Transcription

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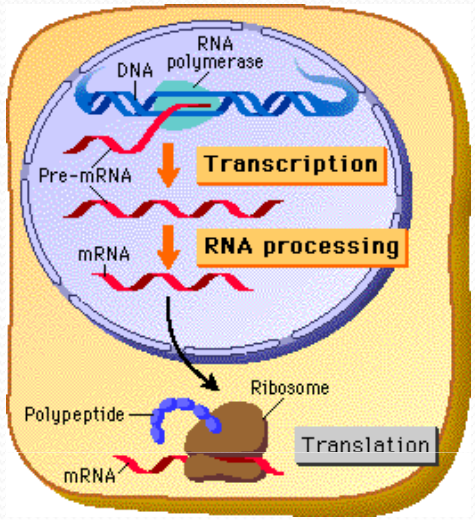
Transcription

It is the process of transcribing or making a copy of Genetic information stored in a DNA strand into a Complementary strand of RNA (messenger RNA or mRNA) with the aid of RNA polymerase.

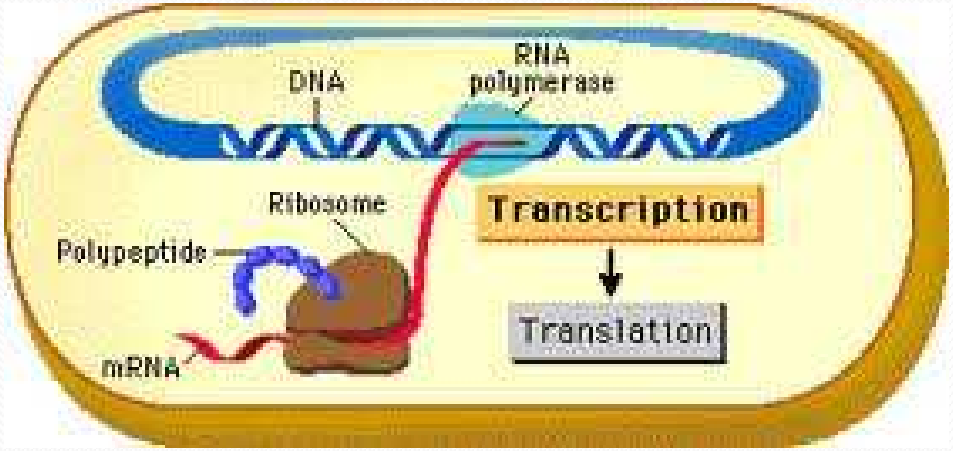
Or

The process by which RNA molecules are initiated, elongated and terminated is called transcription.

Transcription site in Prokaryotes & Eukaryotes



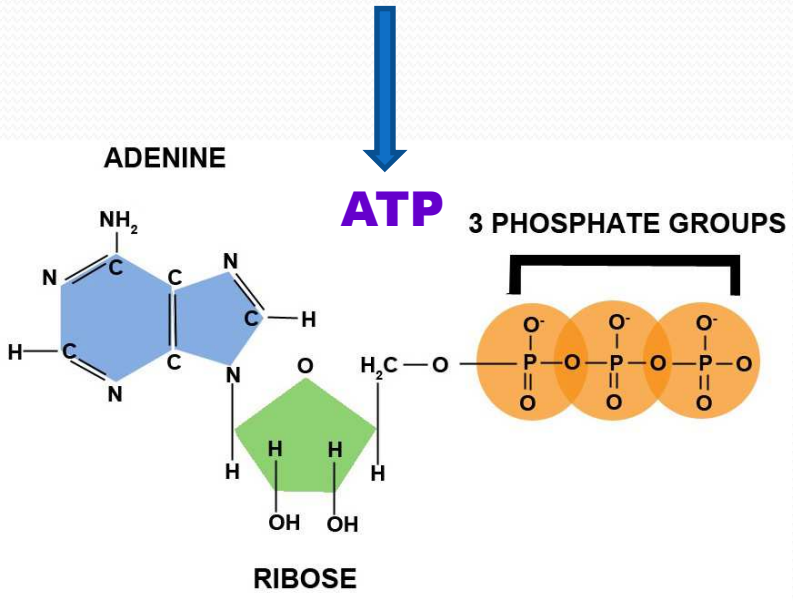
Eukaryotic transcription takes place in nucleus.



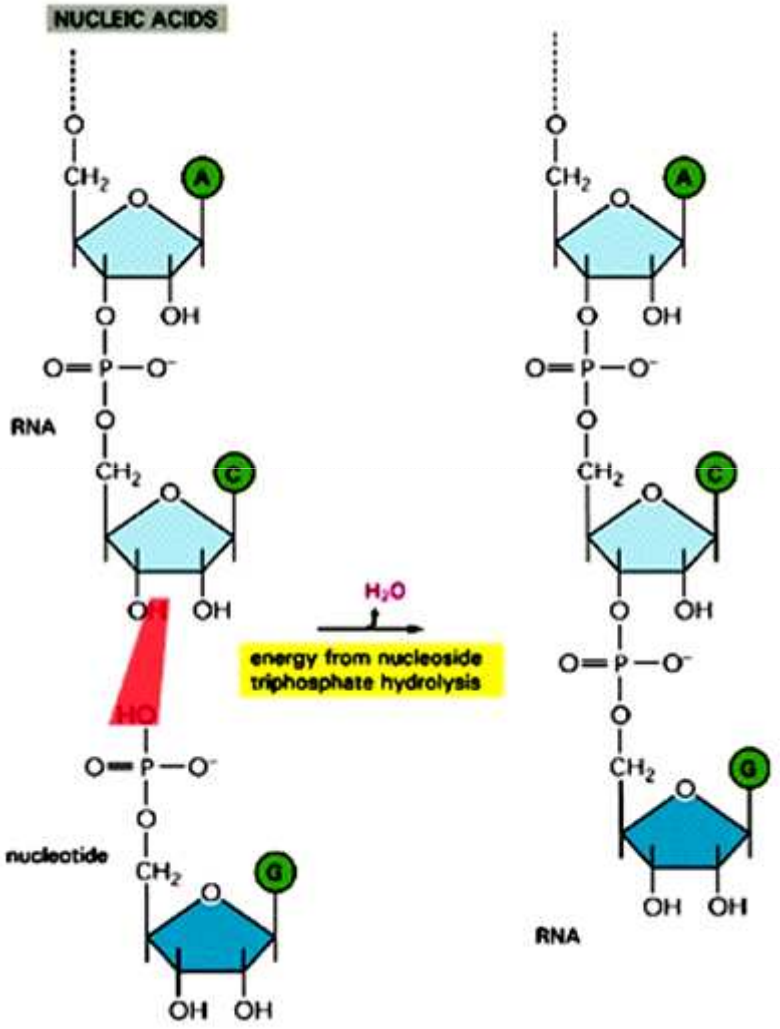
Prokaryotic transcription takes place in side the cell as there is no separation by nuclear membrane.

Basic Features of RNA Synthesis

1.Precursors i.e., the four ribonucleoside 5'-triphosphates (**ATP, GTP, CTP, UTP) are required**

