UNIT 7: Structure and function of nucleic acids.
7.1. NUCLEOTIDES
Basics and biological roles.
Nitrogenous bases: purine or pyrimidine bases.
Nucleosides and nucleotides: structure and nomenclature.
Polynucleotides: structure and properties.

7.2 NUCLEIC ACIDS
Functions and classification.
Discovery of the functions and structures of the nucleic acids.
Characteristics.
Secondary and tertiary DNA structure.
DNA stores genetic information.
RNA: Structure and function.
Physico-chemical properties of the nucleic acids. DNA denaturation and renaturation.

7.3 CHROMOSOMES AND GENOMES
Chromosomes and genomes: sizes and shapes.
Eukaryotic chromosomes condensation: nuclear structure.
a) Metabolism: energy carriers:

- ATP: Ester (14 kJ/mol)
- ATP: Anhydride (30 kJ/mol)

Abbreviations of deoxyribonucleoside 5'-phosphates:

<table>
<thead>
<tr>
<th>Base</th>
<th>Mono-</th>
<th>Di-</th>
<th>Tri-</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adenine</td>
<td>dAMP</td>
<td>dADP</td>
<td>dATP</td>
</tr>
<tr>
<td>Guanine</td>
<td>dGMP</td>
<td>dGDP</td>
<td>dGTP</td>
</tr>
<tr>
<td>Cytosine</td>
<td>dCMP</td>
<td>dCDP</td>
<td>dCTP</td>
</tr>
<tr>
<td>Thymine</td>
<td>dTMP</td>
<td>dTDP</td>
<td>dTTP</td>
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Abbreviations of ribonucleoside 5'-phosphates:

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<td>CTP</td>
</tr>
<tr>
<td>Uracil</td>
<td>UMP</td>
<td>UDP</td>
<td>UTP</td>
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</table>
b) Enzymatic cofactors components:

- **Coenzyme A**
  - Reactive point
  - Adenosine

Flavin adenine dinucleotide (FAD)

Nicotinamide adenine dinucleotide (NAD^+)

β-Mercaptoethylamine
Pantothenic acid

3'-Phosphoadenosine diphosphate (3'-P-ADP)
c) Second messengers ⇒ Cells interact with their environment by means of hormones and other chemical signals. These extracellular chemical signals (first messengers) interact with plasma membrane receptor generating and intracellular second messenger.
d) Genetic information storage (DNA and RNA polymers):
NITROGENOUS BASES: PURINES AND PYRIMIDINES

NOMENCLATURE AND STRUCTURE

Purine

Pyrimidine

Adenine

Guanine

Cytosine

Thymine (DNA)

Uracil (RNA)

Pyrimidines
NITROGENOUS BASES: PURINES AND PYRIMIDINES

NOMENCLATURE AND STRUCTURE

• Some minor purine and pyrimidine bases:

(DNA)
- 5-Methylcytidine
- \( N^6 \)-Methyladenosine
- \( N^2 \)-Methylguanosine
- 5-Hydroxymethylcytidine
- 7-Methylguanosine

(RNA)
- Ribose
- Inosine
- Pseudouridine
- 4-Thiouridine
NITROGENOUS BASES: PURINES AND PYRIMIDINES

PROPERTIES

- They are hydrophobic and highly water insoluble at physiological pH.

- Conjugated molecules (Most of the bond have double bond properties)
  \[
  \begin{align*}
  \text{pyrimidines} & \rightarrow \text{planar molecules} \\
  \text{purines} & \rightarrow \text{almost planar molecules}.
  \end{align*}
  \]

- Free nitrogenous bases exist in two or more than two tautomeric forms.

- They absorb UV light because of the resonance phenomena.

What do you have to know?
Meaning of tautomer
Meaning of resonance
**NUCLEOSIDES AND NUCLEOTIDES: STRUCTURE**

**Nucleoside**: nitrogenous base + pentose (β-furanose)

N-1 of pyrimidines is joined covalently to the C 1’ of the pentose
N-9 of purines are joined covalently to the C 1’ of the pentose

**Nucleotide**: nitrogenous base + pentose (β-furanose) + phosphate group

Esterified to the C 5’

**What do you have to know?**
- What a pentose is
- What a phosphate group is
- What a N-β-glycosyl bond is
7.1 Nucleotides

(a) Deoxyribonucleotides

Nucleotide: Deoxyadenylate (deoxyadenosine 5'-monophosphate)
Symbols: A, dA, dAMP
Nucleoside: Deoxyadenosine

Nucleotide: Deoxyguanylate (deoxyguanosine 5'-monophosphate)
Symbols: G, dG, dGMP
Nucleoside: Deoxyguanosine

Nucleotide: Deoxythymidylate (deoxythymidine 5'-monophosphate)
Symbols: T, dT, dTMP
Nucleoside: Deoxythymidine

Nucleotide: Deoxycytidylate (deoxycytidine 5'-monophosphate)
Symbols: C, dC, dCMP
Nucleoside: Deoxycytidine

