L # 2: From DNA to Protein Synthesis

Definition and Composition of DNA

- Deoxyribonucleic acid (DNA) is a polymer composed of monomer units called **nucleotides**. Each nucleotide consists of three components:
  1) **Phosphate Group:** Provides structural integrity and links nucleotides via phosphodiester bonds.
  2) **Deoxyribose Sugar:** A five-carbon sugar that forms the backbone of the DNA strand.
  3) **Nitrogenous Base:** There are four types—adenine (A), thymine (T), cytosine (C), and guanine (G).
- These are the nucleotides that make up DNA. Each nucleotide in DNA has a sugar called **deoxyribose**, a phosphate group, and one of four nitrogenous bases:

A- Purines (double-ring structure):

1. **Adenine (A)**
2. **Guanine (G)**

B- Pyrimidines (single-ring structure):

3. **Cytosine (C)**
4. **Thymine (T)**

------Figure 1: Nucleotide Structure------

Features of DNA Structure

1) The Double Helix Model

DNA has two intertwined strands forming a helical structure. This model [1] explains the mechanism of DNA replication and [2] the storage of genetic information.

----Figure 2: The Double Helix Structure of DNA---

2) Base Pairing and Hydrogen Bonds

The two strands of DNA are held together by hydrogen bonds between complementary bases:

- **Adenine (A)** pairs with **Thymine (T)** via two hydrogen bonds.
- **Cytosine (C)** pairs with **Guanine (G)** via three hydrogen bonds.

This specific base pairing ensures accurate replication and transcription of genetic information.

---Figure 3: Base Pairing and Hydrogen Bonding in DNA---

3) Antiparallel Strands

The two DNA strands run in opposite directions, termed antiparallel orientation. One strand runs in the 5’ to 3’ direction, while the complementary strand runs 3’ to 5’. This antiparallel arrangement is crucial for replication and enzymatic processes.

Figure 4: Antiparallel Nature of DNA Strands

Transcription of DNA to mRNA

Transcription is the process by which a specific segment of DNA is copied into RNA by the enzyme RNA polymerase. This process is the first step in gene expression, leading to the synthesis of proteins that perform various functions within the cell. Transcription ensures that genetic information is accurately conveyed from DNA to RNA, which then serves as a blueprint for protein synthesis during translation.

The Central Dogma of Molecular Biology

The central dogma outlines the flow of genetic information within a biological system:

DNA $\xrightarrow{\text{Transcription}}$ RNA $\xrightarrow{\text{Translation}}$ Protein

This framework emphasizes the sequential nature of genetic information transfer, highlighting transcription as the bridge between DNA and protein synthesis.

Mechanism of Transcription

Transcription involves three main stages: initiation, elongation, and termination. Each stage is orchestrated by various proteins and regulatory elements to ensure accurate and efficient RNA synthesis.

1- Initiation

During initiation, RNA polymerase binds to a specific DNA sequence known as the promoter. In prokaryotes, the sigma factor facilitates this binding, whereas eukaryotes rely on a suite of transcription factors.

Elongation

Elongation involves the unwinding of the DNA double helix and the synthesis of a complementary RNA strand. RNA polymerase moves along the DNA template, adding ribonucleotides in the 5’ to 3’ direction.

2- Termination

Termination occurs when RNA polymerase encounters a specific sequence signaling the end of transcription. In eukaryotic cells termination involves cleavage of the pre-mRNA.

---Figure 5: Transcription of DNA---

RNA Processing in Eukaryotes

In eukaryotic cells, the primary RNA transcript (pre-mRNA) undergoes several processing steps to become mature mRNA, which can be translated into protein.

1. 5’ Capping

A modified guanine nucleotide is added to the 5’ end of the pre-mRNA, protecting it from degradation and assisting in ribosome binding during translation.

2. **Splicing**

Introns (non-coding regions) are removed, and exons (coding regions) are joined together to form a continuous coding sequence.

3. **Polyadenylation**

A poly-A tail is added to the 3’ end of the mRNA, enhancing its stability and facilitating nuclear export.