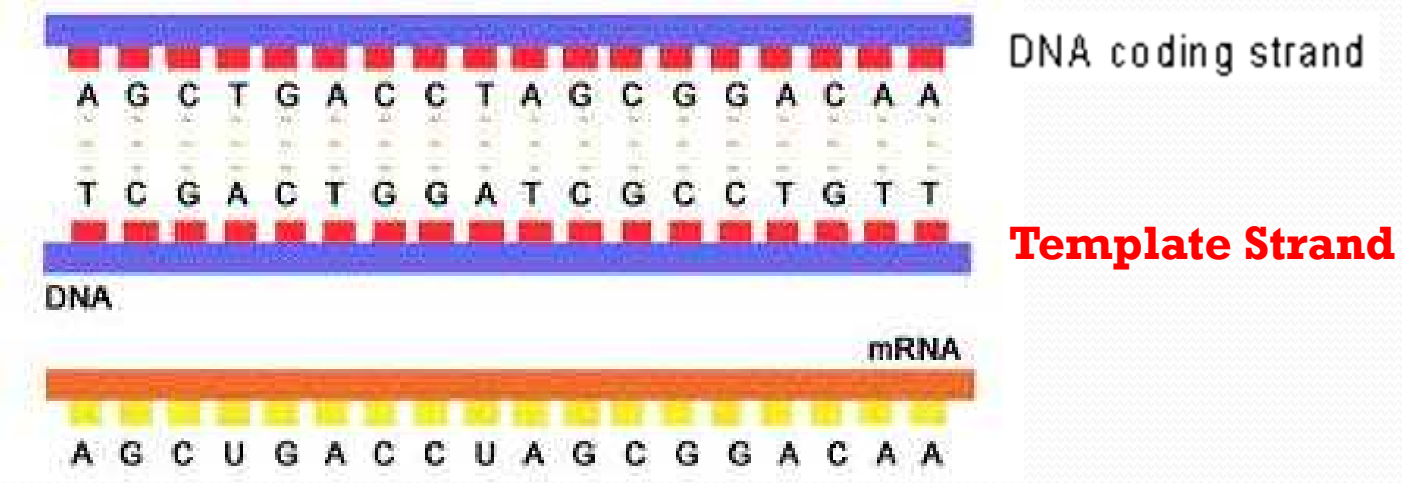


2. Polymerization Reaction

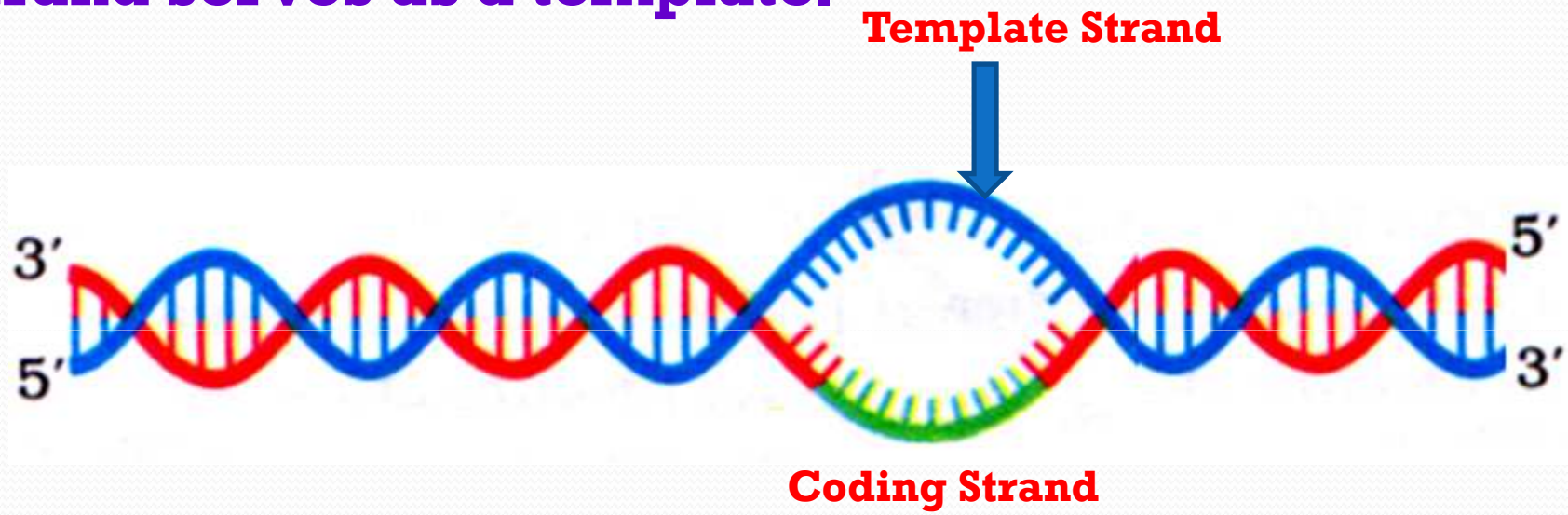


A 3'-OH group of one nucleotide reacts with the 5'-triphosphate of a second nucleotide;

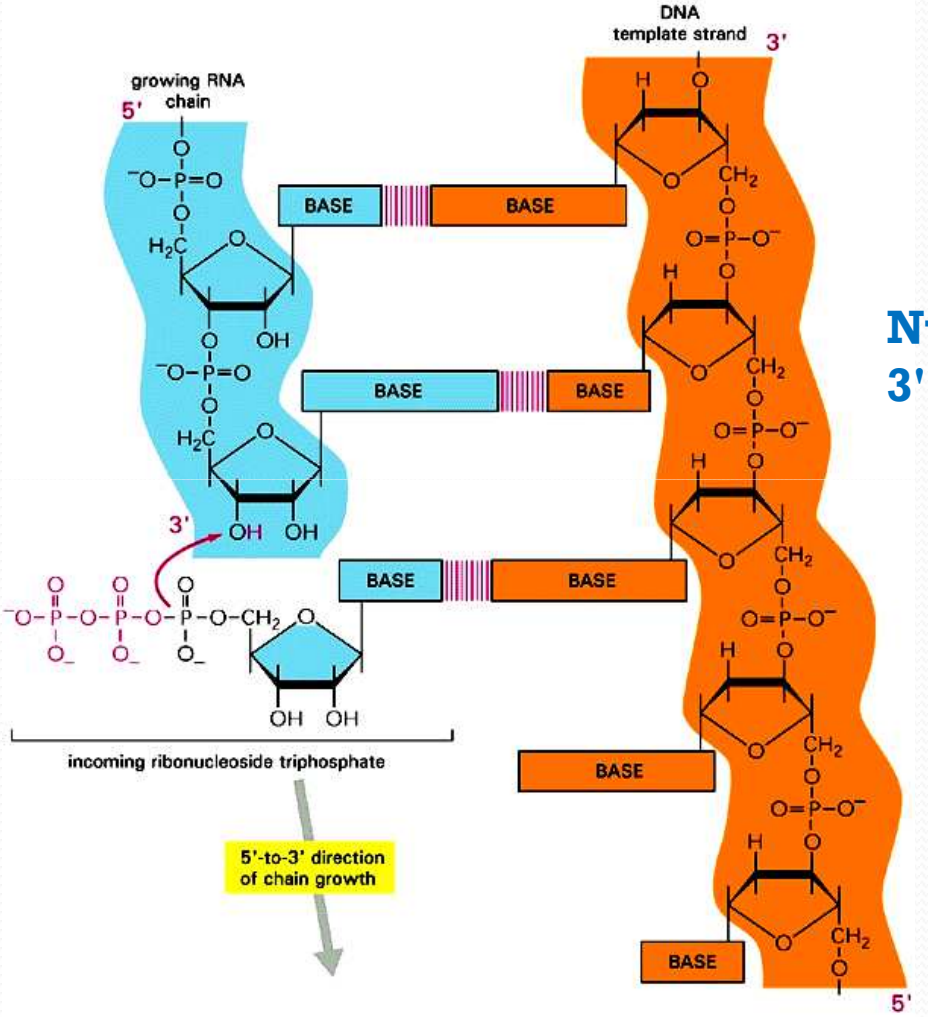
3.The sequence of bases in RNA molecule is determined by the base sequence of the DNA.



4. The DNA molecule being transcribed is double-stranded, yet in any particular region only one strand serves as a template.



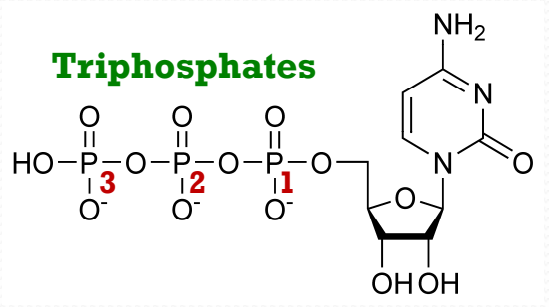
5. Chain Growth is 5’-3’

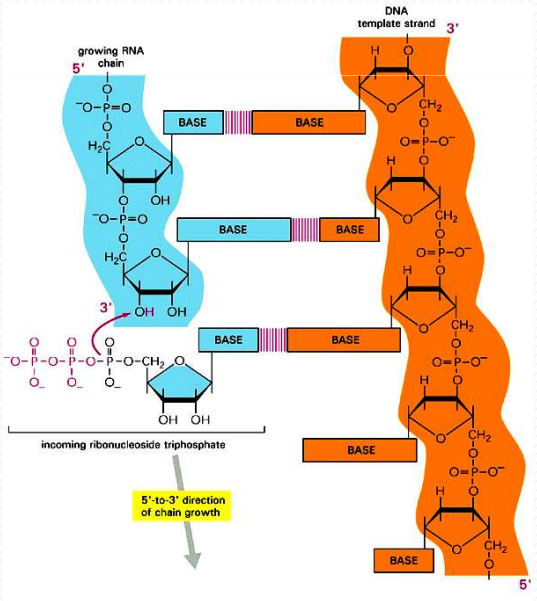


Nucleotides are added only to the 3' -OH end of the growing chain.

