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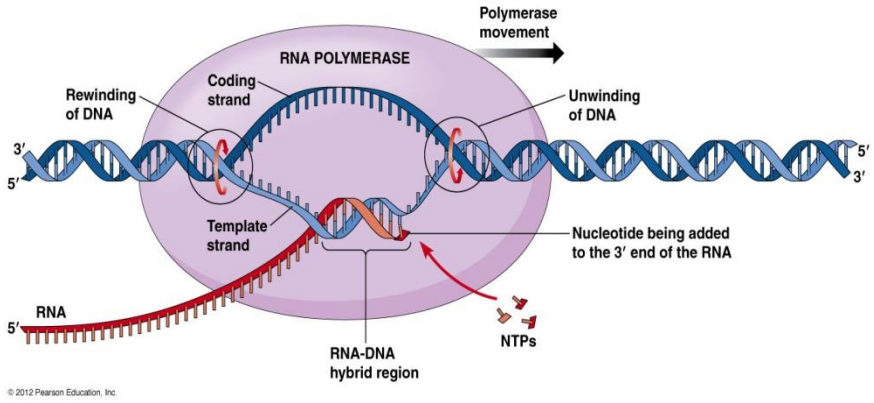
UNIT-V :GENETIC MECHANISMS

Transcription in prokaryotes : Initiation-Recognition,Binding,Elongation, and Termination.

Transcription in Prokaryotes

- The process of synthesis of RNA by copying the template strand of DNA is called transcription.
- During replication entire genome is copied but in transcription only the selected portion of genome is copied.
- The enzyme involved in transcription is RNA polymerase. Unlike DNA polymerase it can initiate transcription by itself, it does not require primase. More exactly it is a DNA dependent RNA polymerase.



The steps of transcription

transcription is an enzymatic process. the mechanism of transcription completes in three major steps

1. Initiation:

- closed complex formation
- Open complex fromation
- Tertiary complex formation

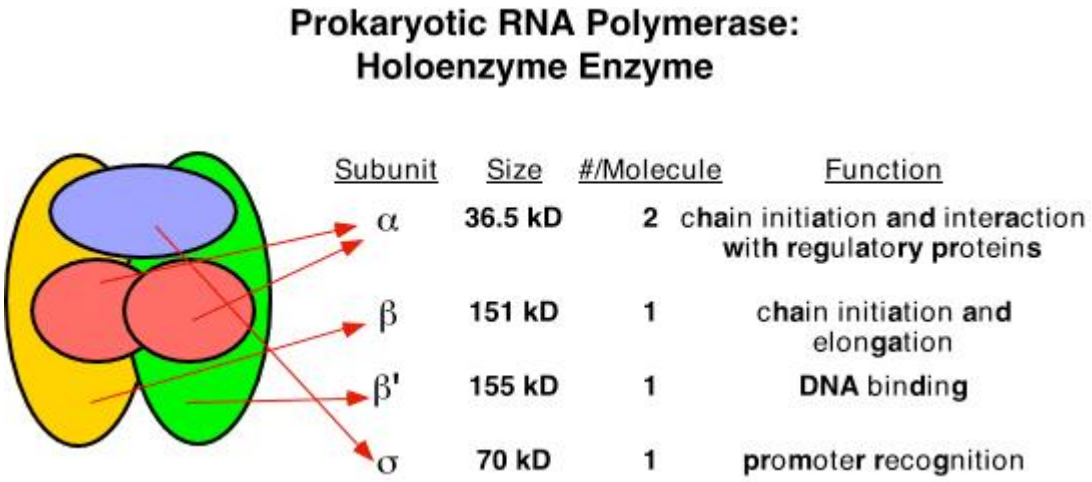
2. Elongation

3. Termination:

- **Rho- dependent**
- **Rho-indepdent**

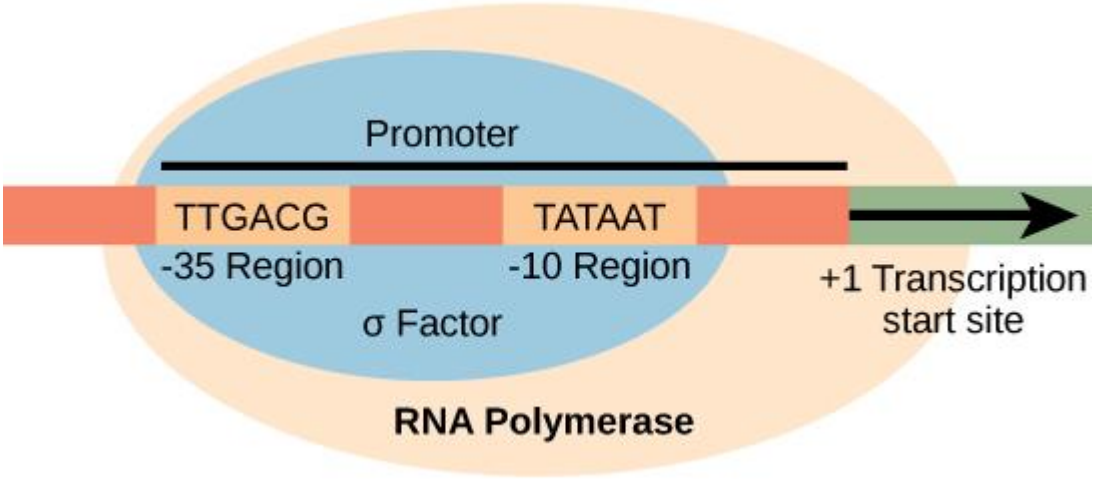
1. Initiation:

- **The transcription is initiated by RNA polymerase holoenzyme from a specific point called promotor sequence.**
- **Bacterial RNA polymerase is the principle enzyme involved in transcription.**
- **Single RNA polymerase is found in a bacteria which is called core polymerase and it consists of α, β, β' and ω sub units.**
- **The core enzyme bind to specific sequence on template DNA strand called promotor. The binding of core polymerase to promotor is facilitates and specified by sigma (σ) factor. (σ70 in case of E. coli).**



- The core polymerase along with σ-factor is called Holo-enzyme ie. RNA polymerase holoenzyme.
- In case of e. coli, promotor consists of two conserved sequences 5'-TTGACA-3' at -35 element and 5'-TATAAT-3' at -10 element. These sequence are upstream to the site from which transcription begins. Binding of holoenzyme to two conserve sequence of promotor form close complex.

