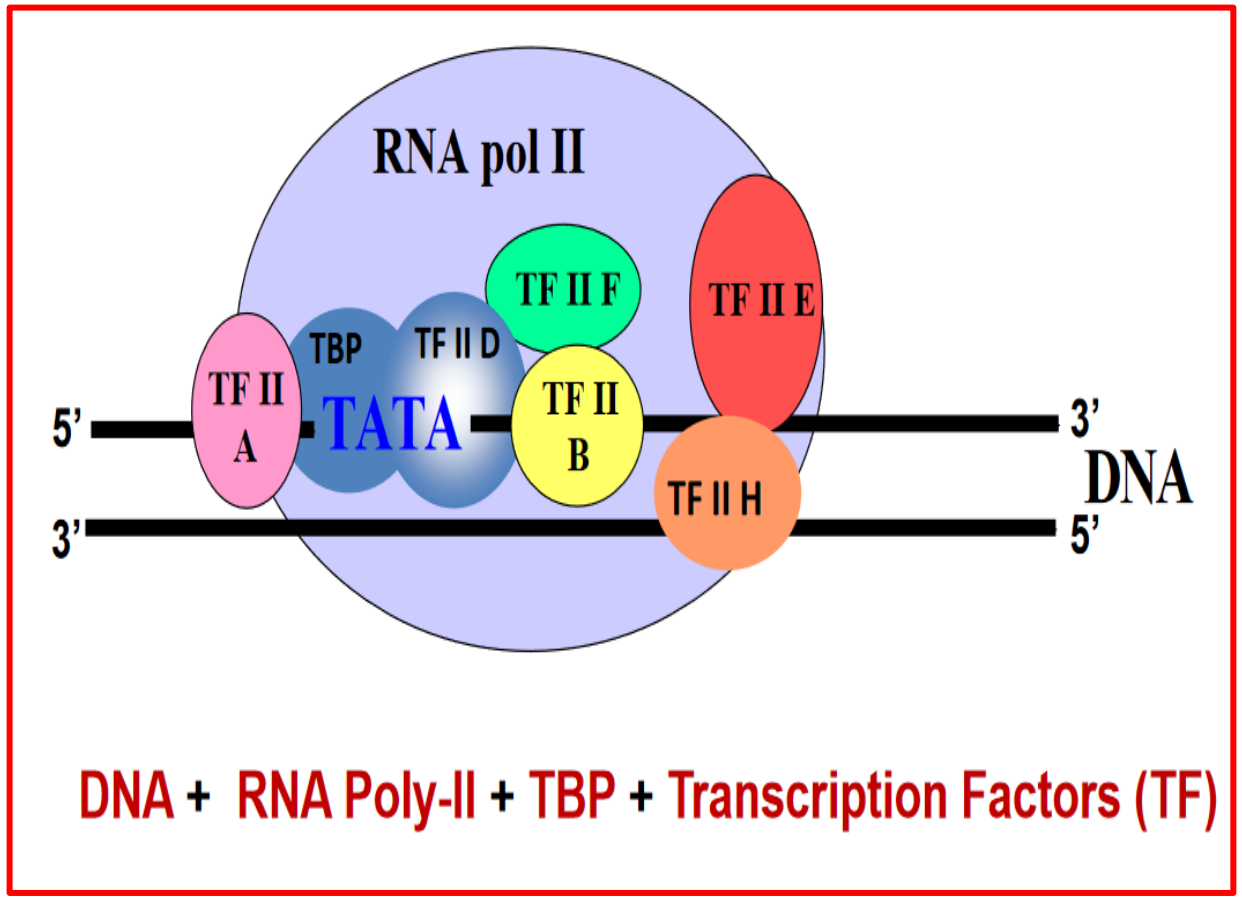


Transcription factors

- RNA-pol II does not bind to the promoter sequences directly.
- RNA-pol II associates with six transcription factors.
- TFII A, TFII B, TFII D, TFII E, TFII F and TFII H.

