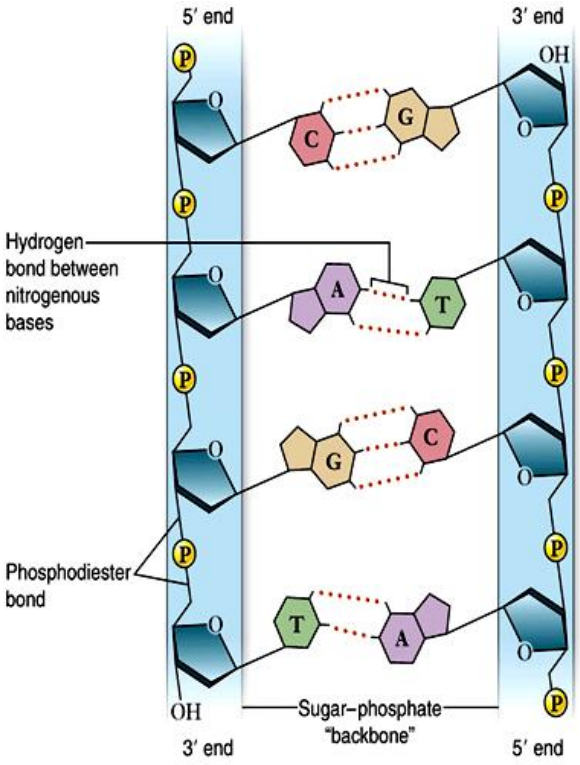
- **The central dogma (principle) of molecular biology:**
  - Information from DNA are transcribed to mRNA which will be further translated to synthesize a protein.
- **What is DNA and its characteristics?**
  - **Definition**: it is a double stranded, polymer of deoxyribonucleotides which is carrying genetic information and found in cell nucleus (notice that a small amount of DNA is also found in mitochondria).
  - **DNA is composed of nucleotides which are further constituted from:**
    - ✓ Nitrogenous base.
    - ✓ Sugar (deoxyribose).
    - ✓ Phosphate group.

      Notice: there are two types of nucleotides:
      - ❖ *Purines*: Adenine (A) and Guanine (G).
      - ❖ *Pyrimidines*: Cytosine (C) and Thymine (T).
  - The DNA is a double helix which was discovered by Watson and Crick in 1953. Before them, Franklin discovered the base-pairing property by x-ray (known as image 51).
  - **The double helix structure of DNA:**
    - ✓ Two sugar phosphate backbones (sugars are joined together by phosphodiester bonds).
    - ✓ Nucleotides are present in the middle and held together by hydrogen bonds:
      - ❖ *Three hydrogen bonds* between Guanine (G) and Cytosine (C).
      - ❖ *Two hydrogen bonds* between Adenine (A) and Thymine (T).
    - ✓ The two strands of DNA are aligned anti-parallel to each other (في اتجاه معاكس للآخر): one will be oriented from 5'→3' while the other strand will be oriented from 3'→5'



  - **The DNA double helix is rapped around proteins known as histones** → forming structures known as nucleosomes (nucleosomes are the main packaging structures of DNA in eukaryotic chromosomes). DNA has to be wrapped and packed because it is long and big while the nucleus is small. **Nucleosomes –together- will form chromatin:**
    - ✓ Euchromatin: genetically-active DNA.
    - ✓ Heterochromatin: genetically-inactive DNA.
- **What is RNA and its characteristics?**
  - **Definition**: it is single stranded and nucleotides are composed of:
    - ✓ Nitrogenous base.
    - ✓ Ribose sugar (instead of deoxyribose sugar in DNA).
    - ✓ Phosphate group.