6. No primer Required

7. 5’ po4 & 3’OH…… The first base to be laid down in the initiation event is a triphosphate. Its 3' -OH group is the point of attachment of the subsequent nucleotide. Thus, the 5' end of a growing RNA molecule terminates with a triphosphate.

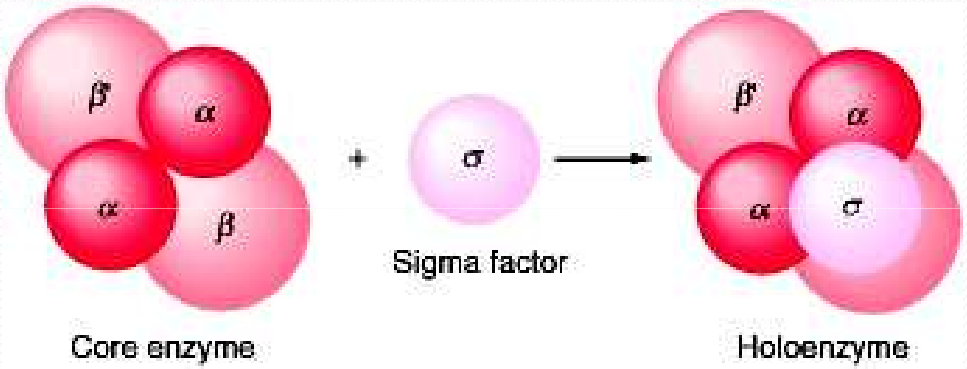


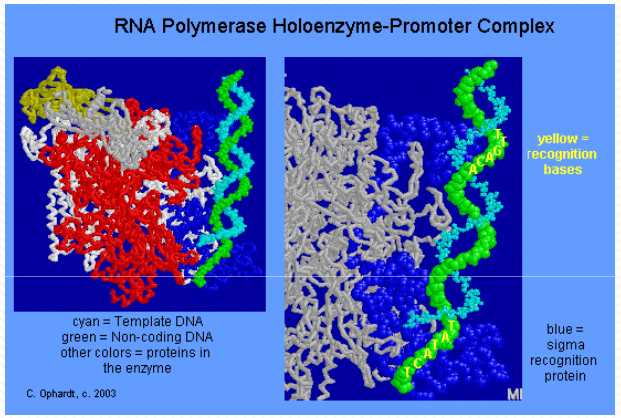


Requirements

1. **Template- Double stranded DNA**
2. **Activated precursors:- ATP, GTP, UTP, CTP**
3. **Divalent Metal Ions:- mg^{++}, mn^{++}**
4. **RNA polymerase:- Enzyme**

RNA polymerase is a key enzyme and was isolated from *E. coli* by Chamberlin and Berg (1962)





The term core enzyme is used to describe the σ-free unit-namely, α_2, β, β'.

The complete enzyme, α_2, β, β' σ is called the holoenzyme.

Sub Units of RNA polymerase and their functions

Sl. No.	Sub Unit	M.W.	Function
1	β’	1,60,000	**Binding of RNA polymerase to DNA *i.e.*, Template strand**
2	β	1,50,000	**Helps in binding with incoming nucleotides & forms 1st phosphodiester bond formation**
3	2 α	40,000	**Needed for assembly of core enzyme & Promoter Recognition**
4		90,000	**Recognition of start signal & directs to bind to the promoter region of DNA**

Promoter : A site in a DNA molecule at which RNA polymerase and transcription factors bind to initiate transcription of mRNA.

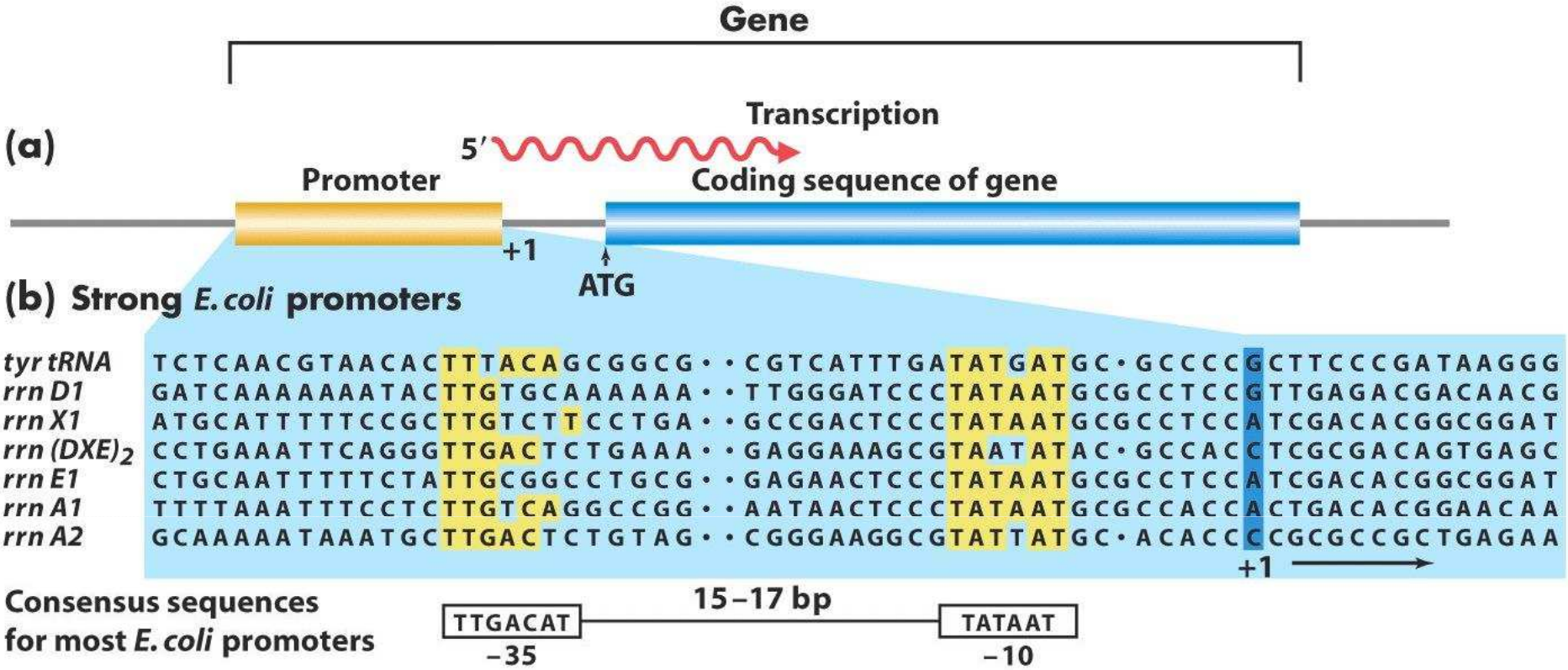
Transcription

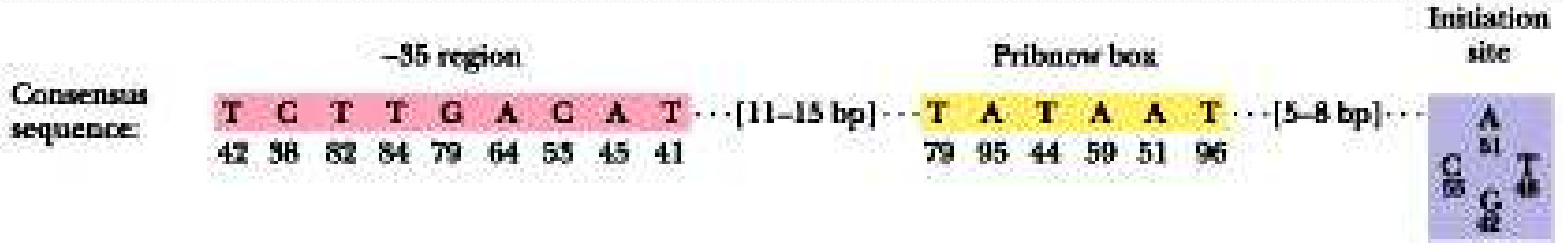
1. Initiation
2. Elongation
3. Termination

