DNA structure and chromatin organization

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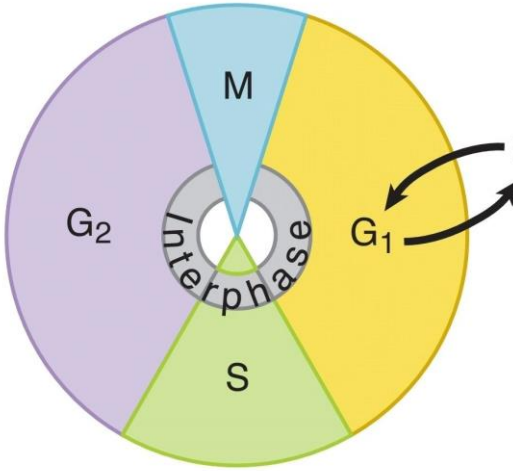
DNA structure and chromatin organization

- Objectives
- Define the nucleoside, nucleotide
- Define nuclear DNA
- Name its different forms and describe the structure of its B form
- Define nucleosome, describe its structure
- List the different levels of chromatin condensation till chromosome
- Differentiate between euochromatin and heterochromatin

Comparison of Gene expression and DNA Replication

Gene Expression	DNA Replication
Produces all the proteins an organism requires	Duplicates the chromosomes before cell division
Transcription of DNA: RNA copy of a small section of a chromosome (average size of human gene, 10^4–10^5 nucleotide pairs)	DNA copy of entire chromosome (average size of human chromosome, 10^8 nucleotide pairs)
Transcription occurs in the nucleus throughout interphase	Occurs during S-phase
Translation of RNA (protein synthesis) occurs in the cytoplasm throughout the cell cycle	Replication in nucleus

Cell Cycle



G1 phase (gap 1) is a period of cellular growth preceding DNA synthesis.

- Cells that have stopped cycling, such as muscle and nerve cells, are said to be in a special state called G0 .

S phase (DNA synthesis) is the period of time during which DNA replication occurs.

- At the end of S phase, each chromosome has doubled its DNA content and is composed of 2 identical sister chromatids linked at the centromere.

G2 phase (gap 2) is a period of cellular growth after DNA synthesis but preceding mitosis.

Replicated DNA is checked for any errors before cell division

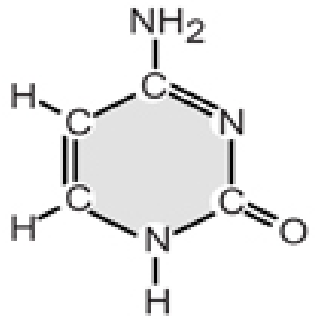
Clinical application

- Many chemotherapeutic agents function by targeting specific phases of the cell cycle.
- Some commonly tested agents with phase of cell cycle they target:
- S-phase: methotrexate, 5-fluorouracil, hydroxyurea
- G2 phase: bleomycin
- M phase: paclitaxel, vincristine, vinblastine
- Non cell-cycle specific: cyclophosphamide, cisplatin