- In some bacteria, the altered promotor may exist which contain UP-element and some may contain extended -10 element rather than -35 element.



σ70 of E. coli has four region.

1. Region1: it includes 1.2 and 1.1 region. Region 1.1 acts as molecular mimic of DNA
2. Region2: it recognizes -10 element in promotor. α-helix recognizes -10 element.
3. Region 3: it recognizes extended -10 element.
4. Region 4: it recognizes -35 element in promotor by a structure called helix-turn-helix.

The UP-element is recognized by a carboxyl terminal domain of α-sub unit called αCTD (carboxyl terminal domain) which is connected to αNTD (Amino terminal domain) by flexible linker.

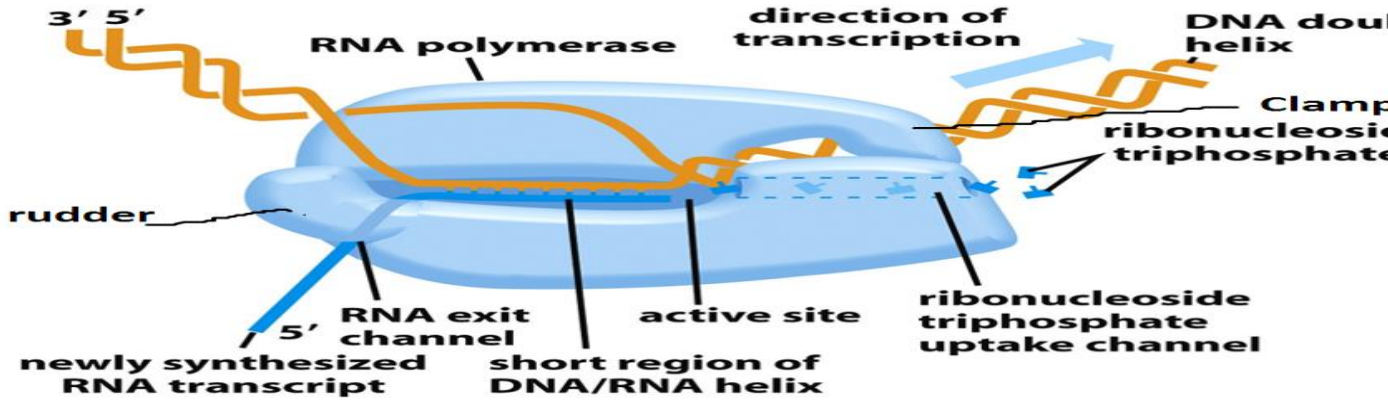
i. closed complex:

- Binding of RNA polymerase holoenzyme to the promotor sequence form closed comolex

ii. Open complex:

- after formation of closed complex, the RNA polymerase holoenzyme separates 10-14 bases exztending from -11 to +3 called melting. So that open complex is formed. This changing from closed complex to open complex is called **isomerization.**

iii. Tertiary complex:



- RNA polymerase starts synthesizing nucleotide. It doesnot require the help of primase.
- If the enzyme synthesize short RNA molecules of less than 10 bp, it does not further elongates which is called abortive initiation. This is because σ 3.2 acting as mimic of RNA and it lies at middle of RNA exit channel in open complex.
- When the RNA polymerase manage to synthesize RNA more than 10 bp long, it eject the σ 3.2 region and RNA further elongates and exit from RNA exit channel. This is the formation of tertiary complex.

2. Elongation:

- After synthesis of RNA more than 10 bp long, the σ-factor is ejected and the enzyme move along 5'-3' direction continuously synthesizing RNA.
- The synthesized RNA exit from RNA exit channel.
- The synthesized RNA is proof reads by Hydrolytic editing. For this the polymerase back track by one or more nucleotide and cleave the RNA removing the error and synthesize the correct one. The Gre factor enhance this proof reading process.
- Pyrophospholytic editing another mechanism of removing altered nucleotide.

3. Termination:

There are two mechanism of termination.

i. Rho independent:

- In this mechanism, transcription is terminated due to specific sequence in terminator DNA.
- The terminator DNA contains invert repeat which cause complimentary pairing as transcript RNA form hair pin structure.
- This invert repeat is followed by larger number of TTTTTTTT(~8 bp) on template DNA. The uracil appear in RNA. The load of hair pin structure is not tolerated by A=U base pair so the RNA get separated from RNA-DNA heteroduplex.

ii. Rho dependent:

- In this mechanism, transcription is terminated by rho (ρ) protein.
- It is ring shaped single strand binding ATpase protein.
- The rho protein bind the single stranded RNA as it exit from polymerase enzyme complex and hydrolyse the RNA from enzyme complex.
- The rho protein does not bind to those RNA whose protein is being translated. Rather it bind to RNA after translation.
- In bacteria transcription and translation occur simultaneously so the rho protein bind the RNA after translation has completed but transcription is still ON

Transcription in Eukaryotes –promoters Initiation With Polymerase |, ||, ||| Elongation And Termination.