Figure 6: RNA Processing Steps in Eukaryotes

Types of RNA

Understanding the different types of RNA and their functions is fundamental to grasping the importance of RNA processing and modification.

1- **Messenger RNA (mRNA)**

mRNA serves as the intermediary between DNA and protein synthesis. It carries the genetic information from DNA to the ribosome, where it is translated into a specific protein.

2- **Transfer RNA (tRNA)**

tRNA is responsible for bringing amino acids to the ribosome during protein synthesis. Each tRNA molecule recognizes specific codons on the mRNA through its anticodon region.

3- **Ribosomal RNA (rRNA)**

rRNA is a structural and functional component of ribosomes, the molecular machines that facilitate protein synthesis. rRNA ensures the proper alignment of mRNA and tRNA and catalyzes peptide bond formation.

4- **Non-coding RNAs**

Non-coding RNAs (ncRNAs) perform various regulatory and catalytic functions without being translated into proteins. Examples include microRNAs (miRNAs), small interfering RNAs (siRNAs), and long non-coding RNAs (lncRNAs).

Translation

Translation is the process by which ribosomes synthesize proteins using mRNA as a template. It involves decoding the nucleotide sequence of mRNA into a specific sequence of amino acids, forming polypeptide chains that fold into functional proteins.

The Central Dogma of Molecular Biology: The Genetic Code

Codons and Amino Acids

The genetic code is a set of rules by which the information encoded in DNA is translated into proteins. Each set of three nucleotides, called a **codon**, specifies a particular amino acid.

Table: The Genetic Code

Second letter

First letter	U	C	A	G	Third letter
U	UUU, UUC: Phe UUA, UUG: Leu	UCU, UCC, UCA, UCG: Ser	UAU, UAC: Tyr UAA: STOP UAG: STOP	UGU, UGC: Cys UGA: STOP UGG: Trp	U C A G
C	CUU, CUC, CUA, CUG: Leu	CCU, CCC, CCA, CCG: Pro	CAU, CAC: His CAA, CAG: Gln	CGU, CGC, CGA, CGG: Arg	U C A G
A	AUU, AUC, AUA: Ile AUG: Met	ACU, ACC, ACA, ACG: Thr	AAU, AAC: Asn AAA, AAG: Lys	AGU, AGC: Ser AGA, AGG: Arg	U C A G
G	GUU, GUC, GUA, GUG: Val	GCU, GCC, GCA, GCG: Ala	GAU, GAC: Asp GAA, GAG: Glu	GGU, GGC, GGA, GGG: Gly	U C A G

(Note: You DO NOT HAVE to memorize them)

Redundancy and Universality of the Genetic Code

The genetic code is redundant, meaning multiple codons can encode the same amino acid. Additionally, it is nearly universal across all organisms, highlighting a common evolutionary heritage.

Start and Stop Signals

Certain codons signal the start and termination of protein synthesis:

- **Start Codon:** AUG (Methionine)
- **Stop Codons:** UAA, UAG, UGA

The central dogma describes the flow of genetic information within a biological system:

[DNA]→*Transcription* [RNA]→*Translation* [Protein]

This framework emphasizes that while DNA is transcribed into RNA, RNA is subsequently translated into proteins, which perform various cellular functions.

The Process of Translation

Translation comprises three main stages: initiation, elongation, and termination. Each stage involves specific molecular interactions and enzymatic activities to ensure accurate protein synthesis.

1- Initiation

Initiation marks the beginning of translation, where the ribosome assembles around the start codon (AUG) on the mRNA. Key steps include:

1. **Assembly of the Initiation Complex:** The small ribosomal subunit binds to the mRNA near the 5' cap.
2. **Recruitment of Initiator tRNA:** The initiator tRNA carrying methionine (fMet in prokaryotes) binds to the start codon.
3. **Binding of the Large Ribosomal Subunit:** The large ribosomal subunit joins to form a complete ribosome, positioning the initiator tRNA in the P (peptidyl) site.