Template binding at a polymerase recognition site

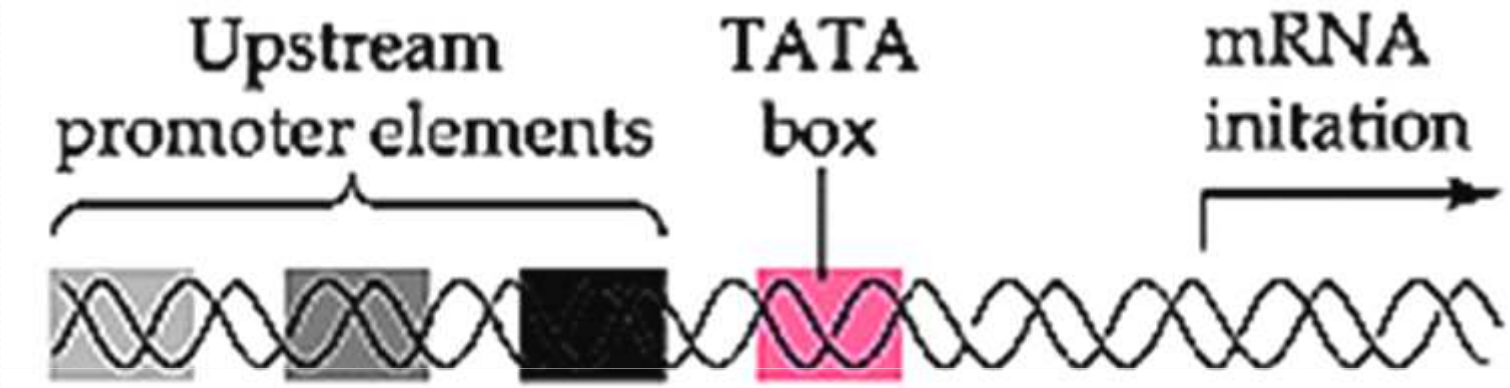
Binding to initiation site

Establishment of open promoter complex

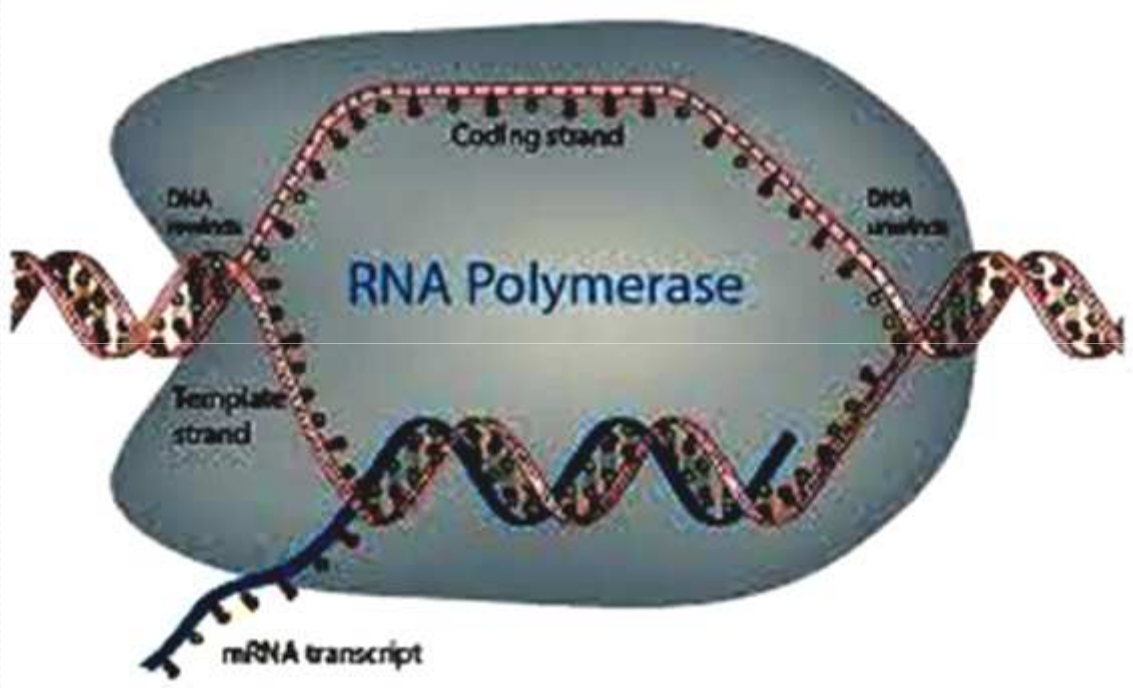

Gene
(a)
Transcription
5′
Promoter
Coding sequence of gene
+1
ATG
(b) Strong E. coli promoters
tyr tRNA TCTCAACGTAACACTTTACAGCGGCG··CGTCATTTGATATGATGC·GCCCCGCTTCCCGATAAGGG
rrn D1 GATCAAAAAAATACTTGTGCAAAAAA··TTGGGATCCCTATAATGCGCCTCCGTTGAGACGACAACG
rrn X1 ATGCATTTTTCCGCTTGTCTTCCTGA··GCCGACTCCCTATAATGCGCCTCCATCGACACGGCGGAT
rrn (DXE)2 CCTGAAATTCAGGGTTGACTCTGAAA··GAGGAAAGCGTAATATAC·GCCACCTCGCGACAGTGAGC
rrn E1 CTGCAATTTTTCTATTGCGGCCTGCG··GAGAACTCCCTATAATGCGCCTCCATCGACACGGCGGAT
rrn A1 TTTTAAATTTCCTCTTGTCAGGCCGG··AATAACTCCCTATAATGCGCCACCACTGACACGGAACAA
rrn A2 GCAAAAATAAATGCTTGACTCTGTAG··CGGGAAGGCGTATTATGC·ACACCCCGCGCCGCTGAGAA
+1
Consensus sequences for most E. coli promoters
TTGACAT
15–17 bp
TATAAT
–35
–10

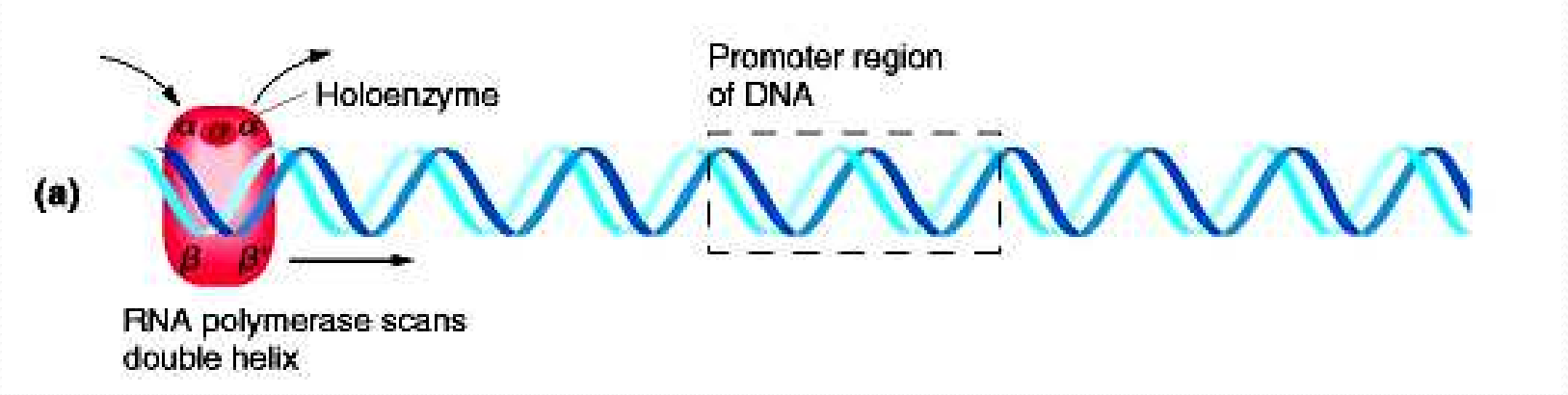

–35 region
Pribnow box
Initiation site
Consensus sequence:
T C T T G A C A T
42 38 82 84 79 64 55 45 41
[11–15 bp]
T A T A A T
79 95 44 59 51 96
[5–8 bp]
A 51
C 55
T 48
G 42