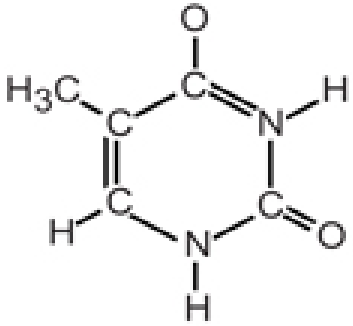
Nitrogenous Bases

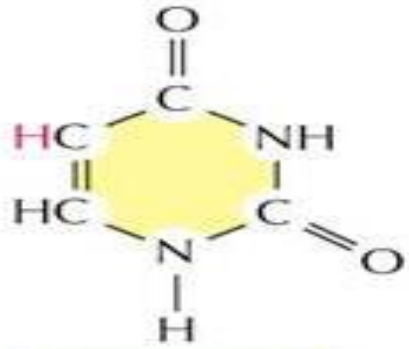
Pyrimidines

C Cytosine



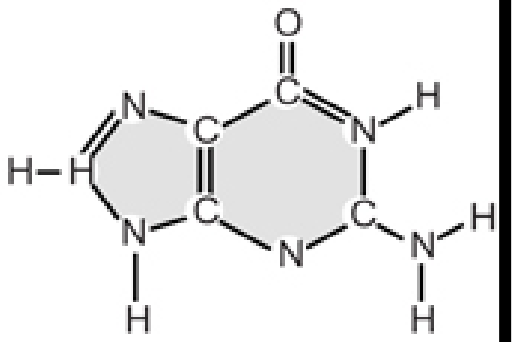
T Thymine



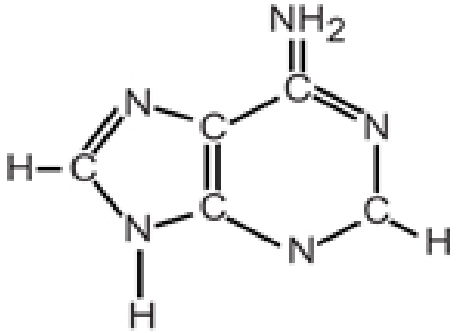


Purines

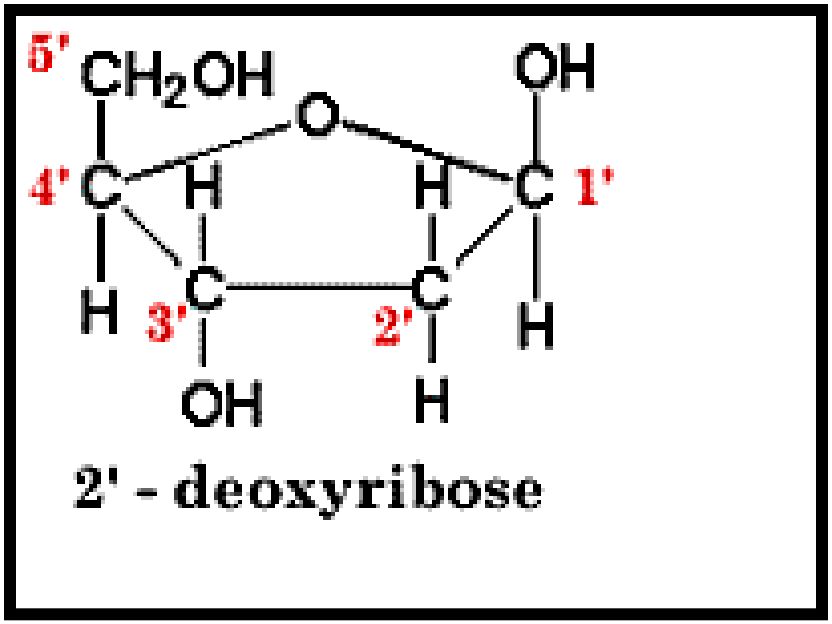
G Guanine



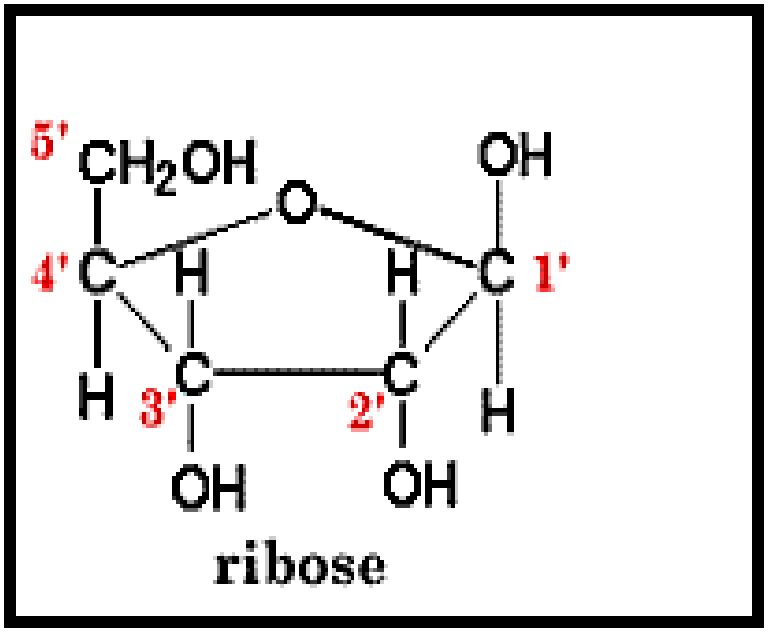
A Adenine



Pentose Sugar ‘Ribose’



2' - deoxyribose



ribose

Nucleotide

Nucleotide composed of:

1- nitrogenous base.

2- Pentose sugar.

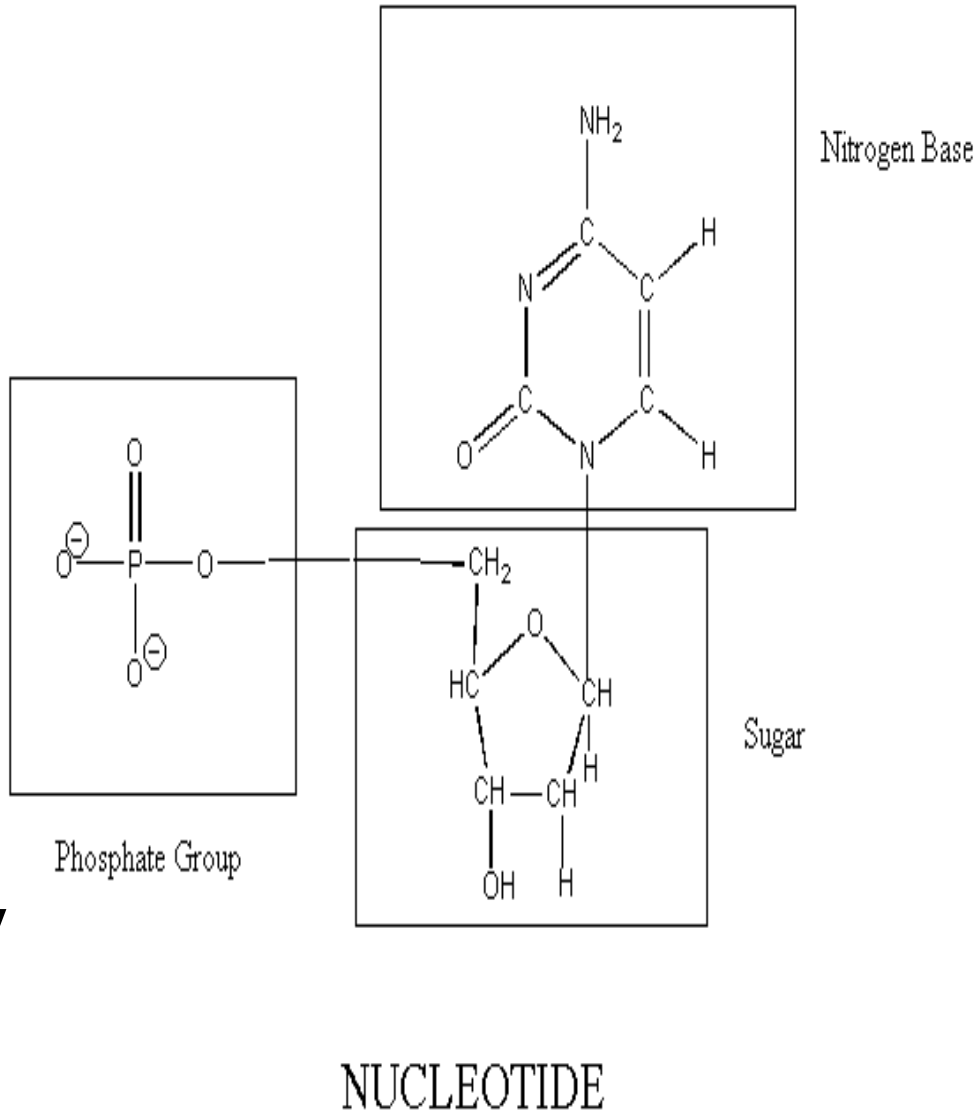
2-deoxyribose in DNA

ribose in RNA

attached by its 1' Carbon to N1 of pyrimidine or N9 of purines by *a glycosidic bond*.

3- *phosphate group* attached to 5 ' -Carbon of the sugar.

***A Nucleic acid*: is a polynucleotide; composed of nucleotides linked together by phosphodiester linkages**



NUCLEOTIDE

Nucleosides & Nucleotides

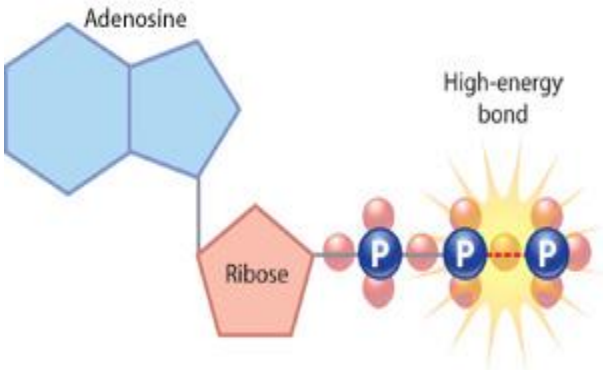
Nitrogenous Base[NB]	NB+Ribose Sugar[RS] =Nucleoside	NB+RS+P =Nucleotide
Adenine →	Adenosine →	AMP [adenosine monophosphate, adenylic acid]
Guanine →	Guanosine →	GMP [Guanosine Monophosphate, Guanylic Acid]
Cytosine →	Cytidine →	CMP [Cytidine monophosphate, Cytidylic Acid]
Thymine →	Thymidine →	TMP [Thymidine monophosphate, Thymidilic Acid]
Uracil →	Uridine →	UMP [Uridine Monophosphate, Uridylic Acid]