Eukaryotic Transcription:

- Transcription is the process by which the information in a strand of DNA is copied into a new molecule of **RNA**.
- It is the first step of gene expression, in which a particular segment of DNA is copied into RNA (especially mRNA) by the enzyme RNA polymerase.
- It results in a complementary, antiparallel RNA strand called a primary transcript.

Transcription occurs in eukaryotes in a way that is similar to prokaryotes with reference to the basic steps involved. However, some major differences between them include:

- Initiation is more complex.
- Termination does not involve stem-loop structures.
- Transcription is carried out by three enzymes (RNA polymerases I, II and III).
- The regulation of transcription is more extensive than prokaryotes.

Enzyme(s) Involved in Eukaryotic Transcription

Unlike prokaryotes where all RNA is synthesized by a single RNA polymerase, the nucleus of a eukaryotic cell has three RNA polymerases responsible for transcribing different types of RNA.

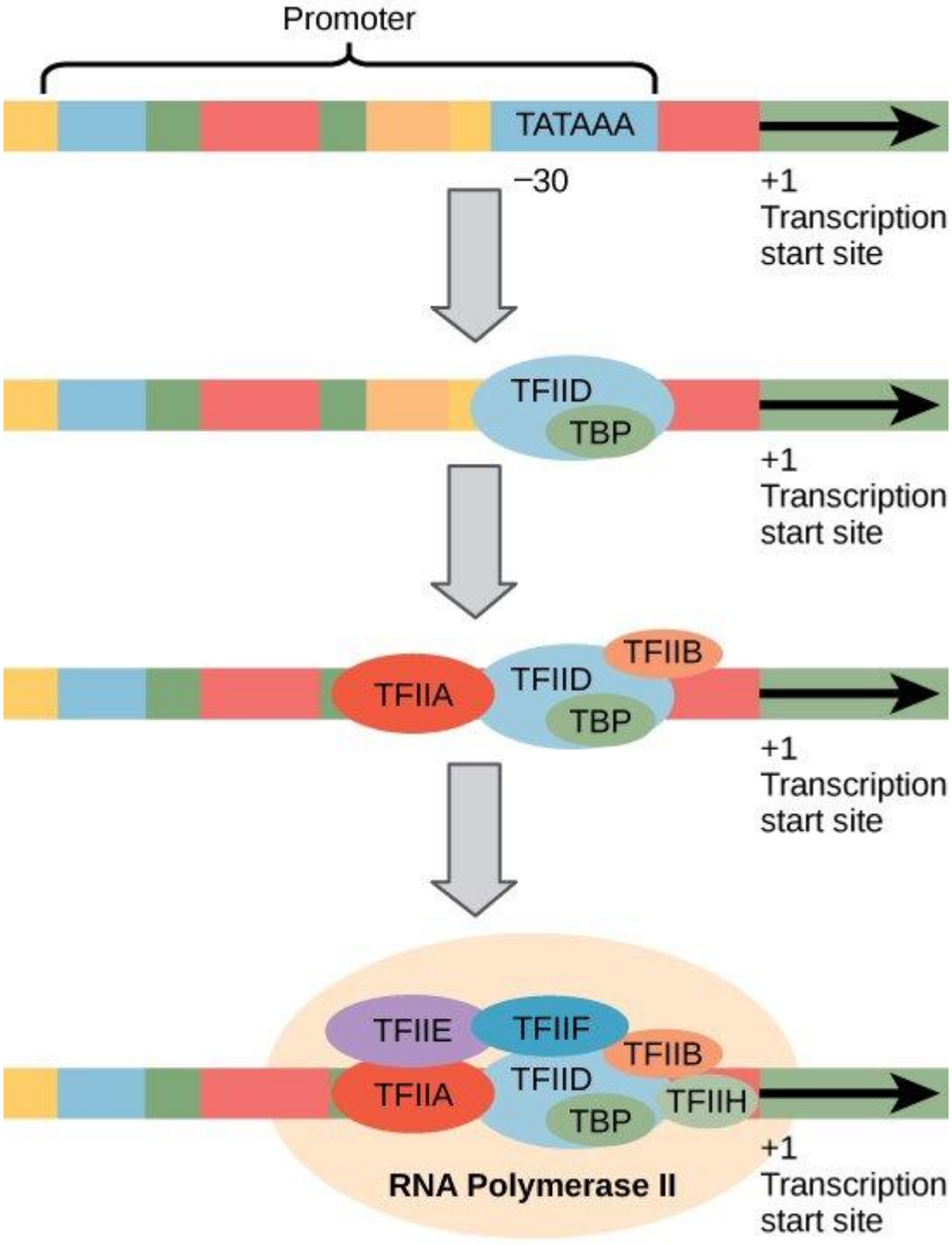
- **RNA polymerase I (RNA Pol I)** is located in the nucleolus and transcribes the 28S, 18S, and 5.8S rRNA genes.
- **RNA polymerase II (RNA Pol II)** is located in the nucleoplasm and transcribes protein-coding genes, to yield pre-mRNA, and also the genes encoding small nucleolar RNAs (snoRNAs) involved in rRNA processing and small nuclear RNAs (snRNAs) involved in mRNA processing, except for U6 snRNA.

- **RNA polymerase III (RNA Pol III)** is also located in the nucleoplasm. It transcribes the genes for tRNA, 5S rRNA, U6 snRNA, and the 7S RNA associated with the signal recognition particle (SRP) involved in the translocation of proteins across the endoplasmic reticulum membrane.
- Each of the three eukaryotic RNA polymerases contains 12 or more subunits and so these are large complex enzymes.
- The genes encoding some of the subunits of each eukaryotic enzyme show DNA sequence similarities to genes encoding subunits of the core enzyme of *E. coli* RNA polymerase.
- However, four to seven other subunits of each eukaryotic RNA polymerase are unique in that they show no similarity either with bacterial RNA polymerase subunits or with the subunits of other eukaryotic RNA polymerases.