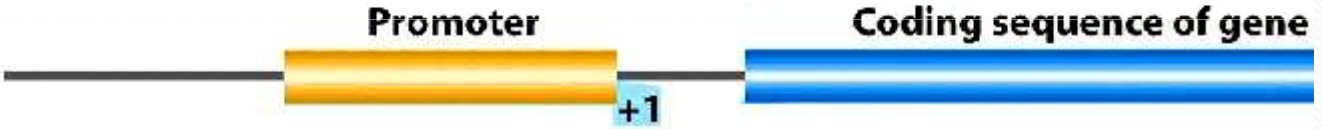
INITIATION OF SYNTESIS

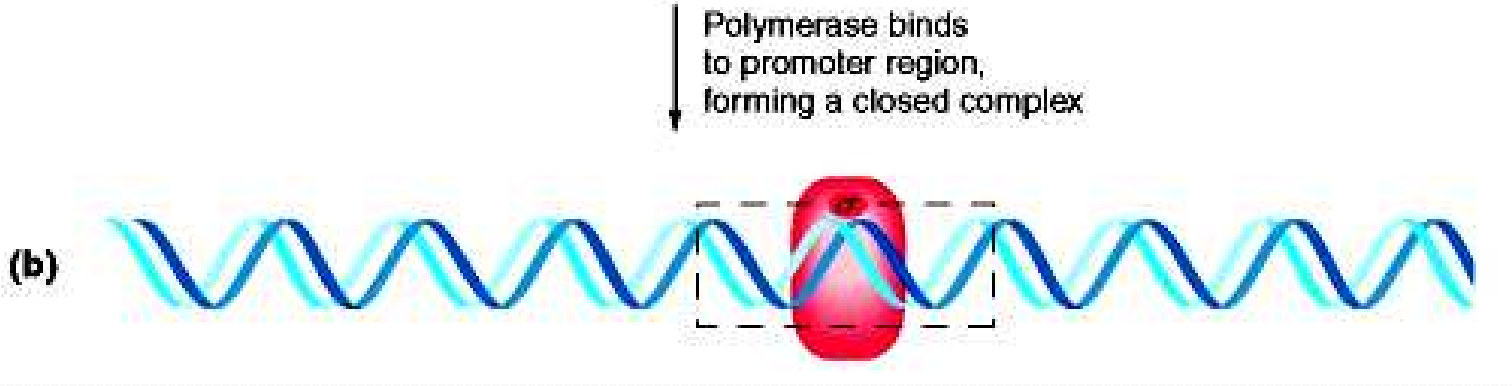


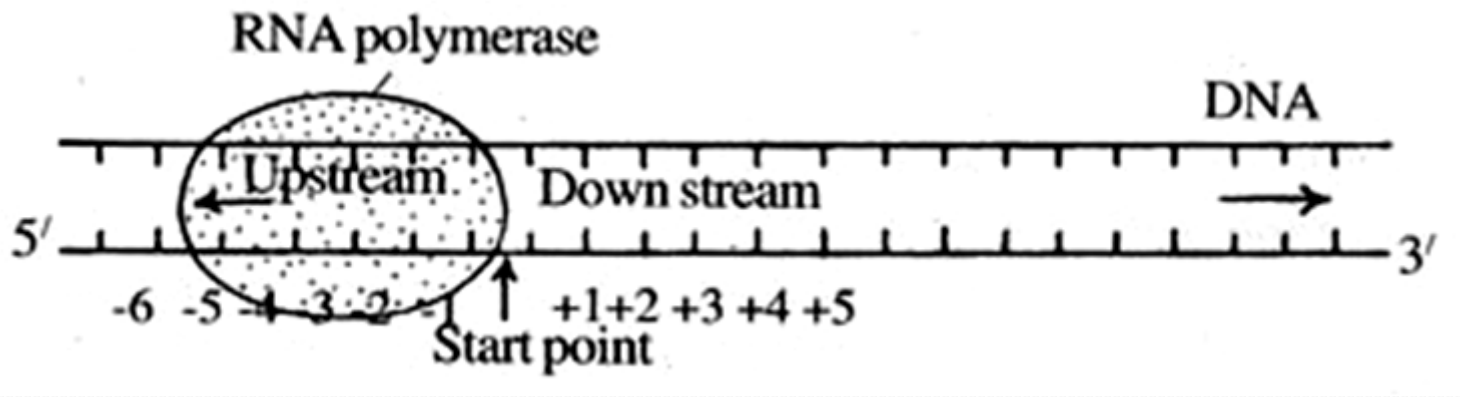
RNA polymerase is a huge enzyme it binds around 60 nucleotides at a time




(a)
Holoenzyme
Promoter region
of DNA
RNA polymerase scans
double helix


Promoter
Coding sequence of gene
+1


Polymerase binds
to promoter region,
forming a closed complex
(b)


RNA polymerase
DNA
Upstream
Down stream
5′
3′
-6 -5 -4 -3 -2 -1 +1 +2 +3 +4 +5
Start point