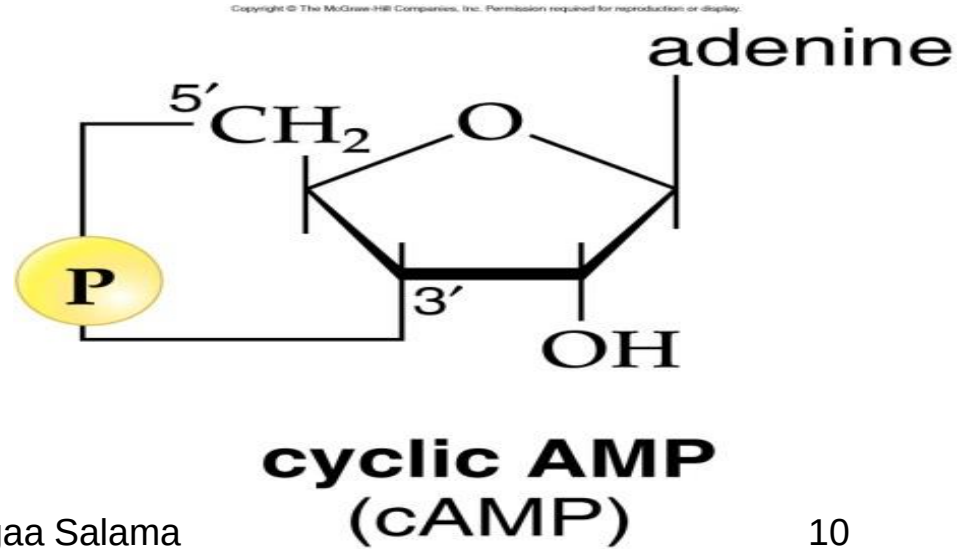
Formation and breakdown of cAMP:

Adenylate cyclase **Phosphodiesterase**

ATP ⇒ 3,5-cAMP + PPi ⇒ 5'-AMP

- It functions as a second messenger for many hormone receptors (group IIa)
- acts as a regulator for carbohydrate and lipid metabolism.

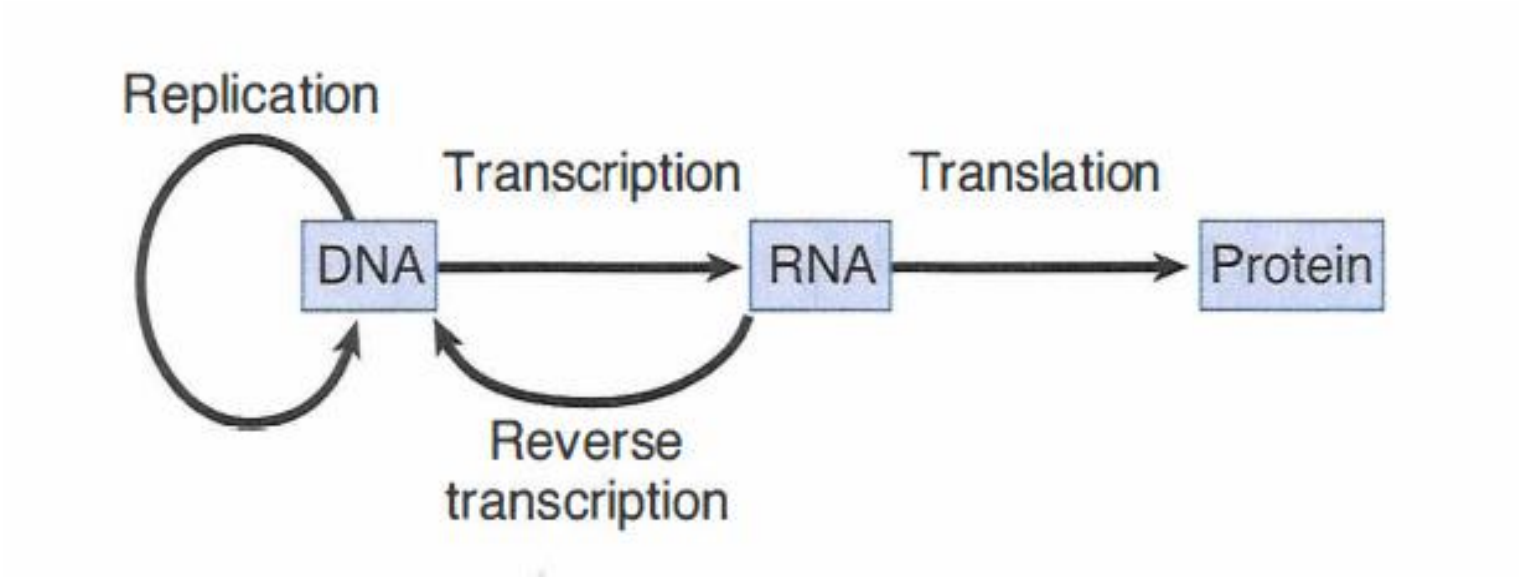




DNA functions

1.Storage of genetic information and transferring it to daughter cells during mitosis by *replication.*

2.Transcription: Formation of rRNA, tRNA, mRNA during protein synthesis [Translation].



DNA Structure (orders)

1.Primary: Linear sequence of deoxyribonucleotide units
2.Secondary: Double Stranded Helix
3.Tertiary:double helix wrapped around histone
→Nucleosome
4.Higher Orders: Formation of chromatin, (30nmFibers), Chromosomes

Using Chargaff's Rules

- In dsDNA (or dsRNA) (ds = double-stranded)
- % A = % T (% U) % G = % C
- % purines = % pyrimidines

A sample of DNA has 10% G; what is the % T? 10% G + 10% C = 20% therefore, % A + % T must total 80% 40% A and 40% T Ans: 40%

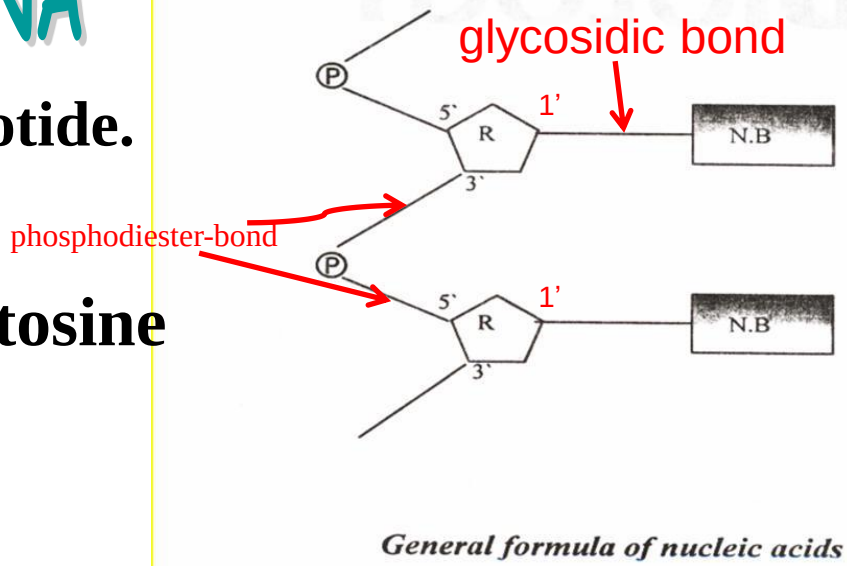
The primary structure of DNA

-Each strand of DNA is a polynucleotide.