Process of Eukaryotic Transcription:

The basic mechanism of RNA synthesis by these eukaryotic RNA polymerases can be divided into the following phases:

Initiation Phase



- During initiation, RNA polymerase recognizes a specific site on the DNA, upstream from the gene that will be transcribed, called a **promoter site** and then unwinds the DNA locally.
- Most promoter sites for RNA polymerase II include a highly conserved sequence located about 25–35 bp upstream (i.e. to the 5 side) of the start site which has the consensus TATA(A/T)A(A/T) and is called the TATA box.
- Since the start site is denoted as position +1, the TATA box position is said to be located at about position -25.
- The TATA box sequence resembles the -10 sequence in prokaryotes (TATAAT) except that it is located further upstream.

- Both elements have essentially the same function, namely recognition by the RNA polymerase in order to position the enzyme at the correct location to initiate transcription.
- The sequence around the TATA box is also important in that it influences the efficiency of initiation. Transcription is also regulated by upstream control elements that lie 5′ to the TATA box.
- Some eukaryotic protein-coding genes lack a TATA box and have an initiator element instead, centered around the transcriptional initiation site.
- In order to initiate transcription, RNA polymerase II requires the assistance of several other proteins or protein complexes, called general (or basal) transcription factors, which must assemble into a complex on the promoter in order for RNA polymerase to bind and start transcription.
- These all have the generic name of TFII (for Transcription Factor for RNA polymerase II).
- The first event in initiation is the binding of the transcription factor IID (TFIID) protein complex to the TATA box via one its subunits called TBP (TATA box binding protein).
- As soon as the TFIID complex has bound, TFIIA binds and stabilizes the TFIID-TATA box interaction. Next, TFIIB binds to TFIID.
- However, TFIIB can also bind to RNA polymerase II and so acts as a bridging protein. Thus,
- RNA polymerase II, which has already complexed with TFIIF, now binds.
- This is followed by the binding of TFIIE and H. This final protein complex contains at least 40 polypeptides and is called the **transcription initiation complex.**
- Those protein-coding genes that have an initiator element instead of a TATA box appear to need another protein(s) that binds to the initiator element.
- The other transcription factors then bind to form the transcription initiation complex in a similar manner to that described above for genes possessing a TATA box promoter.

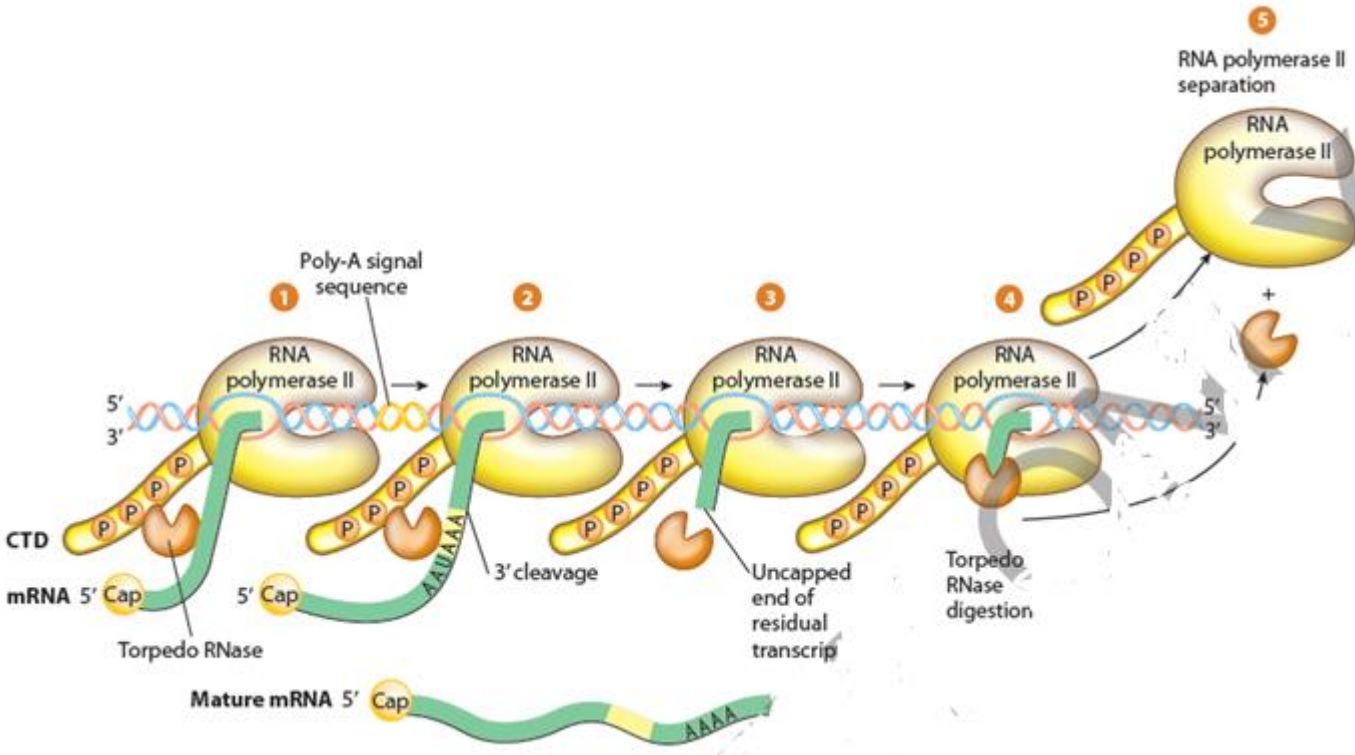
Elongation Phase

TFIIH has two functions:

1. It is a helicase, which means that it can use ATP to unwind the DNA helix, allowing transcription to begin.
2. In addition, it phosphorylates RNA polymerase II which causes this enzyme to change its conformation and dissociate from other proteins in the initiation complex.