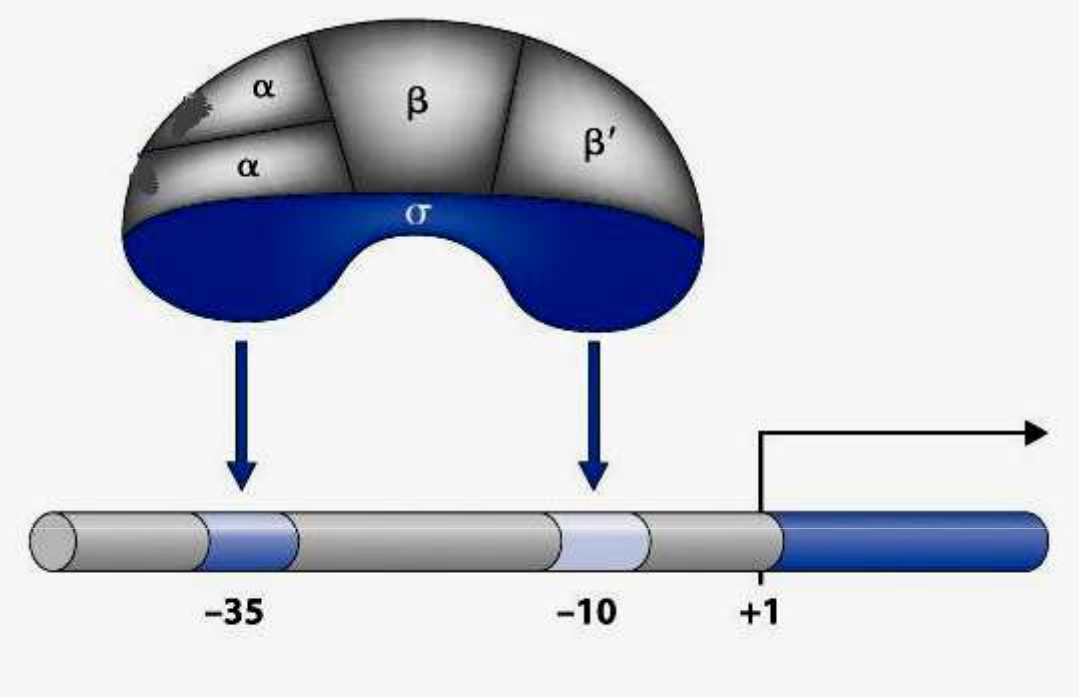
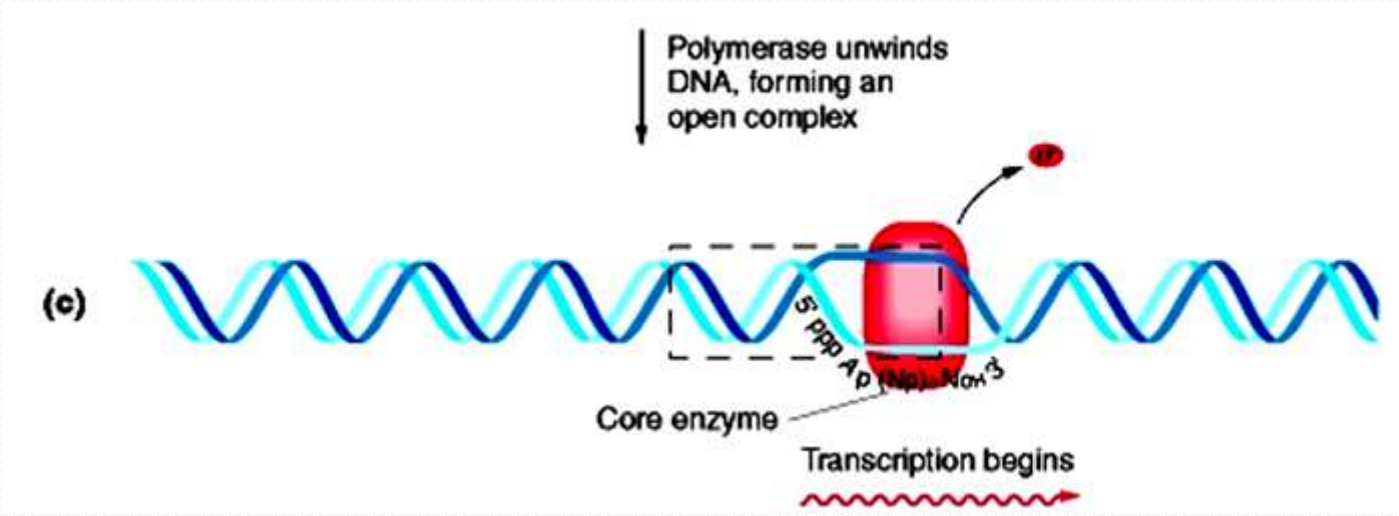
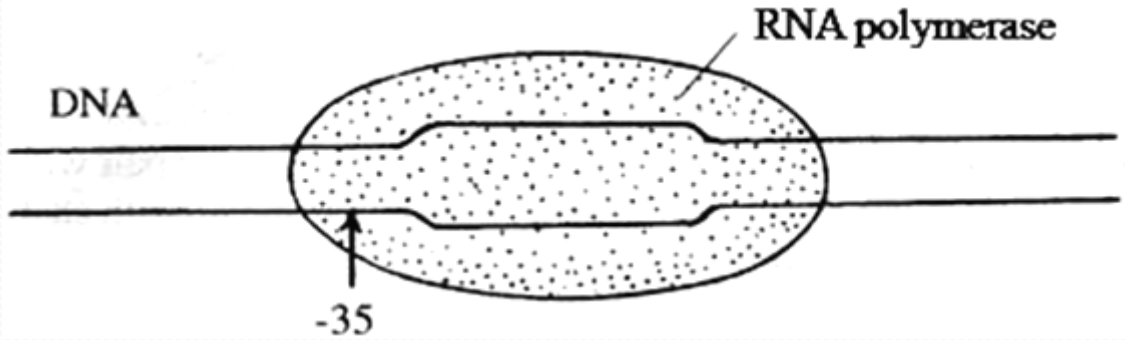
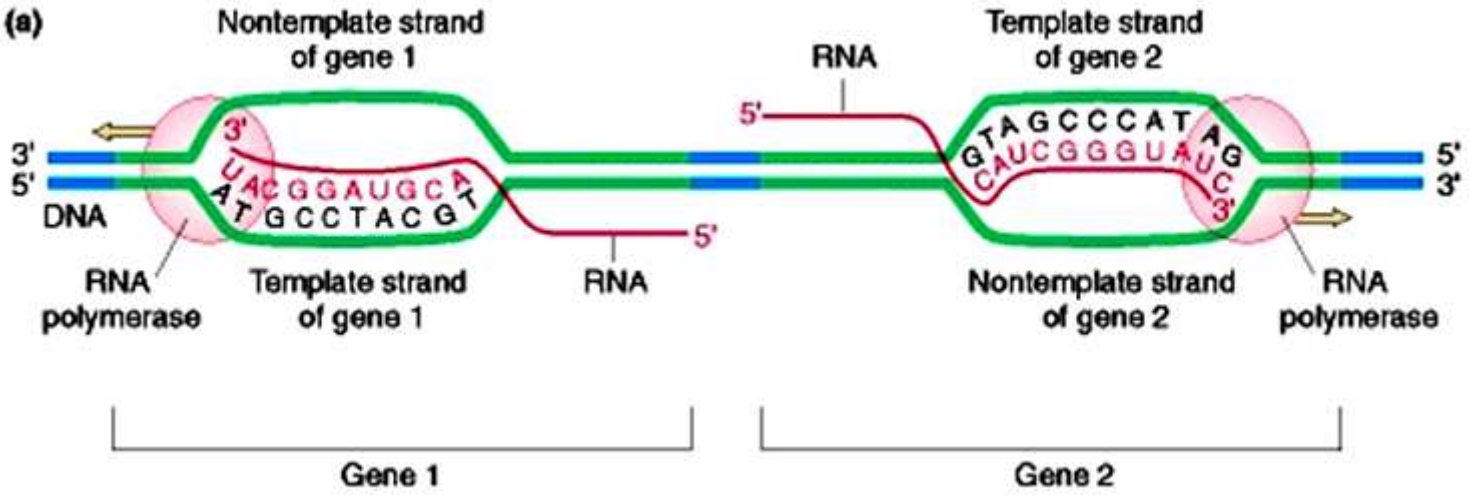
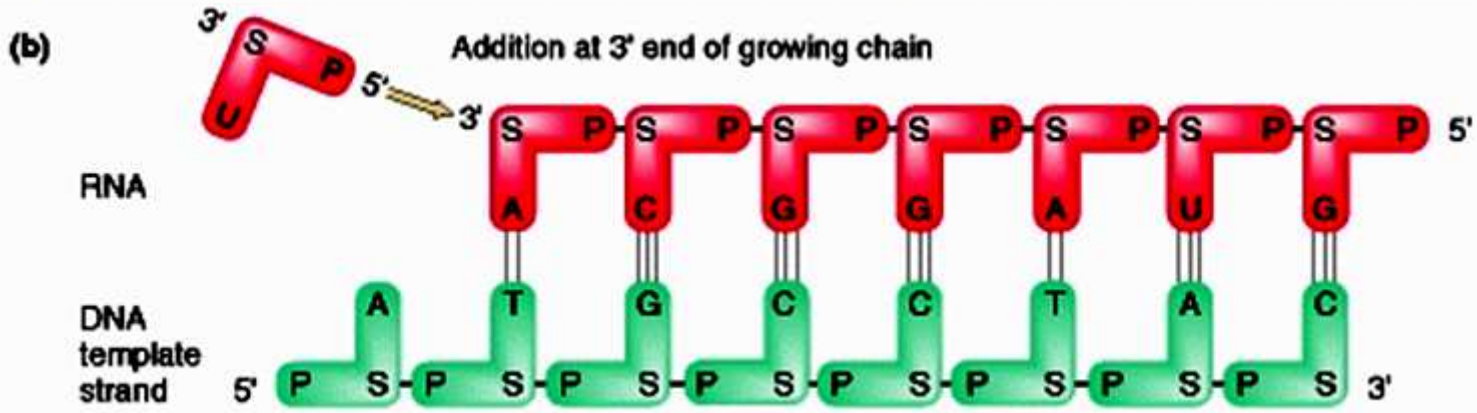


Figure shows a general promoter and where the RNA Polymerase sigma factor binds.