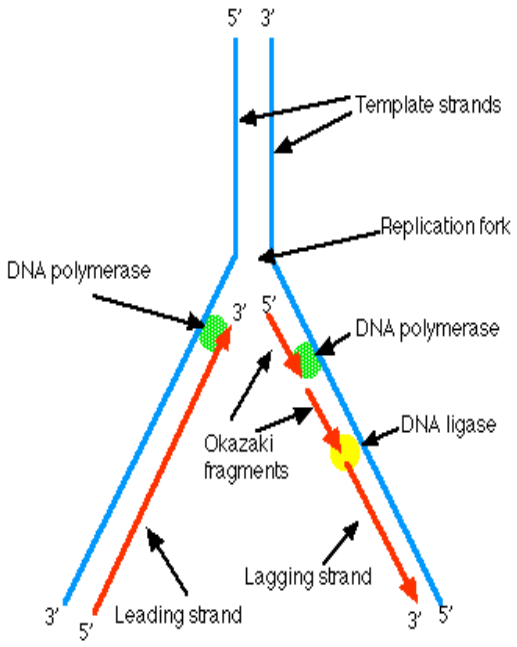
      Notice: nucleotides of RNA are:
      - ❖ Adenine (A).

- ❖ Guanine (G).
- ❖ Cytosine (C).
- ❖ Uracil (U) → instead of Thymine (T) which is found in DNA.

- **Secondary and tertiary structures of RNA:** RNA molecules can fold back on themselves to make complex secondary and tertiary structures which are essential to perform their biological role.
- **There are three major types of RNA:**
  - ✓ mRNA: which is the only one coding for proteins (other types of RNA are non-coding but they are important for other functions such as storage, regulation… etc).
  - ✓ tRNA: it binds amino acids and transfer them to mRNA-ribosome complex (for the process of translation).
  - ✓ ribosomal RNA: the site where protein is synthesized.

- **Protein:**
  - **Definition**: it is a chain-like polymers of a few or many thousands of amino acids. Each three nucleotides will make a codon for one amino acid.
  - **There are four levels of protein structure:**
    - ✓ Primary: sequence of a chain of amino acid.
    - ✓ Secondary: a chain of amino acids linked by hydrogen bonds.
    - ✓ Tertiary: it occurs when certain attraction occurs between α-helices and pleated sheets.
    - ✓ Quaternary: a protein containing more than one amino acid chain.

- **DNA replication:**
  - Starts at origin of replication through the action of the enzyme helicase which unwinds the two parental strands generating what is known as the Y-shaped replication fork (the unwinding process is stabilized by: single-strand binding proteins).
  - Each DNA strand directs the synthesis of a complementary identical daughter DNA strand (nucleotides are added by complementary base-pairing with the template parental strand).
  - Remember that DNA-polymerase adds nucleotides only in the 5'→3' direction.
  - **DNA-synthesis is semi-discontinuous which means:**
    - ✓ Leading strand is synthesized continuously.
    - ✓ Lagging strand is synthesized discontinuously as Okazaki fragments (which will be joined together by the enzyme DNA-ligase).



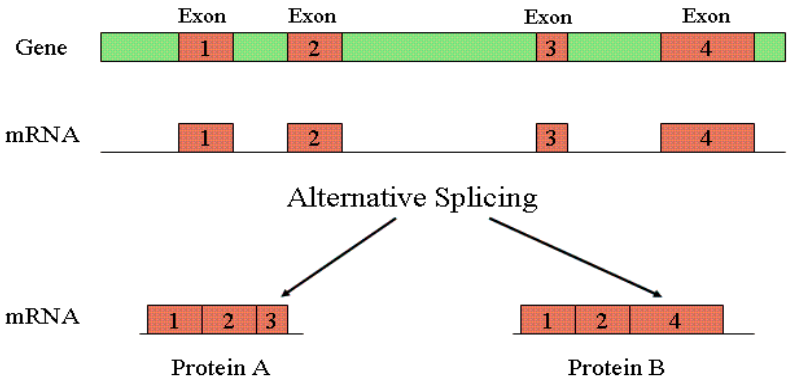
- **Gene expression:**
  - **Definition**: it is the process of transcribing a copy of DNA and then translating it into a protein.

