

Introduction to Molecular biology

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Lab-1

Practical molecular biology

Learning outcomes

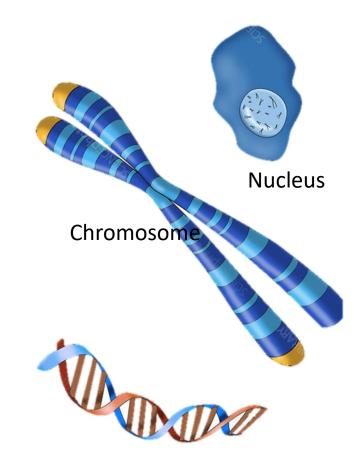
In this lab, you will be able to

- Learn materials; including DNA,RNA and Protein.
- Learn laboratory instrument; including Pipette, Vortex, and Centirfuge

Materials DNA, RNA and Protein.

Structure and organization of chromosomes

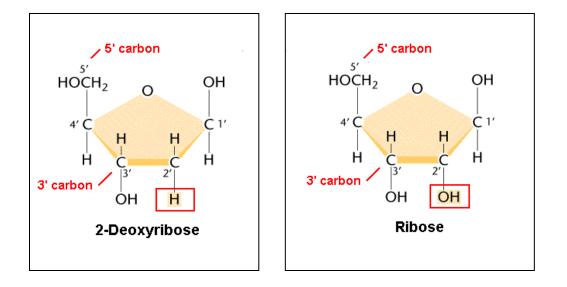
- **Nucleus** is the basic compartment of the cell that is known as the control center of the cell.
- Chromosomes: are invisible structures in the cell's nucleus. Each chromosome is made up of DNA tightly coiled many times around proteins called histones that support its structure.
- DNA: is made up of two long chains of polynucleotides wound together. DNA encodes genetic instructions and is present within the nucleus of the cell. It helps in regulating gene regulation



Gene and DNA

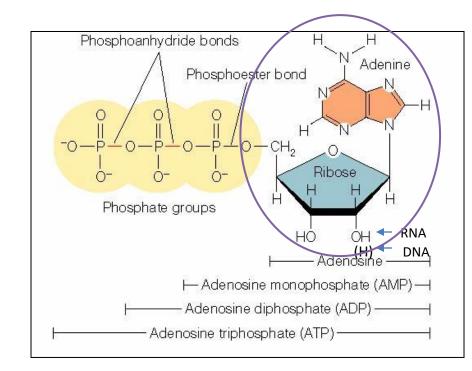
Types of ncleic acids; DNA and RNA

- **DNA** (deoxyribonucleic acid) and **RNA** (ribonucleic acid).
- DNA contains the bases A, G, C and T, but RNA contains A, G and C, but T is replaced by (U).
- **DNA** contains related deoxyribose, but **RNA** contains 5-carbon sugar ribose



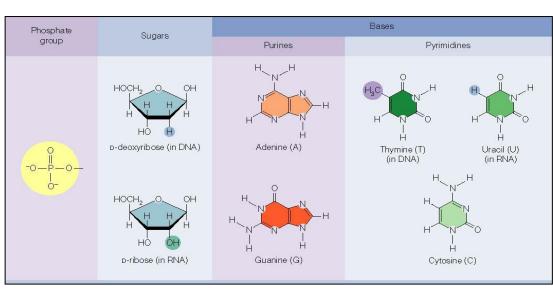
Building blocks of DNA and RNA

- Nucleotide is a building block of DNA or RNA, consisting of a pentose sugar, a base, a phosphate group.
- Nucleoside is the union of a purine or pyrimidine base with a pentose sugar.



Basic units of DNA and RNA

- Phosphate group.
- Sugar: DNA has Deoxyribose, but RNA has Ribose
- **Bases** are the main components that carries genetic information.
- a) Purine = Adenine (A) & Guanine (G)
- b) Pyrimidine = Cytosine (C), Thymine (T) and Uracil (U).