-The nitrogenous bases

adenine, guanine, thymine and cytosine

Deoxyribose and phosphat



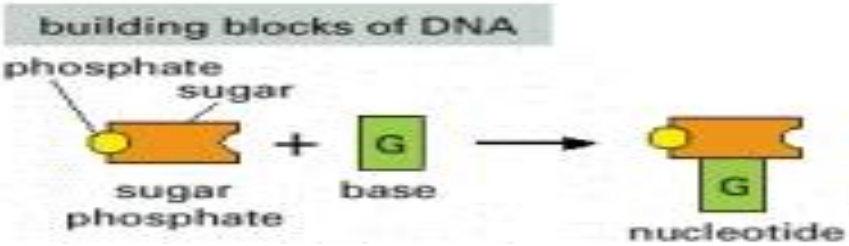
General formula of nucleic acids

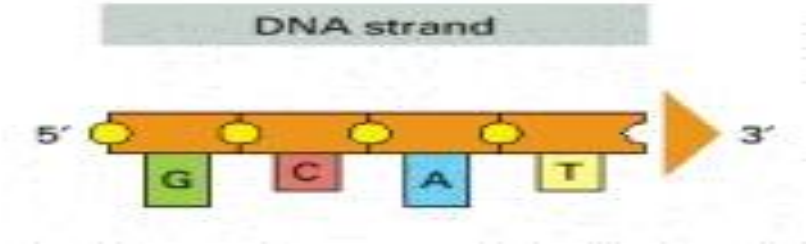
- DNA nucleotides are held in a polymeric form by ***phosphodiester-bonds*** between 3'hydroxyl of the sugar of one nucleotide to the 5' hydroxyl of the sugar of the next nucleotide through a phosphate molecule.
- 4 deoxyribonucleotides [dAMP, dGMP, dTMP, dCMP].

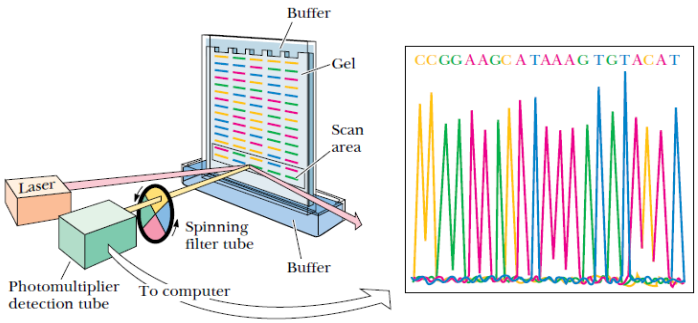
The *sugar-phosphate* linkages form the backbone of the polymer to which the variable bases stick.

The nucleotide polymer has a free phosphate group attached to 5' position of sugar and a free 3' hydroxyl-group; this is called polarity of DNA molecule.

The base sequence of the polymer is always read, written from 5' to 3' direction e.g. 5' GCAT 3'.