- The key phosphorylation occurs on a long C-terminal tail called the C-terminal domain (CTD) of the RNA polymerase II molecule.
- Interestingly, only RNA polymerase II that has a non-phosphorylated CTD can initiate transcription but only an RNA polymerase II with a phosphorylated CTD can elongate RNA.
- RNA polymerase II now starts moving along the DNA template, synthesizing RNA, that is, the process enters the elongation phase.
- RNA synthesis occurs in the 5′ → 3′ direction with the RNA polymerase catalyzing a nucleophilic attack by the 3-OH of the growing RNA chain on the alpha-phosphorus atom on an incoming ribonucleoside 5-triphosphate.
- The RNA molecule made from a protein-coding gene by RNA polymerase II is called a primary transcript.

Termination Phase



- Elongation of the RNA chain continues until termination occurs.
- Unlike RNA polymerase in prokaryotes, RNA polymerase II does not terminate transcription at a specific site but rather transcription can stop at varying distances downstream of the gene.
- RNA genes transcribed by RNA Polymerse II lack any specific signals or sequences that direct RNA Polymerase II to terminate at specific locations.
- RNA Polymerase II can continue to transcribe RNA anywhere from a few bp to thousands of bp past the actual end of the gene.
- The transcript is cleaved at an internal site before RNA Polymerase II finishes transcribing. This releases the upstream portion of the transcript, which will serve as the initial RNA prior to further processing (the pre-mRNA in the case of protein-encoding genes.)
- This cleavage site is considered the "end" of the gene. The remainder of the transcript is digested by a 5′-exonuclease (called Xrn2 in humans) while it is still being transcribed by the RNA Polymerase II.
- When the 5′-exonulease "catches up" to RNA Polymerase II by digesting away all the overhanging RNA, it helps disengage the polymerase from its DNA template strand, finally terminating that round of transcription.

Post transcriptional modification in mRNA Mechanism-Capping,Splicing.

Post-transcriptional modification or **co-transcriptional modification** is a set of biological processes common to most eukaryotic cells by which an RNA primary transcript is chemically altered following transcription from a gene to produce a mature, functional RNA molecule that can then leave the nucleus and perform any of a variety of different functions in the cell. There

are many types of post-transcriptional modifications achieved through a diverse class of molecular mechanisms.

One example is the conversion of precursor messenger RNA transcripts into mature messenger RNA that is subsequently capable of being translated into protein. This process includes three major steps that significantly modify the chemical structure of the RNA molecule: the addition of a 5' cap, the addition of a 3' polyadenylated tail, and RNA splicing. Such processing is vital for the correct translation of eukaryotic genomes because the initial precursor mRNA produced by transcription often contains both exons (coding sequences) and introns (non-coding sequences); splicing removes the introns and links the exons directly, while the cap and tail facilitate the transport of the mRNA to a ribosome and protect it from molecular degradation.

Post-transcriptional modifications may also occur during the processing of other transcripts which ultimately become transfer RNA, ribosomal RNA, or any of the other types of RNA used by the cell.

Capping:

Capping of the pre-mRNA involves the addition of 7-methylguanosine (m^7G) to the 5' end. To achieve this, the terminal 5' phosphate requires removal, which is done with the aid of a phosphatase enzyme. The enzyme guanosyl transferase then catalyses the reaction, which produces the diphosphate 5' end. The diphosphate 5' end then attacks the alpha phosphorus atom of a GTP molecule in order to add the guanine residue in a 5'5' triphosphate link. The enzyme (guanine-N^7-)-methyltransferase ("cap MTase") transfers a methyl group from S-adenosyl methionine to the guanine ring. This type of cap, with just the (m^7G) in position is called a cap 0 structure. The ribose of the adjacent nucleotide may also be methylated to give a cap 1. Methylation of nucleotides downstream of the RNA molecule produce cap 2, cap 3 structures and so on. In these cases the methyl groups are added to the 2' OH groups of the ribose sugar. The cap protects the 5' end of the primary RNA transcript from attack by ribonucleases that have specificity to the 3'5' phosphodiester bonds.

Splicing

RNA splicing is the process by which introns, regions of RNA that do not code for proteins, are removed from the pre-mRNA and the remaining exons connected to re-form a single continuous molecule. Exons are sections of mRNA which become "expressed" or translated into a protein. They are the coding portions of a mRNA molecule. Although most RNA splicing occurs after the complete synthesis and end-capping of the pre-mRNA, transcripts with many exons can be spliced co-transcriptionally.The splicing reaction is catalyzed by a large protein complex called the spliceosome assembled from proteins and small nuclear RNA molecules that recognize splice sites in the pre-mRNA sequence. Many pre-mRNAs, including those encoding antibodies, can be spliced in multiple ways to produce different mature mRNAs that encode different protein sequences. This process is known as alternative splicing, and allows production of a large variety of proteins from a limited amount of DNA.

TRANSLATION:

translation is the process in which ribosomes in the cytoplasm or endoplasmic reticulum synthesize proteins after the process of transcription of DNA to RNA in the cell's nucleus. The entire process is called gene expression.

In translation, messenger RNA (mRNA) is decoded in a ribosome, outside the nucleus, to produce a specific amino acid chain, or polypeptide. The polypeptide later folds into an active protein and performs its functions in the cell. The ribosome facilitates decoding by inducing the binding of complementary tRNA anticodon sequences to mRNA codons. The tRNAs carry specific amino acids that are chained together into a polypeptide as the mRNA passes through and is "read" by the ribosome.