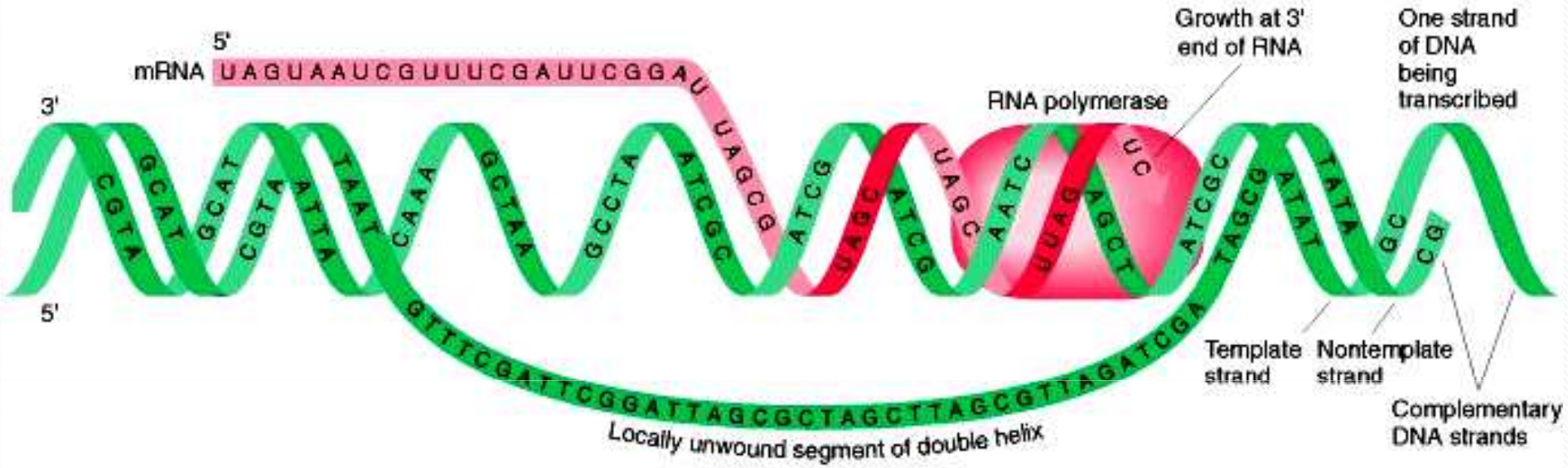

Polymerase unwinds
DNA, forming an
open complex
(c)
Core enzyme
Transcription begins


RNA polymerase
DNA
-35

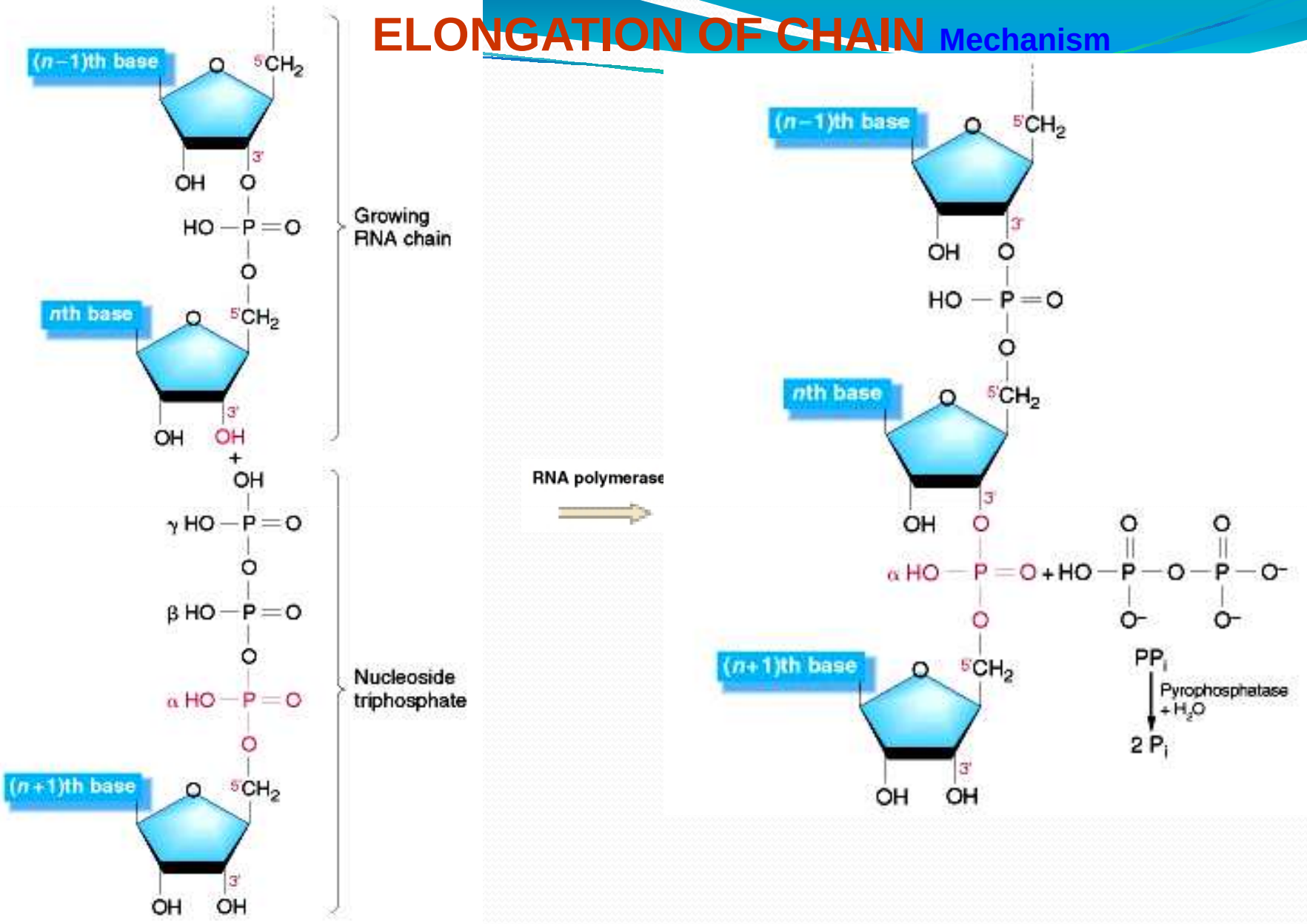

(a)
Nontemplate strand of gene 1
RNA
Template strand of gene 2
3'
5'
DNA
5'
3'
5'
RNA polymerase
Template strand of gene 1
RNA
Nontemplate strand of gene 2
RNA polymerase
Gene 1
Gene 2


(b)
Addition at 3' end of growing chain
3'
5'
RNA
DNA template strand
5'
3'

Synthesis continues



ELONGATION OF CHAIN Mechanism



TERMINATION OF SYNTHESIS

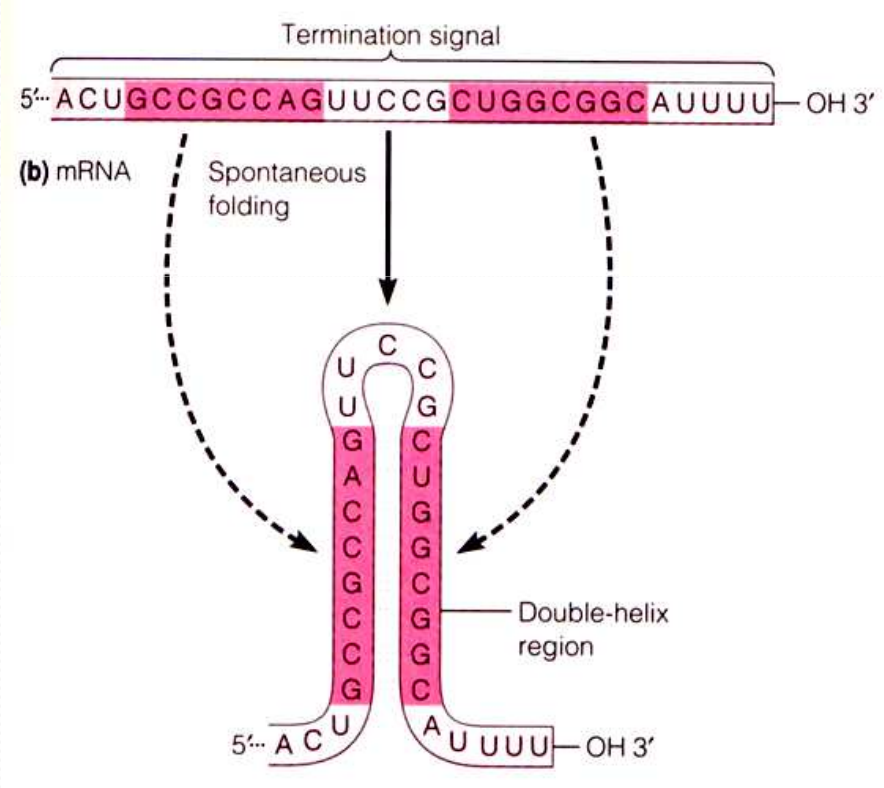
Termination of RNA synthesis occurs at specific base sequences within the DNA molecule. These sequences are of two types,