2- Elongation

During elongation, amino acids are sequentially added to the growing polypeptide chain. Key steps include:

1. **Codon Recognition:** An aminoacyl-tRNA enters the A (aminoacyl) site, matching its anticodon with the mRNA codon.
2. **Peptide Bond Formation:** The ribosome catalyzes the formation of a peptide bond between the amino acid in the P site and the amino acid in the A site.
3. **Translocation:** The ribosome moves along the mRNA, shifting the peptidyl-tRNA to the P site and freeing the A site for the next aminoacyl-tRNA.

Important Note: in the ribosome, there is an are called The **E site** stands for the **Exit site**, and it plays an important role in the process of elongation during protein synthesis. After **translocation**, when the ribosome moves along the mRNA:

1. The **tRNA** that was in the **P site** (and already donated its amino acid to the growing chain) is now empty—it's no longer carrying an amino acid or the growing polypeptide chain.
2. This empty tRNA is shifted to the **E site** (the exit site) of the ribosome.
3. Once in the E site, the empty tRNA detaches from the ribosome and leaves, making room for the next aminoacyl-tRNA to enter the A site.

3- Termination

Termination occurs when a stop codon (UAA, UAG, UGA) is encountered. Key steps include:

1. **Release Factor Binding:** Release factors recognize the stop codon and bind to the A site.
2. **Polypeptide Release:** The completed polypeptide chain is released from the tRNA in the P site.
3. **Ribosome Disassembly:** The ribosomal subunits dissociate, freeing the mRNA and tRNA for future translation cycles.

Figure 7: Process of translation

Post-Translational Modifications (PTMs)

After synthesis, proteins often undergo PTMs, which are **chemical modifications** that alter protein function, localization, stability, and interactions. PTMs are essential for the dynamic regulation of cellular processes.

DNA Packaging and Chromosomes

DNA is packaged into chromosomes within the cell nucleus. The organization involves wrapping DNA around histone proteins, forming nucleosomes, and further coiling to create compact structures. This packaging facilitates efficient storage and regulation of gene expression.

Figure 8: DNA Packaging from Nucleosomes to Chromosomes

Mutations and Their Effects on the Genetic Code

Mutations are changes in the DNA sequence that can affect the genetic code. Types of mutations include:

	Mutation Type	**Description**	**Effect on Protein**
1.	Missense	Substitution of one amino acid	Changed amino acid
2.	Nonsense	Introduction of a stop codon	Truncated protein
3.	Silent	Substitution does not change amino acid	No change in protein
4.	Frameshift	Insertion or deletion alters reading frame	Altered protein sequence

Table 1: Comparison of DNA and RNA Nucleotides

DNA	Component	RNA
Deoxyribose	Sugar	Ribose
Adenine (A), Thymine (T), Cytosine (C), Guanine (G)	Nitrogenous Bases	Adenine (A), Uracil (U), Cytosine (C), Guanine (G)
Double-stranded helix	Structure	Typically, single-stranded

Review Questions of Lecture #2

1) Describe the structure of a nucleotide and its role in the DNA polymer.
2) What is the genetic code?
3) Differentiate between missense, nonsense, and silent mutations.
4) What are codons, and how do they determine the sequence of amino acids in proteins?
5) Discuss the role of start and stop codons in translation.
6) Explain how mutations can lead to genetic disorders.
7) What are the three components of a DNA nucleotide, and how do they contribute to the structure and function of DNA?
8) Explain the role of hydrogen bonds in DNA base pairing and describe the significance of the antiparallel orientation of DNA strands.
9) What are the three main stages of transcription?
10) Outline the key steps in RNA processing in eukaryotic cells.
11) Compare the roles of mRNA, tRNA, and rRNA in protein synthesis.
12) Describe the three stages of translation (initiation, elongation, and termination) and the roles of ribosome sites (A, P, and E) in this process.
13) Illustrate the flow of genetic information from DNA to protein and discuss the importance of the central dogma in molecular biology.
14) Why are post-translational modifications critical for protein function, and how is DNA packaged into chromosomes to facilitate storage and regulation?
15) What are the major differences between DNA and RNA?