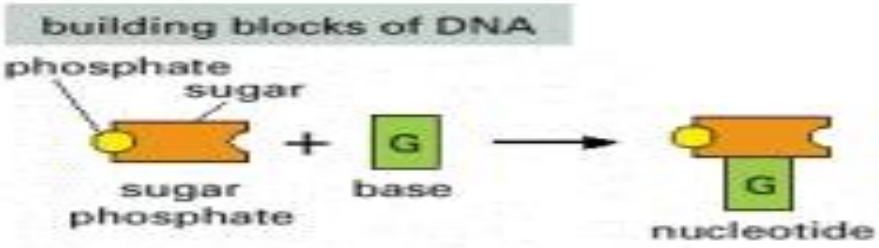


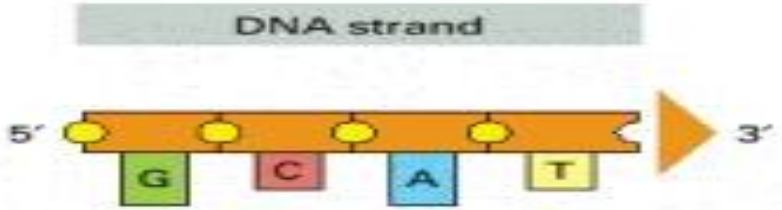


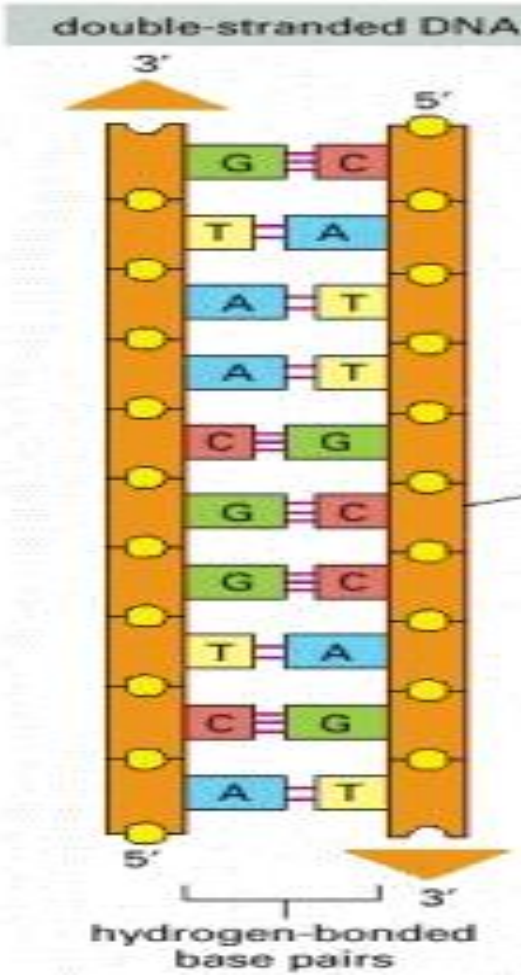
DNA Sequencer

The secondary structure of DNA

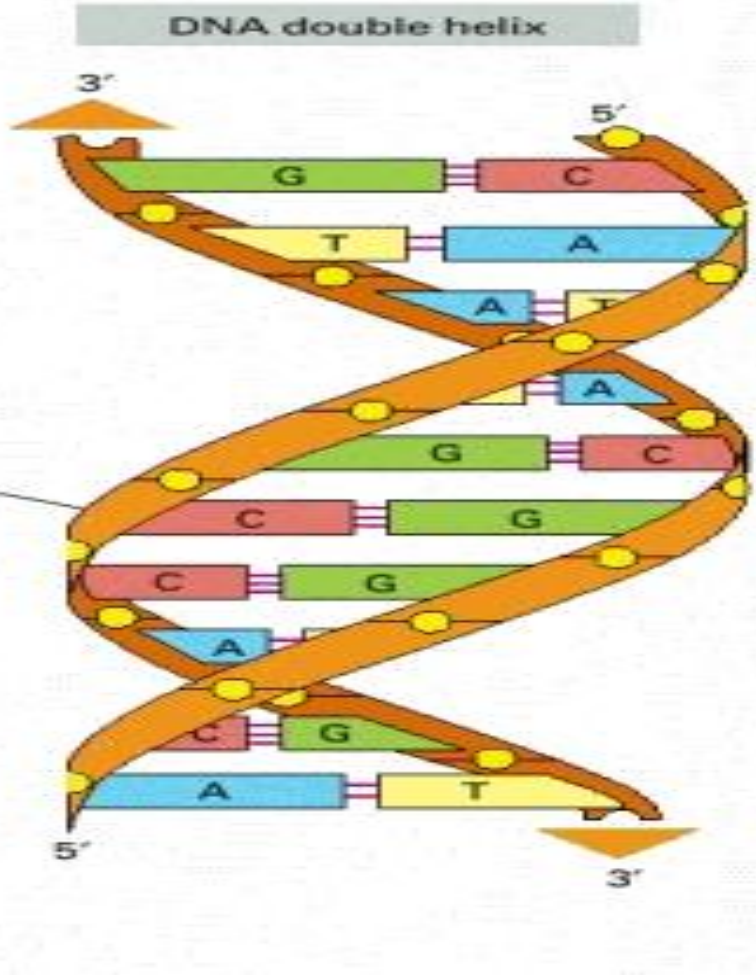
- **DNA is a double stranded helix formed of 2 polynucleotide chains.**
- **The 2 chains are connected by hydrogen bonds.**
- **Adenine on one strand binds to thymine (A=T) of the opposite strand by 2 hydrogen bonds.**
- **Guanine on one strand binds to cytosine (G≡C) of the opposite strand by 3 hydrogen bonds, that is called base pairing [Watson and Crick], and base complementarity.**
- **The 2 strands run antiparallel.**
- **One strand is a coding strand; the other is the template strand.**



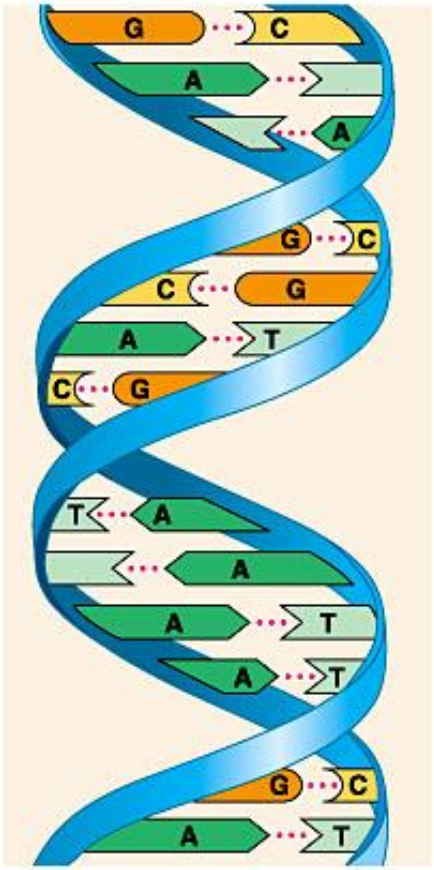




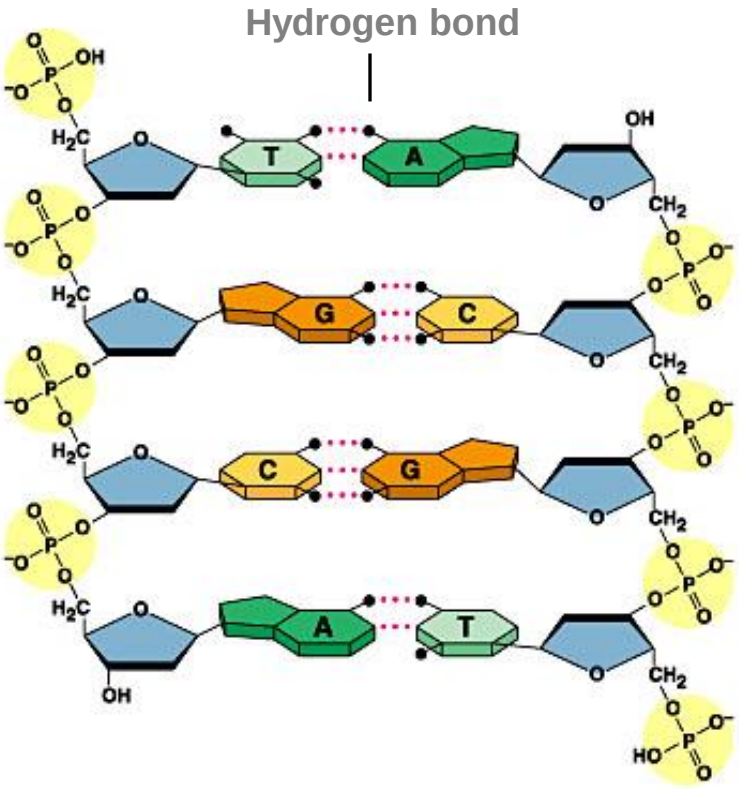
Secondary structure DNA



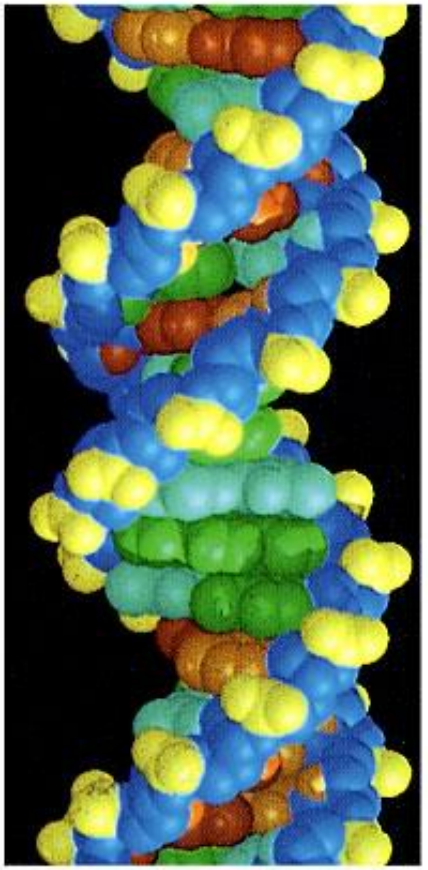
- **Hydrogen bonds between bases hold the strands together: A and T, C and G**