THREE STEPS OF TRANSLATION:

Initiation: sets the stage for polypeptide synthesis.

Elongation: causes the sequential addition of amino acids to the polypeptide chain in a colinear fashion as determined by the sequence of mRNA.

Termination: Brings the polypeptide synthesis to a halt.

Protein Folding-Chaperone-Modiated Abnormal Folding-CJD disease.

PROTEIN FOLDING:

Protein folding is the physical process by which a protein chain is translated to its native three-dimensional structure, typically a "folded" conformation by which the protein becomes biologically functional. Via an expeditious and reproducible process, a polypeptide folds into its characteristic three-dimensional structure from a random coil. Each protein exists first as an unfolded polypeptide or random coil after being translated from a sequence of mRNA to a linear chain of amino acids. At this stage the polypeptide lacks any stable (long-lasting) three-dimensional structure (the left hand side of the first figure). As the polypeptide chain is being synthesized by a ribosome, the linear chain begins to fold into its three-dimensional structure.

Folding of many proteins begins even during translation of the polypeptide chain. Amino acids interact with each other to produce a well-defined three-dimensional structure, the folded protein (the right hand side of the figure), known as the native state. The resulting three-dimensional structure is determined by the amino acid sequence or primary structure (Anfinsen's dogma).

The correct three-dimensional structure is essential to function, although some parts of functional proteins may remain unfolded, so that protein dynamics is important. Failure to fold into native structure generally produces inactive proteins, but in some instances misfolded proteins have modified or toxic functionality. Several neurodegenerative and other diseases are believed to result from the accumulation of amyloid fibrils formed by misfolded proteins. Many allergies are caused by incorrect folding of some proteins, because the immune system does not produce antibodies for certain protein structures.

Denaturation of proteins is a process of transition from the folded to the unfolded state. It happens in cooking, in burns, in proteinopathies, and in other contexts.

The duration of the folding process varies dramatically depending on the protein of interest. When studied outside the cell, the slowest folding proteins require many minutes or hours to fold primarily due to proline isomerization, and must pass through a number of intermediate states, like checkpoints, before the process is complete. On the other hand, very small single-domain proteins with lengths of up to a hundred amino acids typically fold in a single step. Time scales of milliseconds are the norm and the very fastest known protein folding reactions are complete within a few microseconds.

Chaperones:

Molecular chaperones are a class of proteins that aid in the correct folding of other proteins *in vivo*. Chaperones exist in all cellular compartments and interact with the polypeptide chain in order to allow the native three-dimensional conformation of the protein to form; however, chaperones themselves are not included in the final structure of the protein they are assisting in. Chaperones may assist in folding even when the nascent polypeptide is being synthesized by the ribosome. Molecular chaperones operate by binding to stabilize an otherwise unstable structure of a protein in its folding pathway, but chaperones do not contain the necessary information to know the correct native structure of the protein they are aiding; rather, chaperones work by preventing incorrect folding conformations.In this way, chaperones do not actually increase the rate of individual steps involved in the folding pathway toward the native structure; instead, they work by reducing possible unwanted aggregations of the polypeptide chain that might otherwise slow down the search for the proper intermediate and they provide a more efficient pathway for the polypeptide chain to assume the correct conformations.Chaperones are not to be confused with folding catalyst proteins, which catalyze chemical reactions responsible for slow steps in folding pathways. Examples of folding catalysts are protein disulfide isomerases and peptidyl-prolyl isomerases that may be involved in formation of disulfide bonds or interconversion between cis and trans stereoisomers of peptide group.Chaperones are shown to be critical in the process of protein folding *in vivo* because they provide the protein with the aid needed to assume its proper alignments and conformations efficiently enough to become "biologically relevant". This means that the polypeptide chain could theoretically fold into its native structure without the aid of chaperones, as demonstrated by protein folding experiments conducted *in vitro*; however, this process proves to be too inefficient or too slow to exist in biological systems; therefore, chaperones are necessary for protein folding *in vivo.* Along with its role in aiding native structure formation, chaperones are shown to be involved in various roles such as protein transport, degradation, and even allow denatured proteins exposed to certain external denaturant factors an opportunity to refold into their correct native structures.

A fully denatured protein lacks both tertiary and secondary structure, and exists as a so-called random coil. Under certain conditions some proteins can refold; however, in many cases, denaturation is irreversible. Cells sometimes protect their proteins against the denaturing influence of heat with enzymes known as heat shock proteins (a type of chaperone), which assist other proteins both in folding and in remaining folded. Heat shock proteins have been found in all species examined, from bacteria to humans, suggesting that they evolved very early and have an important function. Some proteins never fold in cells at all except with the assistance of chaperones which either isolate individual proteins so that their folding is not interrupted by interactions with other proteins or help to unfold misfolded proteins, allowing them to refold into the correct native structure. This function is crucial to prevent the risk of precipitation into insoluble amorphous aggregates. The external factors involved in protein denaturation or disruption of the native state include temperature, external fields (electric, magnetic), molecular crowding, and even the limitation of space (i.e. confinement), which can have a big influence on the folding of proteins. High concentrations of solutes, extremes of pH, mechanical forces, and the presence of chemical denaturants can contribute to protein denaturation, as well. These individual factors are categorized together as stresses. Chaperones are shown to exist in increasing concentrations during times of cellular stress and help the proper folding of emerging proteins as well as denatured or misfolded ones.