- **Transcription:**
  - **Definition**: synthesis of an RNA-transcript that is complementary to one of the strands of DNA. This process occurs in the nucleus and mitochondria and is catalyzed by the enzyme (DNA-dependent RNA polymerase).
  - **Requirements of transcription:**
    - ✓ DNA.
    - ✓ RNA-polymerase.
    - ✓ Enhancers.

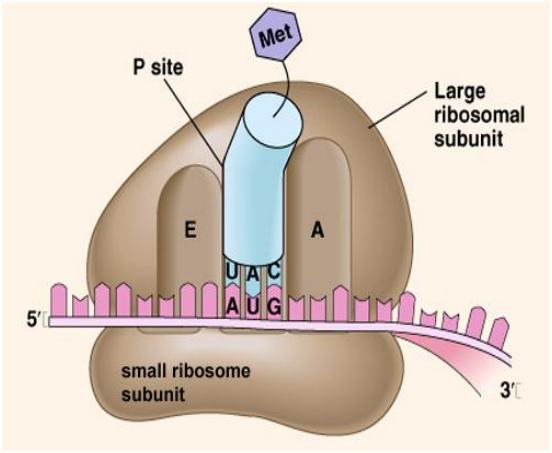
- ✓ ATP (energy).

- **There are three stages of transcription (remember that the RNA-transcript is synthesized as a single-strand in the 5'→3' direction):**
  - ✓ Initiation: RNA-polymerase binds to the DNA at the promoter region and the DNA helix unwinds.
  - ✓ Elongation: RNA-polymerase moves downstream and adds ribonucleotides.
  - ✓ Termination: RNA-polymerase reaches a sequence of DNA after the gene ends (termination sequence).

    Notes:
    - ❖ The DNA template strand is called anti-sense strand.
    - ❖ The opposite non-template strand is called the sense strand.
- **RNA-polymerase is the main but not the only molecule/enzyme needed for transcription:**
  - ✓ Promoters: these are found near the genes which are needed to be transcribed. TATA box is the most common promoter.
  - ✓ Enhancers: enhances transcriptional activity.
  - ✓ Silencers: inhibiting the process of transcription.
  - ✓ Insulators: preventing genes from being incorrectly transcribed.
  - ✓ Basal factors: they form a complex which will initiate transcription.
- **Reverse transcription:**
  - ✓ Definition: transcribing cDNA copies from RNA (reversing the process).
  - ✓ Found in:
    - ❖ *Telomerase*: in eukaryotic cells.
    - ❖ *RNA-viruses*: such as HIV virus.
- **Post-transcriptional modifications:**
  - ✓ Why do we need these modifications? → because mRNAs are not ready to be translated into protein directly after being transcribed from DNA.
  - ✓ These modifications include:
    - ❖ *Capping*: of 5' end by methylated guanosine triphosphate. This guides the ribosome to the site of attachment and determines when translation begins.
    - ❖ *PolyA tail*: of 3' end. This facilitates the transport of mRNA out of the nucleus and protects it from degradation by hydrolytic enzymes.
    - ❖ *Splicing:*
      - ➢ Pre-mRNA contains both introns (which are not needed) and exons. Introns will be removed by splicesome to produce mature mRNA.
      - ➢ Splicesome: it is a large RNA-protein complex found in cytosol of eukaryotic cells.
      - ➢ Alternative splicing: this occurs in eukaryotes when different types of proteins are needed to be made from the same gene (it is important in gene regulation).