(b) Ribonucleotides

Nucleotide: Adenylate (adenosine 5'-monophosphate)
Symbols: A, AMP
Nucleoside: Adenosine

Nucleotide: Guanylate (guanosine 5'-monophosphate)
Symbols: G, GMP
Nucleoside: Guanosine

Nucleotide: Uridylate (uridine 5'-monophosphate)
Symbols: U, UMP
Nucleoside: Uridine

Nucleotide: Cytidylate (cytidine 5'-monophosphate)
Symbols: C, CMP
Nucleoside: Cytidine
Nucleotides containing phosphate groups esterified to carbons located in position different to C 5’

Hydrolysis of RNA under alkaline conditions
'Phosphate group bridges' ⇒ **PHOSPHODIESTER BONDS** ⇒ Link successive nucleotides in nucleic acids (covalent bond).
• Covalent backbone of alternating pentose and phosphate groups.
• Side groups ⇒ Nitrogenous bases.
Oligonucleotides: Short nucleic acid (less than 50 nucleotides).

Polynucleotides: Long nucleic acids.
Covalent backbones: **hydrophilic**.

- Phosphate groups: ionised at physiological pH ($pK_a < 1$).

- Linear polynucleotide strands: specific **POLARITY** and 5’ and 3’ ends well defined.

- The base-stacking interactions make the major contribution to the stability of the double helix.

**Watson and Crick model**
**Definition:** Polymers constituted by nucleotides covalently linked by phosphodiester bonds.

**Functions:** To store, transmit and express the genetic information from one generation to the next.

*Genetic information flow: Molecular Biology Dogma*
FUNCTIONS AND CLASSIFICATION

DNA

Code for all the RNAs and proteins.

RNA

- Ribosomic (rRNA) ➔ structural component of the ribosomes ➔ involved in the protein synthesis.
- Messenger (mRNA) ➔ carries genetic information from DNA (gene) to the ribosome.
- Transfer (tRNA) ➔ translate genetic information code by mRNA into amino acid sequences.
- Other minor RNA: heterogeneous nuclear (hnRNA).
1865- Mendel: published the basic rules of the inheritance.

1869- Friedrich Meischer: discovery of the nuclein (acid molecule containing high phosphate concentration).

Kossel (1882-1889) y Levene (1920s) described the chemical composition of DNA (tetranucleotide).

1928- Fred Griffith observed the transformation of non pathogenic bacteria into pathogenic bacteria.

1944- Avery-Mc Leod and Mc Carty identified DNA as the transforming agent previously described by Griffith.
1950- Erwin Chargaff and co-workers studied the nitrogenous base composition of the DNA isolated from different organisms.

**Chargaff’s rules:**

1. The base composition of DNA generally varies from one species to another.

2. DNA specimens isolated from different tissues of the same species have the same base composition.

3. The base composition of DNA in a given species does not change with an organism’s age, nutritional state, or changing environment.

4. In all cellular DNAs, regardless of the species, $A = T$ and $G = C$ ⇒ Pur = Pyr ($A + G = T + C$).
1952- Hershey and Chase performed experiments (infection of bacterial cells by a bacteriophage) to demonstrate that DNA and not protein, carried the genetic information.

Early 1950s: Rosalind Franklin and Maurice Wilkins, shed light on the DNA structure using X-ray diffraction (DNA fibers). They deduced that DNA molecules are helical with two periodicities along their long axis.

1953- Watson and Crick relied on the accumulated information about DNA to set about deducing its structure.
WHAT DO YOU HAVE TO KNOW?

Experiments carried out by:

- Griffith
- Avery-McLeod-McCarty
- Hershey and Chase
Nucleic acids have primary, secondary and tertiary structure.

Nucleic acids absorb at wavelengths close to 260 nm (nitrogenous base resonance).

**Hypochromic effect**: decreasing its absorption of UV light relative to that of a solution with the same concentration of free nucleotides.
• Hydrophobic stacking interactions in which two or more bases are positioned with the planes of their rings parallel are stabilise the three-dimensional structure of the nucleic acids (water contact is minimised).

• Second kind of important interaction between nitrogenous bases: hydrogen-bonding patterns in the base pair defined by Watson and Crick.

![Diagram of nucleic acids with Adenine-Thymine and Guanine-Cytosine base pairs]
DNA SECONDARY STRUCTURE

DNA: WATSON AND CRICK MODEL

- Two helical DNA chains wound around the same axis to form a **right-handed double helix**.

- Nitrogenous bases are stacked **inside** the helix, and the covalent backbones are on the outside.

- 10 (10,5) base pair per turn.
  
  
  - Adjacent Bases \( \sim 3.4 \text{ Å} \).
  - Rotation \( \sim 36^\circ \).
  - Diameter \( \sim 20 \text{ Å} \).
  - Pitch of the helix \( \sim 34 \text{ Å} (36 \text{ Å}) \).
Hydrogen bonds between bases from both strands.

There are no restrictions in the nitrogenous bases sequence.