Under some conditions proteins will not fold into their biochemically functional forms. Temperatures above or below the range that cells tend to live in will cause thermally unstable proteins to unfold or denature (this is why boiling makes an egg white turn opaque). Protein thermal stability is far from constant, however; for example, hyperthermophilic bacteria have been found that grow at temperatures as high as 122 °C. which of course requires that their full complement of vital proteins and protein assemblies be stable at that temperature or above.

The bacterium *E. coli* is the host for bacteriophage T4, and the phage encoded gp31 protein (P17313) appears to be structurally and functionally homologous to *E. coli* chaperone

protein GroES and able to substitute for it in the assembly of bacteriophage T4 virus particles during infection. Like GroES, gp31 forms a stable complex with GroEL chaperonin that is absolutely necessary for the folding and assembly in vivo of the bacteriophage T4 major capsid protein gp23.

CJD diseases:

Creutzfeldt–Jakob disease (**CJD**), also known as **subacute spongiform encephalopathy** or **neurocognitive disorder due to prion disease**, is a fatal degenerative brain disorder. Early symptoms include memory problems, behavioral changes, poor coordination, and visual disturbances. Later symptoms include dementia, involuntary movements, blindness, weakness, and coma.[1] About 70% of people die within a year of diagnosis.

CJD is caused by a protein known as a prion. Infectious prions are misfolded proteins that can cause normally folded proteins to become misfolded. About 85% of cases occur for unknown reasons, while about 7.5% of cases are inherited from a person's parents in an autosomal dominant manner. Exposure to brain or spinal tissue from an infected person may also result in spread. There is no evidence that it can spread between people via normal contact or blood transfusions. Diagnosis involves ruling out other potential causes. An electroencephalogram, spinal tap, or magnetic resonance imaging may support the diagnosis.

There is no specific treatment for CJD. Opioids may be used to help with pain, while clonazepam or sodium valproate may help with involuntary movements. CJD affects about one per million people per year. Onset is typically around 60 years of age. The condition was first described in 1920. It is classified as a type of transmissible spongiform encephalopathy. Inherited CJD accounts for about 10% of prion disease cases. Sporadic CJD is different from bovine spongiform encephalopathy (mad cow disease) and variant Creutzfeldt–Jakob disease (vCJD).

Regulation Of Gene Expression In Prokaryotes & Eukaryotes:

Regulation Of Gene Expression In Prokaryotes:

Regulation of gene expression, or **gene regulation**, includes a wide range of mechanisms that are used by cells to increase or decrease the production of specific gene products (protein or RNA). Sophisticated programs of gene expression are widely observed in biology, for example to trigger developmental pathways, respond to environmental stimuli, or adapt to new food sources. Virtually any step of gene expression can be modulated, from transcriptional initiation, to RNA processing, and to the post-translational modification of a protein. Often, one gene regulator controls another, and so on, in a gene regulatory network.

Gene regulation is essential for viruses, prokaryotes and eukaryotes as it increases the versatility and adaptability of an organism by allowing the cell to express protein when needed. Although as early as 1951, Barbara McClintock showed interaction between two genetic loci, Activator (*Ac*) and Dissociator (*Ds*), in the color formation of maize seeds, the first discovery of a gene regulation system is widely considered to be the identification in 1961 of the *lac* operon, discovered by François Jacob and Jacques Monod, in which some enzymes involved in lactose metabolism are expressed by *E. coli* only in the presence of lactose and absence of glucose.

In multicellular organisms, gene regulation drives cellular differentiation and morphogenesis in the embryo, leading to the creation of different cell types that possess different gene expression profiles from the same genome sequence. Although this does not explain how gene regulation

originated, evolutionary biologists include it as a partial explanation of how evolution works at a molecular level, and it is central to the science of evolutionary developmental biology

Gene Expression in Prokaryotes:

Prokaryotes only transcribe genes that their end-proteins are needed at the time. They do this inorder to save up energy and increase efficiency. The regulation of gene expression is depended mainly on their immediate environment, for example on the presence and absence of nutrients. Gene expression in prokaryotes occurs primarily at the level of trascription.

Gene Expression In Eukaryotes:

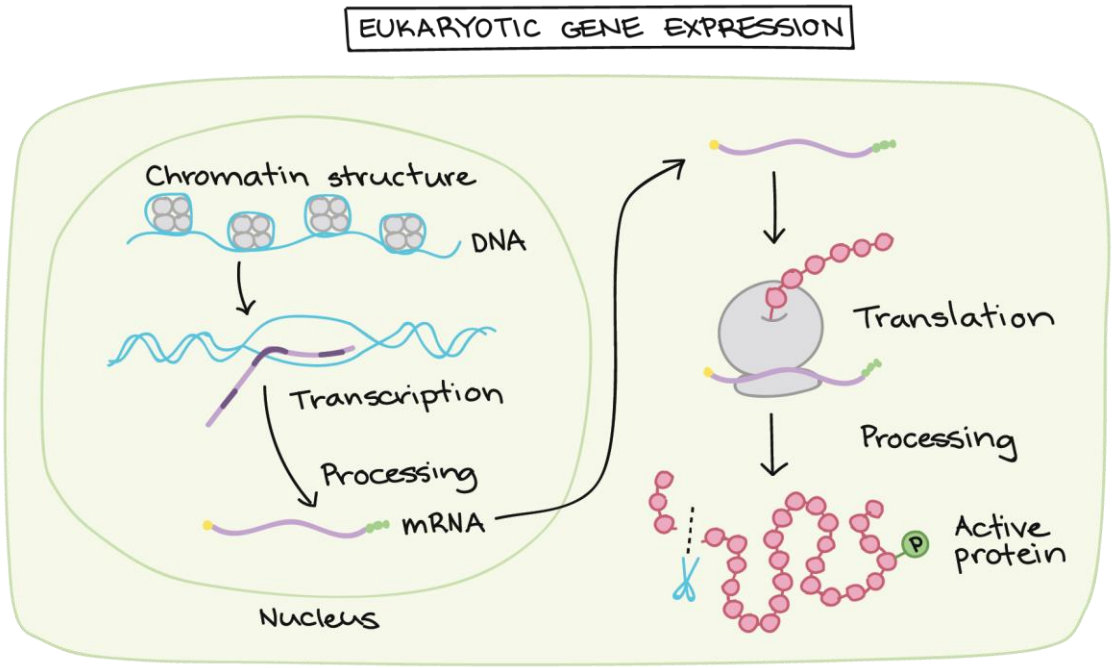
Eukaryotic gene expression can be regulated at many stages