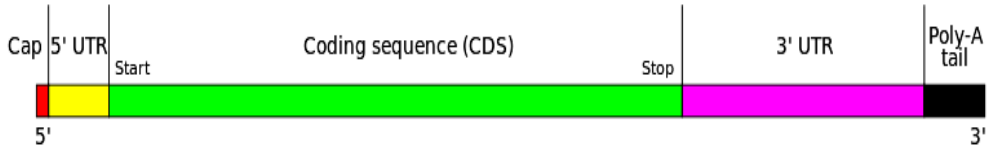


- **Translation:**
  - **Definition**: it is the process at which RNA specifies the synthesis of proteins in ribosomes (which are found in the cytosol of the cell).
  - **Requirements of translation:**
    - ✓ Ribosomes
    - ✓ mRNA
    - ✓ tRNA
    - ✓ Amino acids
    - ✓ Initiation, elongation and termination factors.
    - ✓ ATP (energy).
  - **Genetic information is encoded as a sequence of non-overlapping base triplets (codons):**
    - ✓ The genetic code consists of 64 codons.
    - ✓ Each amino acid is specified by 3 nucleotides.
    - ✓ There are 61 codes for 20 amino acids.
    - ✓ One codon (AUG) is used as a start codon.
    - ✓ Three codons (UAG, UGA, UAA) are considered as stop codons.
  - **Ribosome:**
    - ✓ What is it? → it is the site where proteins are synthesized.
    - ✓ Characteristics:
      - ❖ *It consists of small and large subunits:*
        - ➢ Small subunit (40S).
        - ➢ Large subunit (60S): which contains three tRNA binding sites:
          - A-site: aminoacyl tRNA (a tRNA bound to an amino acid).
          - P-site: peptidyl tRNA (a tRNA bound to the peptide being synthesized).
          - E-site: a free tRNA before it exits the ribosome.



      - ❖ When assembled, it can bind to tRNA and carry amino acids.
      - ❖ There are thousands of ribosomes in each cell!
  - **tRNA:**
    - ✓ Each tRNAcarries a specific amino acid on one end and has an anti-codon on the other end (which is complementary to the codons of mRNA that is being translated).
  - **Stages of translation:**
    - ✓ Initiation: the AUG (start codon) is recognized by methionyl-tRNA. Ribosomes will bind to mRNA and the first amino acid attaches to its tRNA.
    - ✓ Elongation: tRNA delivers the amino acid to be incorporated into the growing peptide chain to the ribosome.
    - ✓ Termination: translation ends when a stop codon is reached (UAA, UAG or UGA). The ribosome will release mRNA and the polypeptide.

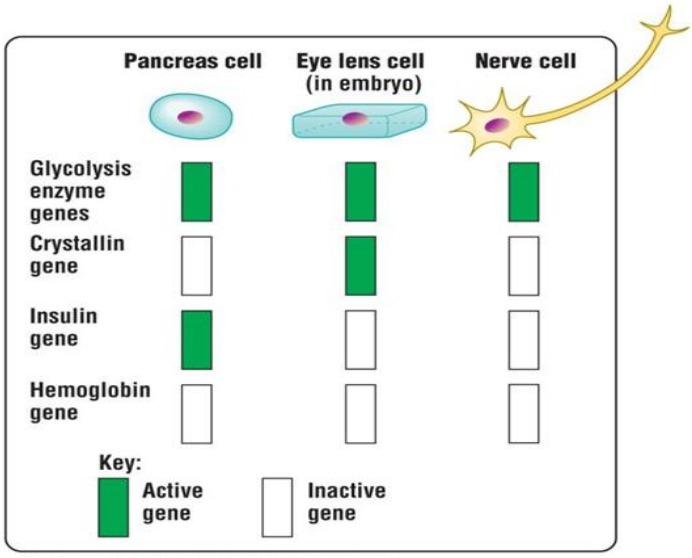
Note: there is an untranslated region at the beginning of mRNA (5'-untranslated region) and another one beyond the stop codon (3'-untranslated region).



- **Post-translational modifications:**
  - ✓ Chemical modification.
  - ✓ Folding.
  - ✓ Localization.

  Note: effects of post-translational modification:
  - ❖ Stability of the protein.
  - ❖ Biochemical activity (activity regulation).
  - ❖ Protein targeting (protein localization).
  - ❖ Protein signaling (protein-protein interaction, cascade amplification).