1. Simple terminators & 2. Termination factors dependent.

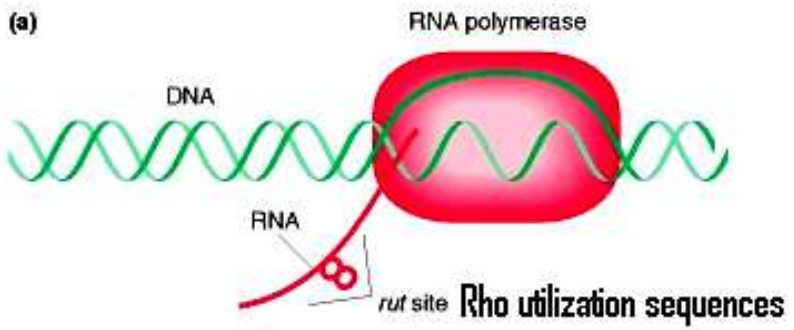
1. Simple Termination

Features are

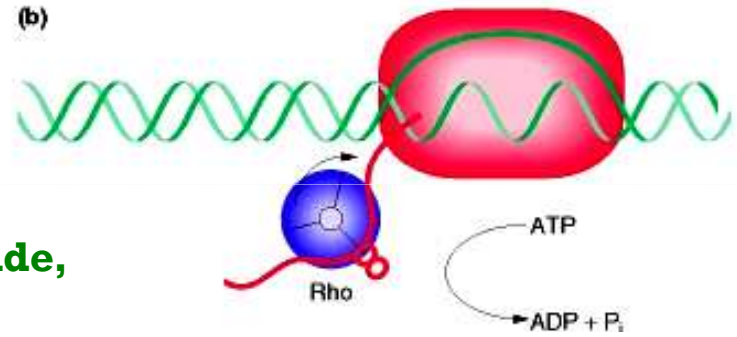
1. Sequence in one DNA strand would read like ABCDE-XYZ-E'D'C'B'A'. Thus, this sequence is capable of intrastrand base pairing, forming a stem-and-loop in the RNA transcript
2. Near the loop end there is a high G + C sequence.
3. This is followed by a sequence of 6-8 U's.



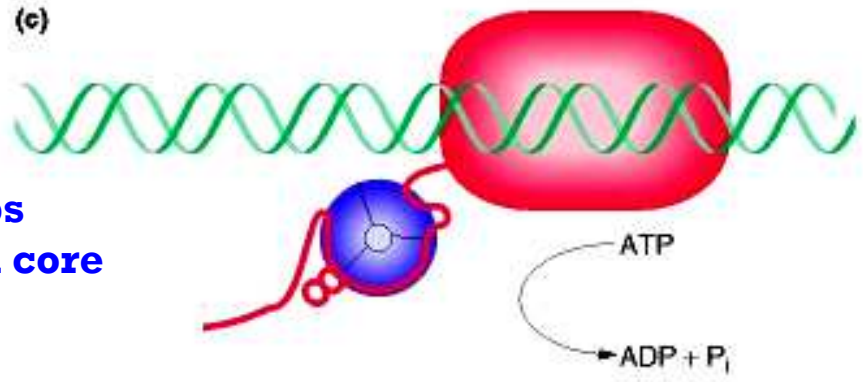
2. Rho dependant Termination



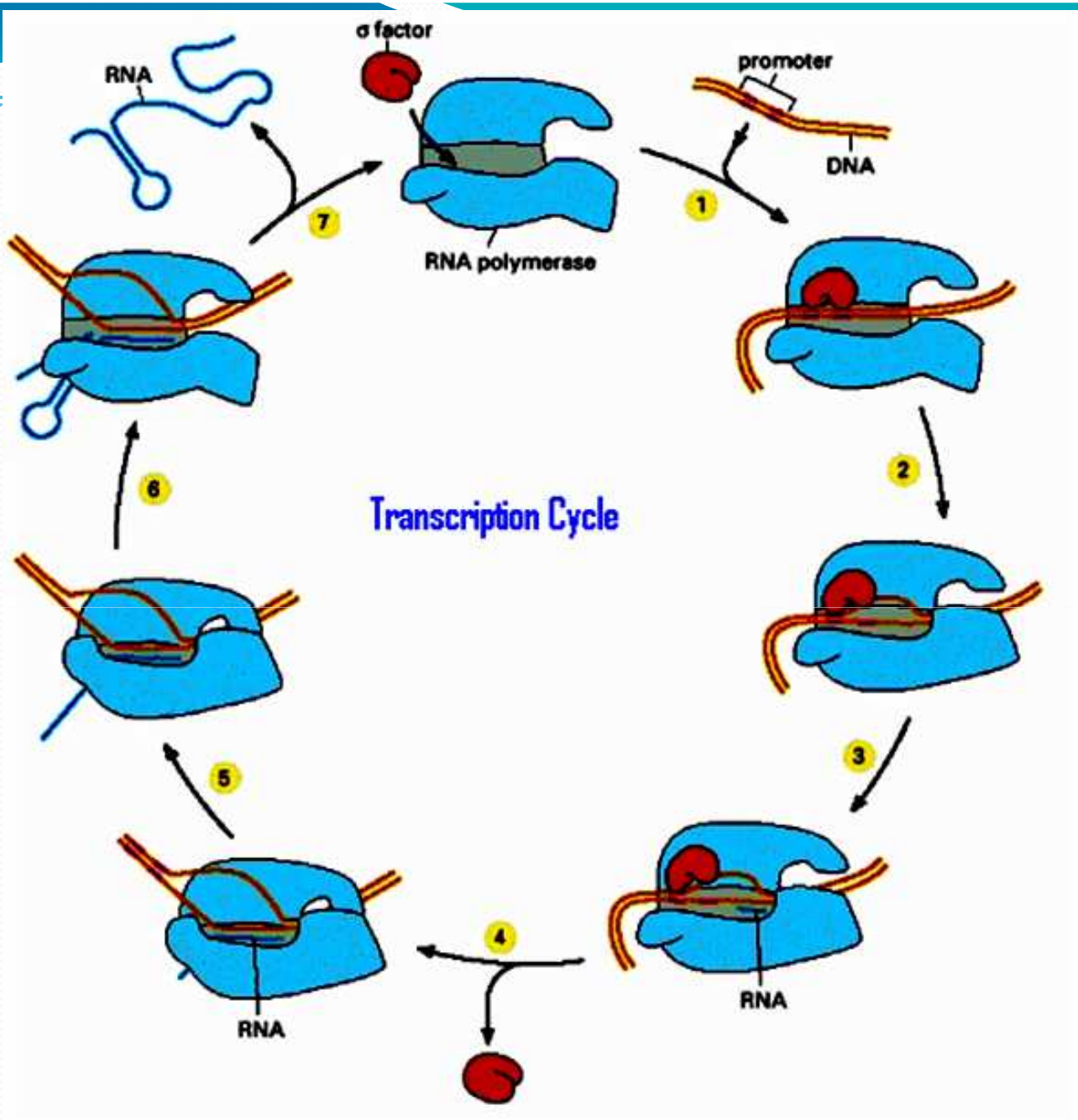
As polymerization proceeds and rut sites are exposed Rho factor recognizes and attaches to it



As polymerization proceeds and C-rich segments are made, the ATPase activity of Rho increases until no further polymerization is possible,



As no polymerization, RNA polymerase stops its action. However, since no sigma factor in core enzyme it can not restart the process Hence separation of all units.


σ factor
RNA
promoter
DNA
RNA polymerase
1
2
3
4
5
6
7
Transcription Cycle
RNA
RNA

Animation on Transcription

Acknowledgements
to

INTERNET

FOR

PICTURES