Ribbon model



Partial chemical structure



Computer model

Figure 10.3D

Different forms of DNA

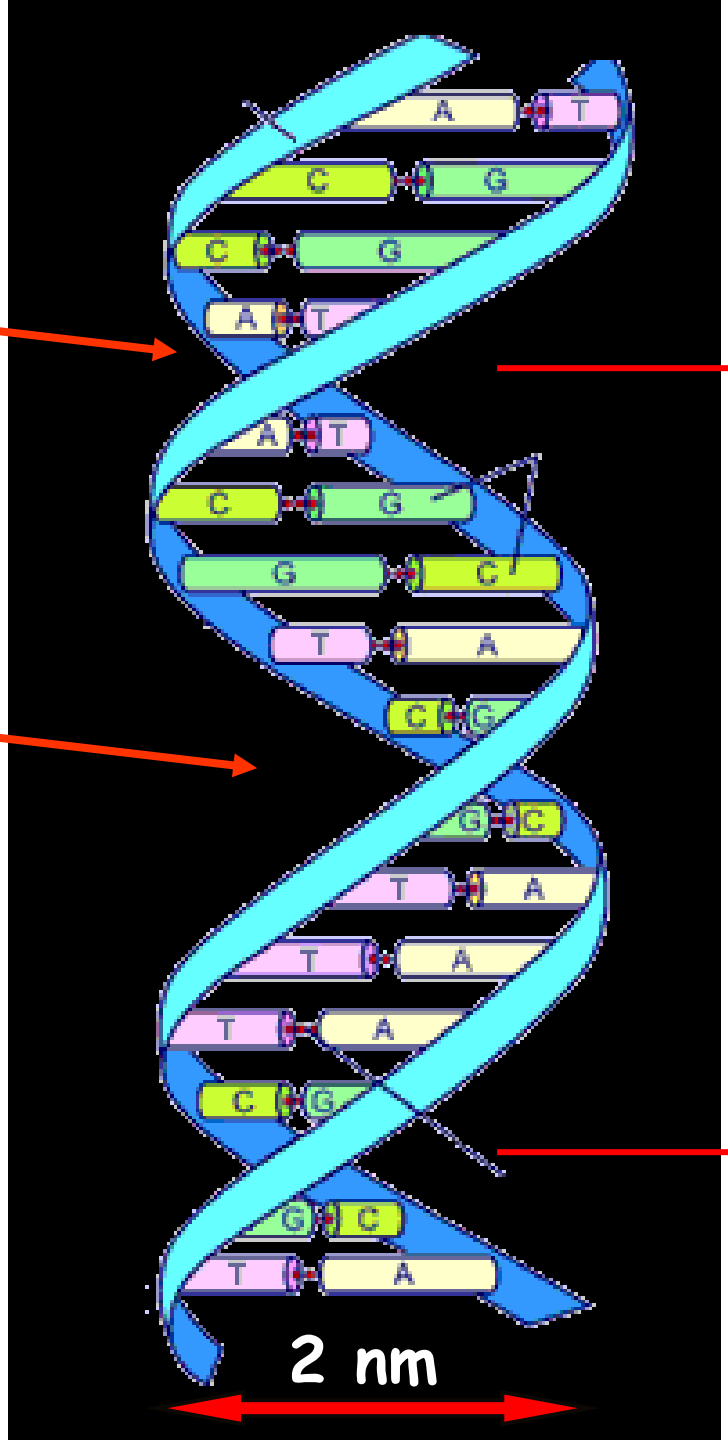
1. ***B- Form [Watson-Crick] DNA structure:***

- **It is right handed double helix.**
- **There are 2 grooves, major and minor [regulatory proteins can bind].**
- **The 2 strands run antiparallel.**
- **The width of dsDNA is 1.9 nm.**
- **There are 10 b.p./turn which repeat every 3.4 nm.**
- **This form is *predominant* under physiological conditions.**

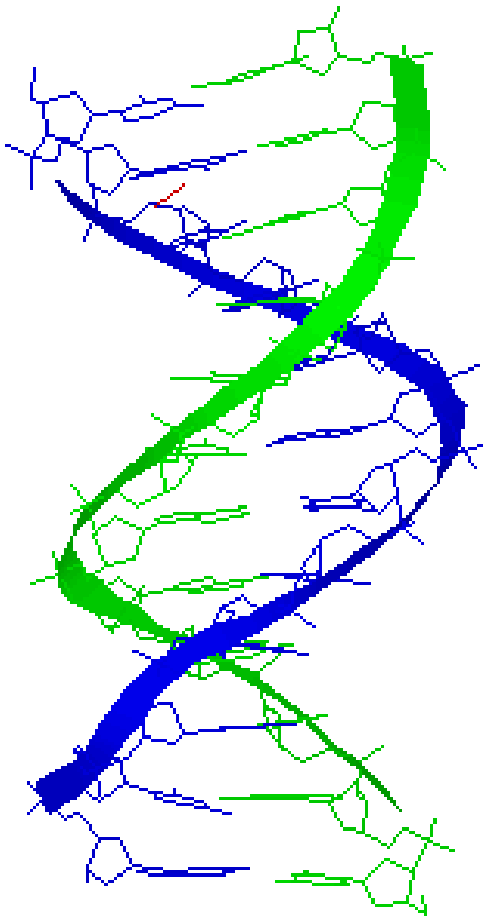
2.A-Form [anhydride form]: Right handed, wider [2.3 nm], condensed, 11 b.p./turn, high salt medium.