Pre-initiation complex (PIC)

- TBP of TFII D binds TATA–Box(-10 sequence)
- TFII A and TFII B bind TFII D
- TFII F- RNA-pol complex binds TFII B
- TFII F and TFII E open the dsDNA
- (helicase and ATPase)
- TFII H: completion of PIC.



Elongation

- During elongation, **the transcription machinery needs to move histones out of the way every time it encounters a nucleosome**.
- TFIIF remains associated with RNA Pol-II throughout elongation.
- The activity of the RNA poly-II is greatly enhanced by proteins called Elongation factors

Termination

- When the RNA Polymerase transcribes the terminator region of the DNA, the polymerase releases the mRNA
- The termination sequence is AATAAA followed by GT repeats.

Post-Transcriptional Modifications

Posttranscriptional modifications to eukaryotic pre-mRNA

Modification	Function
Addition of 5′ cap	Facilitates binding of ribosome to 5′ end of mRNA, increases mRNA stability, enhances RNA splicing
3′ cleavage and addition of poly(A) tail	Increases stability of mRNA, facilitates binding of ribosome to mRNA
RNA splicing	Removes noncoding introns from pre-mRNA, facilitates export of mRNA to cytoplasm, allows for multiple proteins to be produced through alternative splicing
RNA editing	Alters nucleotide sequence of mRNA

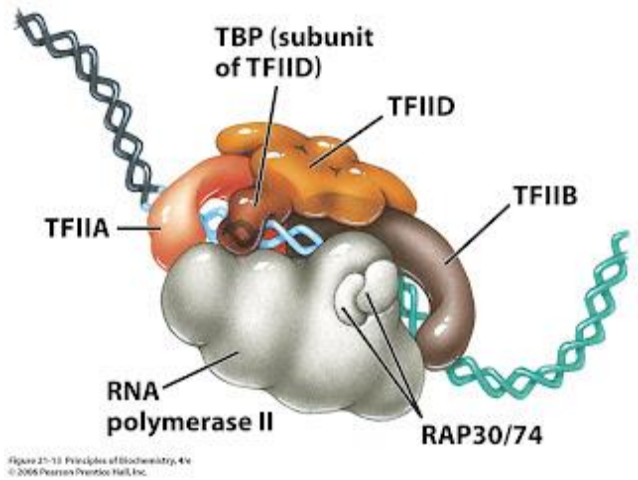
- **Is ALL the DNA transcribed into mRNA?**

- **NO!!!**
- Only certain sections of the DNA are made (transcribed)into message (mRNA)
- AND…
- Only part of the mRNA is actually used and sent out of the nucleus to meet up with a ribosome! This is **EDITING!**

- RNA editing is **a process through which the nucleotide sequence specified in the genomic template is modified to produce a different nucleotide sequence in the transcript**.
- **mRNA has same sequence as non-template strand**

RNA polymerase in Eukaryote

Name	Can be found in	Transcribes
RNA Polymerase I	All Eukaryotes	Large rRNAs
RNA Polymerase II	All Eukaryotes	mRNA, snoRNAs, some snRNAs and miRNAs
RNA Polymerase III	All Eukaryotes	tRNAs, small rRNAs, some snRNAs and miRNAs
RNA Polymerase IV	Plants	some siRNAs
RNA Polymerase V	Plants	RNAs important in heterochromatin formation



Prokaryotic vs Eukaryotic Transcription

Prokaryotic Transcription	Eukaryotic Transcription
Transcription and translation occur simultaneously	Transcription and translation don’t occur simultaneously.
Prokaryotic transcription occurs in the cytoplasm	Eukaryotic transcription occurs in the nucleus and translation occurs in the cytoplasm.
RNAs are released and processed in the cytoplasm	RNAs are released and processed in the nucleus
RNA polymerases are a complex of five polypeptides.	RNA polymerases are a complex of 10 -15 polypeptides.