- **Regulation of gene expression:**
  - **Why is it needed?**
    - ✓ It plays a central role in the development from a zygote to a multicellular organism.
    - ✓ It is essential for cell specialization/ differentiation. Specialized human cells show distinct patterns of gene expression.



  - **Different gene categories:**

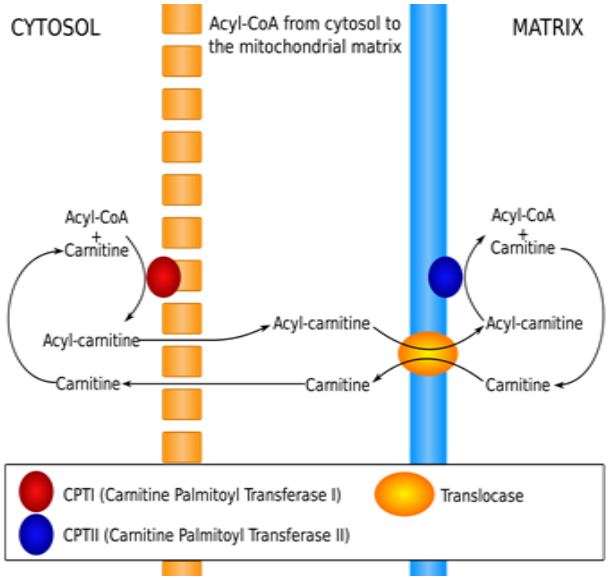
Housekeeping genes	They are activated in all cells at all times (example: transcription machinery)
Cell type specific genes	Activated in each cell that gives a cell its special properties and function
Developmental regulatory genes	Genes specific to certain stages during growth and development of a person
Inducible genes	Genes which are not normally expressed but can be in response to external stimuli

  - **Gene regulation is regulated at several steps:**
    - ✓ Transcription initiation and elongation.
    - ✓ Processing of mRNA.
    - ✓ Translation.
    - ✓ Transport.
    - ✓ Stability.

- **Epigenetic effects on gene regulation:**
  - ✓ Definition: it is the study of heritable changes in gene activity and expression that occur without alteration in DNA sequence.
  - ✓ Each cell turns-on only a fraction of its genes → the rest of genes will be turned-off.
  - ✓ Epigenetic gene regulation mechanisms:
    - ❖ *Histone acetylation*: acetylation of lysine loosens the association between DNA and histone → transcription is initiated. On the other hand, histone deacetylation leads to a silent gene.
    - ❖ *DNA methylation*: mtheylation of cytosine in the gene promoter region → inhibits the initiation of transcription → gene is silent. On the other hand, unmethylated DNA activates the gene.
    - ❖ *microRNAs*

- **The Carnitine Palmitoyl Transferase (CPT) system:**

- Long-Chain Fatty Acids (LCFAs) undergo catabolism through β-oxidation in mitochondria but they require a shuttle system (CPT) to be transported to mitochondrial matrix.



- **CPT1 is encoded by three separated genes in different tissues of the body:**
  - ✓ CPT1a: expressed in almost all cells except brown adipose cells and skeletal muscle cells.
  - ✓ CPT1b: expressed in heart and skeletal muscles and brown adipose cells.
  - ✓ CPT1c: expressed in brain and testes.
- **Regulation of CPT genes:**
  - ✓ CPT1 and CPT2 differ with respect to:
    - ❖ Tightness of their membrane association.
    - ❖ Subcellular targeting.
    - ❖ Sensitivity to malonyl-CoA (which specifically inhibit CPT1 activity) and structurally related compounds.
  - ✓ The additional N-terminal domain in CPT1 is essential for:
    - ❖ Enzyme activity.
    - ❖ Import into the MOM.
    - ❖ Malonyl-CoA sensitivity.
    - ❖ Regulation of enzyme activity by malonyl CoA.
  - ✓ Regulation of mitochondrial fatty acid oxidation mainly involves CPT1. The entire pathway is regulated by malonyl-CoA which provides a signal to the cell on the availability of lipids for energy needs.