3. Z- Form [Zig-zag]: Left handed, elongated [1.8 nm width], phosphodiester bonds are zig-zag.

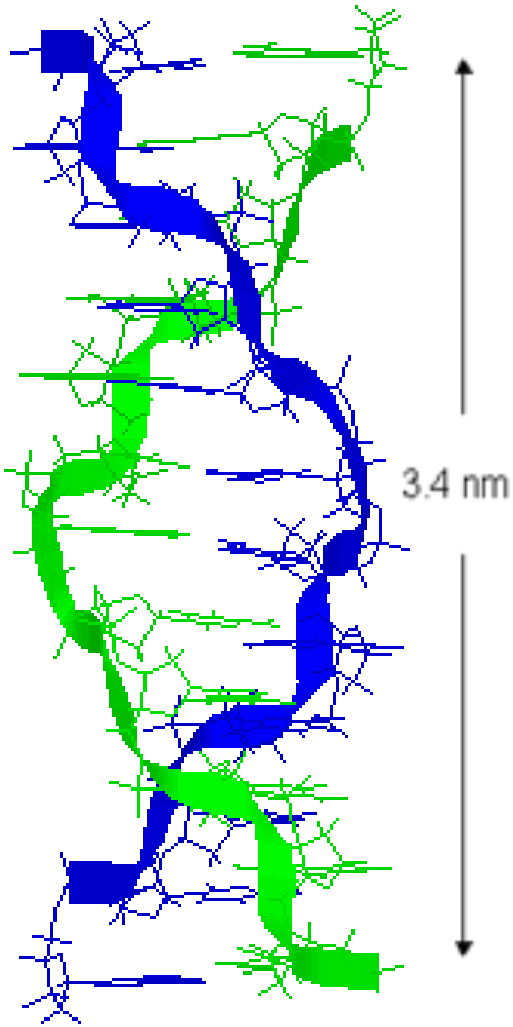
B-Form of DNA



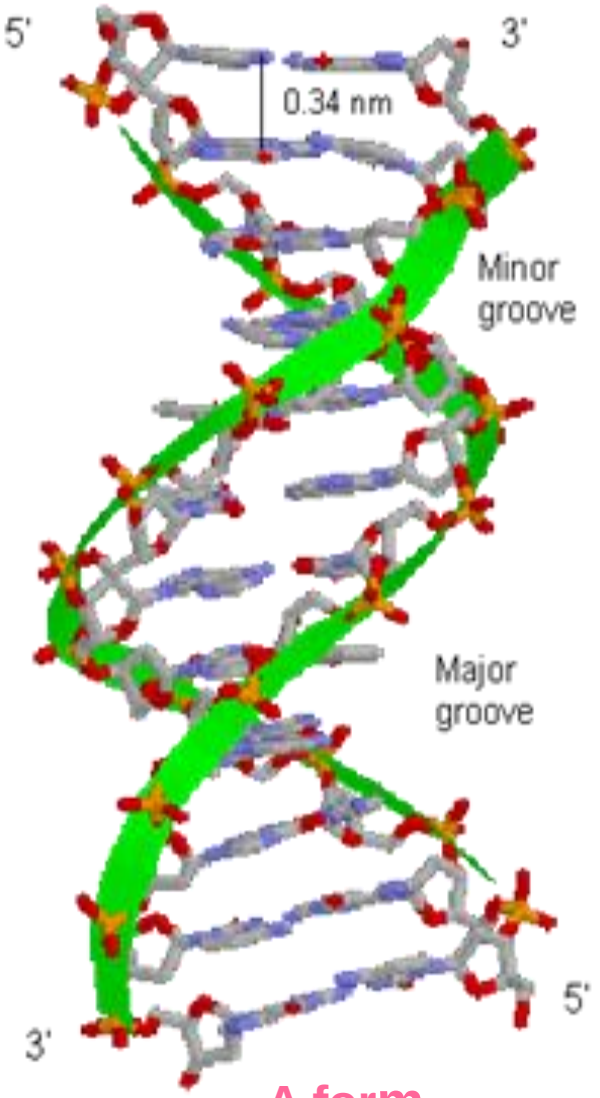
DNA Forms



B Form



Z Form

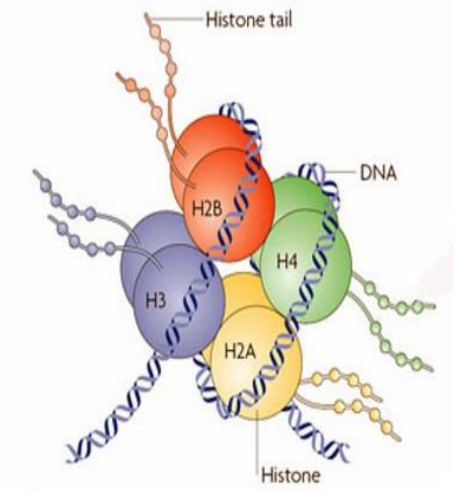


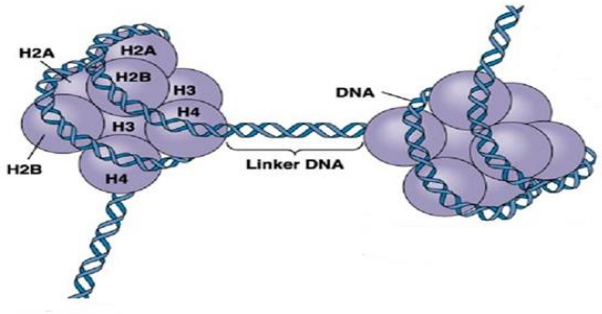
A form

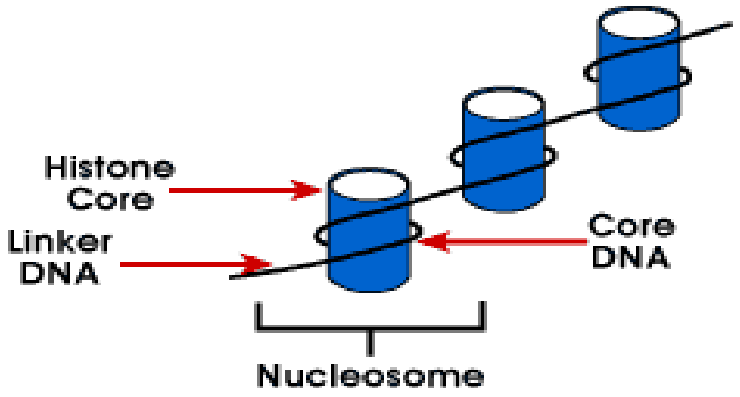
The tertiary structure of DNA

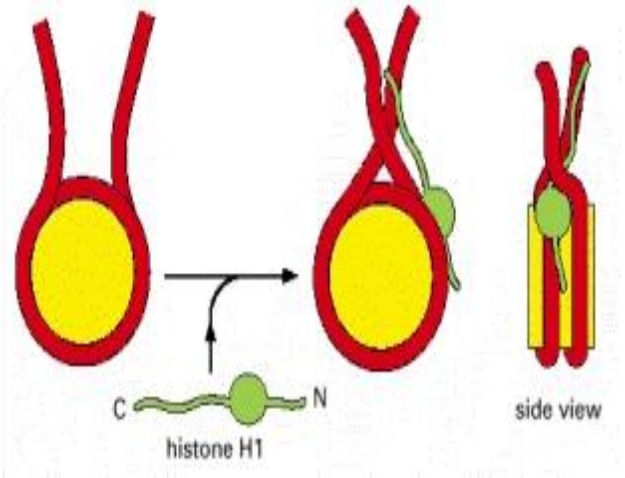
- **The double stranded DNA wraps around a basic protein (histone octamer core) to form the nucleosome [10 nm].**
- Histones are **5** types **H1,H2A , H2B, H3** and **H4**
- **Histones octamer is $[H_2A, H_2B, H_3, H_4]_2$**
- **Nucleosomes are linked by DNA linker wrapped around H_1.**
- **DNA + Histones → nucleosome → chromatin → cromatids →chromosomes.**
- ***Higher orders:***
  - **The Nucleosomes are supercoiled to form the chromatin filaments [30 nm].**
  - **Chromatin filaments are supercoiled to form the cromatids, then the chromosomes.**

The function of nucleosome is to condense and stabilize DNA









30 nm Chromatin fibril

Chromatin is formed of:

1- Double stranded DNA

2- Small amount of RNA

3- Proteins (mainly Histones).