**SPECIFICITY OF THE BASES PAIRED:**

Stearic factors $\Rightarrow$ 10.85 Å distance between the two C 1' (N-\(\beta\)-glycosyl bond) corresponding to two base-paired.

Hydrogen bonding factors $\Rightarrow$ H atoms involved have well defined position within the base structure.
**B-DNA** ⇒ Watson and Crick model.

**A-DNA** ⇒ right-handed double helix wider and shorter than B-form. 11 pb per turn and 26 Å diameter. It is present when the relative humidity is reduced up to 75%.

**Z-DNA** ⇒ left-handed double helix. 12 pb per turn and 18 Å diameter. Dinucleotide(XpYp).
Important roles related to proteins-DNA binding or regulation of the DNA metabolism.

- **Bends**: 4 or more adenosine residues appear sequentially in one strand.

- **Palindrome**: regions of DNA with inverted repeats of base sequence having twofold symmetry over two strands of DNA. They have the potential to form hairpin or cruciform structures.

- **Mirror repeat**: The inverted repeat occurs within each individual strand. They cannot form hairpin or cruciform structures.
The inverted repeats are self-complementary within each strand: **Hairpin (1 strand, RNA)** and **Cruciform structures (DNA)**
WHAT DO YOU HAVE TO KNOW?:
- Why must DNA be supercoiled?
- When is DNA in a relaxed state?
- What are topoisomers?
- Differences between negative and positive supercoils.
- What is the linking number?
- What are topoisomerases?
- Types of topoisomerases and reaction catalysed by them.
DNA STORES GENETIC INFORMATION.

**Structural gene:** gene coding for polypeptides or RNA; i.e., encodes primary sequences related to a genetic product.

**Regulatory sequences:** provide signals that may denote the beginning or the end of genes, or influence the transcription of genes, or function as initiation points for replication and recombination.
**COLINEARITY**  ⇒  Alignment of the coding nucleotide sequences of DNA and mRNA (triplets = codons) and the amino acid sequence of a polypeptide chain.

DNA STORES GENETIC INFORMATION.
• Eukaryotic genes ⇒ INTRONS (they are not transcripted).

• There is no colinearity.

• Coding segments ⇒ EXONS.

In several bacteria and many eukaryotic genes, coding sequences are interrupted at intervals by regions of noncoding sequences.
Genetic information (DNA) $\Rightarrow$ mRNA $\Rightarrow$ proteins

- Transmission of genetic information from the nucleus to the cytoplasm.

- One mRNA molecule per gene or group of genes to be expressed.

- Prokaryotes: one mRNA molecule can code for one or several polypeptide chains.
• Prokaryotes, mRNA coding for just one polypeptide chain ⇒ **MONOCISTRONIC**.

• mRNA coding for two or even more polypeptide chains ⇒ **POLYCI STRONIC**.

• Most of the eukaryotic mRNA are monocistronic.

• No coding RNA involves regulatory sequences of the protein synthesis.
• Simple strand RNA:

The product of transcription of DNA is always single-stranded RNA.

Base pairing between G and U.

Palindromic and self-complementary sequences.

It has no simple, regular secondary structure that serves as a referent point.

• RNA can base-pair antiparallel with complementary regions following the standard Watson and Crick model:

  RNA strand: RNA duplex

  DNA strand: hybrid RNA
Secondary structure of RNA

- Single strands
- Bulge
- Internal loop
- Hairpin

Secondary structure of RNA
Eukaryotes have special-function small RNAs apart from tRNAs, rRNAs and mRNAs:

**Small nuclear RNA (snRNA):** involved in mRNA splicing (introns removal thanks to the spliceosome: RNA-protein complexes). The introns are removed from the primary transcript and the exons are joined to form a continuous sequence that specifies a functional polypeptide.

**MicroRNA (miRNA):** small noncoding RNA molecules (21 nucleotides) complementary in sequence to particular regions of mRNAs. They suppress their translation.

**Small interference RNA (siRNA):** RNA molecules able to facilitate mRNA degradation.

RNA is also a component of the telomerases: enzymes able to keep the structure of the telomeres (the ends of the linear eukaryotic chromosomes) during DNA replication.
Physico-chemical properties depend on the characteristics of the nucleotides.

- Isolated DNA in the native state: high viscosity at pH 7.0 and room temperature.

**DESNAUTURATION = FUSION.**

- Denaturation $\implies$ Hydrogen bonds are and hydrophobic interactions disappeared $\implies$ Nitrogenous bases cleavage became ionised.

- Fusion does not break covalent bonds.
- Tm depends on the bases composition.
- Tm (high % G+C) > Tm (high % A+T).

Separation of the strands → increase of the $A_{260}$ ➞ ➞ HYPERCHROMIC EFFECT.
WHAT DO YOU HAVE TO KNOW?

- Sizes and types of genetic materials
- What are genomes?
- What are chromosomes (prokaryotics and eukaryotics)?
- What are plasmids?
- What is the chromatin?
- What are histones?