In the articles that follow, we'll examine different forms of eukaryotic **gene regulation**. That is, we'll see how the expression of genes in eukaryotes (like us!) can be controlled at various stages, from the availability of DNA to the production of mRNAs to the translation and processing of proteins.

Eukaryotic gene expression involves many steps, and almost all of them can be regulated. Different genes are regulated at different points, and it's not uncommon for a gene (particularly an important or powerful one) to be regulated at multiple steps.

- **Chromatin accessibility**. The structure of chromatin (DNA and its organizing proteins) can be regulated. More open or "relaxed" chromatin makes a gene more available for transcription.
- **Transcription**. Transcription is a key regulatory point for many genes. Sets of **transcription factor** proteins bind to specific DNA sequences in or near a gene and promote or repress its transcription into an RNA.
- **RNA processing**. Splicing, capping, and addition of a poly-A tail to an RNA molecule can be regulated, and so can exit from the nucleus.

Different mRNAs may be made from the same pre-mRNA by **alternative splicing**.



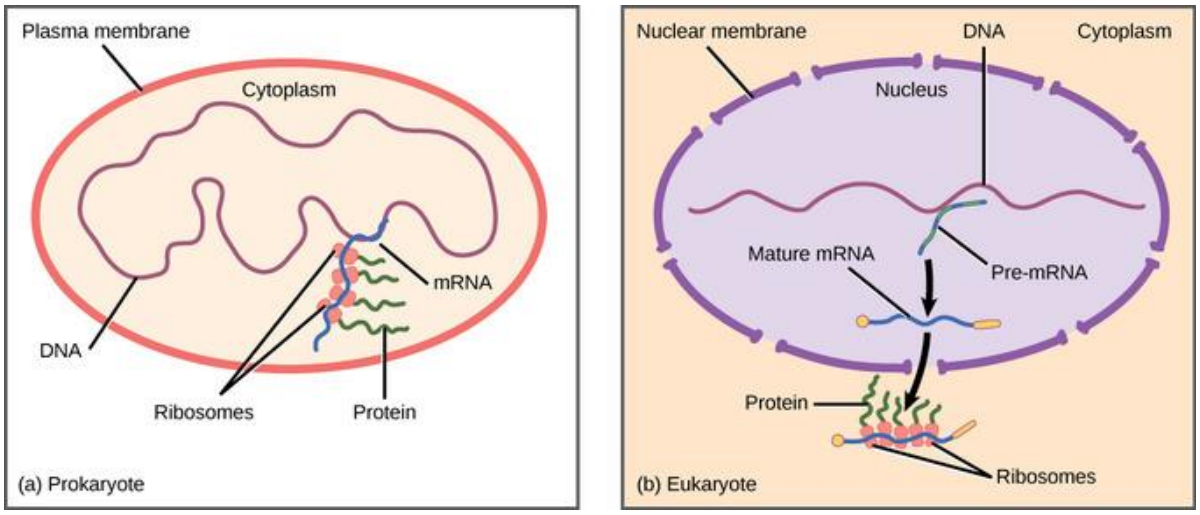
Prokaryotic versus Eukaryotic Gene Expression

To understand how gene expression is regulated, we must first understand how a gene codes for a functional protein in a cell. The process occurs in both prokaryotic and eukaryotic cells, just in slightly different manners.

Prokaryotic organisms are single-celled organisms that lack a defined nucleus; therefore, their DNA floats freely within the cell cytoplasm. To synthesize a protein, the processes of transcription (DNA to RNA) and translation (RNA to protein) occur almost simultaneously. When the resulting protein is no longer needed, transcription stops. Thus, the regulation of transcription is the primary method to control what type of protein and how much of each protein is expressed in a prokaryotic cell. All of the subsequent steps occur automatically. When more protein is required, more transcription occurs. Therefore, in prokaryotic cells, the control of gene expression is mostly at the transcriptional level.

Eukaryotic cells, in contrast, have intracellular organelles that add to their complexity. In eukaryotic cells, the DNA is contained inside the cell's nucleus where it is transcribed into RNA. The newly-synthesized RNA is then transported out of the nucleus into the cytoplasm where ribosomes translate the RNA into protein. The processes of transcription and translation are physically separated by the nuclear membrane; transcription occurs only within the nucleus, and translation occurs only

outside the nucleus within the cytoplasm. The regulation of gene expression can occur at all stages of the process. Regulation may occur when the DNA is uncoiled and loosened from nucleosomes to bind transcription factors (epigenetics), when the RNA is transcribed (transcriptional level), when the RNA is processed and exported to the cytoplasm after it is transcribed (post-transcriptional level), when the RNA is translated into protein (translational level), or after the protein has been made (post-translational level).



Prokaryotic vs Eukaryotic Gene Expression: Prokaryotic transcription and translation occur simultaneously in the cytoplasm, and regulation occurs at the transcriptional level. Eukaryotic gene expression is regulated during transcription and RNA processing, which take place in the nucleus, and during protein translation, which takes place in the cytoplasm. Further regulation may occur through post-translational modifications of proteins.