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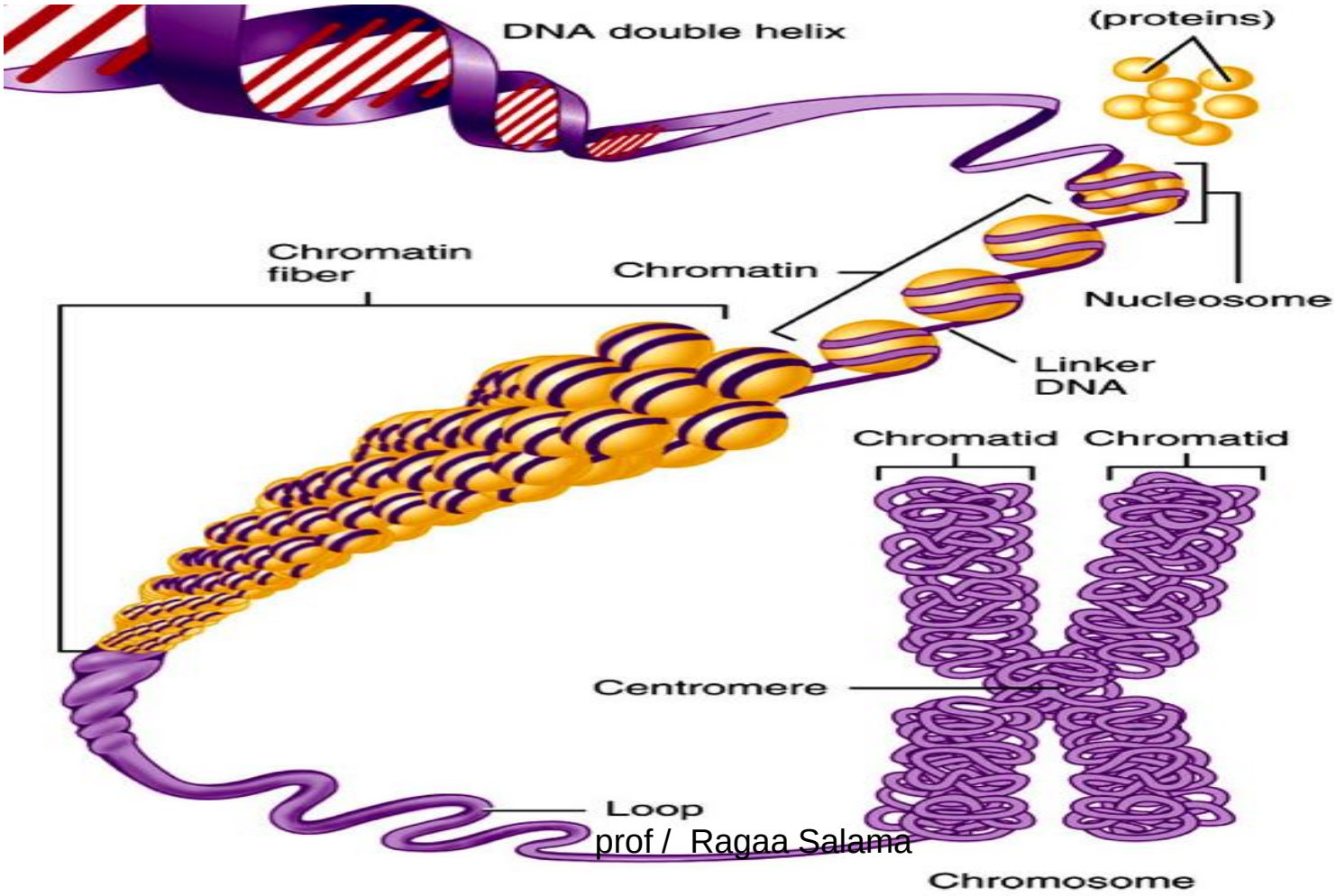
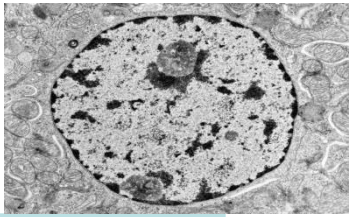
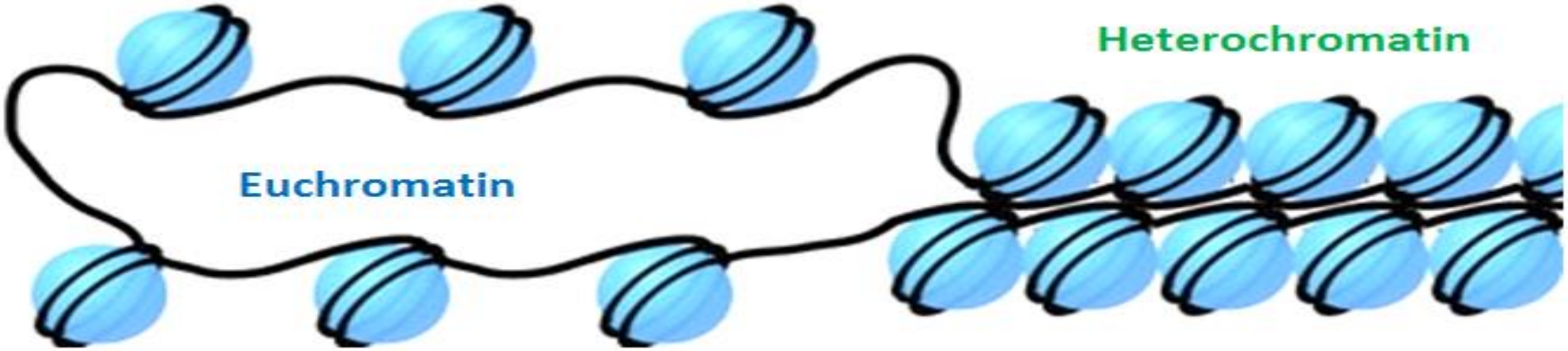


Denaturation and Renaturation of DNA

- Double-helical DNA can be denatured by conditions that disrupt hydrogen bonding and base stacking,→ "melting" of the double helix into 2 single strands that separate from each other.
- No covalent bonds are broken in this process.
- Heat, alkaline pH, and chemicals such as formamide and urea are commonly used to denature DNA.
- Denatured single-stranded DNA can be renatured (annealed) if the denaturing condition is slowly removed.
- If heat denatured DNA is slowly cooled, the two complementary strands can become base-paired again
- in probing a Southern blot and in performing the polymerase chain reaction (PCR)
- When probe DNA binds to target DNA sequences of sufficient complementarity, the process is called hybridization



Euchromatin	Heterochromatin
Loosely packed	Densely packed
Stained light	Stained dark
Transcriptionally active	Transcriptionally inactive



ON

EPIGENETICS

OFF