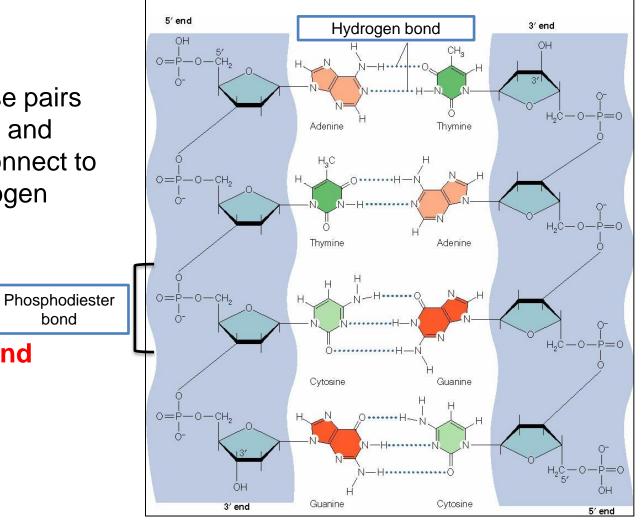
Nucleic acid structure and Hydrogen bonding

Bonds I.Hydrogen Bond

Н.

The complementary base pairs of guanine with cytosine and adenine with thymine connect to one another using hydrogen bonds.

Phosphodiester bond



Types of RNA

- **1. Messengar RNA (mRNA):** is a single-stranded RNA molecule that is complementary to one of the DNA strands of a gene. It encodes a specific protein.
- 2. Non-coding RNA
- A. Housekeeping ncRNA
- **Ribosomal RNA (rRNA)** is involved in synthesis of polypeptide chains and major portion of ribosome.
- Transfer RNA (tRNA) carries amino acids to ribosome and recognizes codons on mRNA.

Types of Non-coding (nc)RNA

- **B. Regulatory ncRNA:**
- I. Long ncRN (200nt): regulates gene expression
- II. Small ncRNA
- a) MicroRNA (miRNA; 18-22nt): regulates gene expression by base pairing to 3'UTR of mRNA.
- b) Small nuclear RNA (snRNA; 150nt) involved in the splicing process of mRNA.
- c) Small nucleolar RNA (snoRNA;60-300nt) facilitates chemical modification of RNAs
- d) Small interference RNA (siRNA;) silencing of gene expression.
- e) Piwi-interacting RNA (piRNA; 24-30nt):

Proteins

- There are about 20 amino acids that encoded by four nucleotides A, U, G & C
- Combinations of 2 nucleotides can only encode
 4²=16 different combinations.
- But combinations of 3 (triplets) can encode
 4³=64 combinations.
- 61 of 64 triplets (codons) code for amino acids.
- One for initiation and 3 for stop.

Second position							
		U	С	А	G		
First position	U	UUU UUC UUA UUA UUG	UCU UCC UCA UCG	(UGU UGC UGA Stop UGG Trp	U C A G	
	С	CUU CUC CUA CUG	CCU CCC CCA CCG	CAU CAC CAA CAA CAG	CGU CGC CGA CGG	U C A G	osition
	A	AUU AUC AUA AUG Met/start	ACU ACC ACA ACG	AAU AAC AAA AAA AAG	AGU AGC AGA AGA AGG	U C A G	Third position
	G	GUU GUC GUA GUG	GCU GCC GCA GCG	GAU GAC GAA GAA GAG	GGU GGC GGA GGG	U C A G	

Proteins Vary in their Biological Roles

- Functionally, proteins divided into four main categories:
- **Regulatory protein:** A protein that regulates the expression of a gene or the activity of another protein.
- **Structural protein:** A protein that forms part of a cellular structure.
- Transport protein: A protein that carries other molecules across membranes or around the body
- **Functional protein :** A protein that alters substrate

Laboratory

Tools and instrument

Laboratory tool

Types of Micropipette

Micropipette: is molecular equipment that is used to measure and transfer the small volumes of the solution by pushing it. it deals with microliter (µl)

A serological pipette is used for transferring milliliter volumes of liquid. It deals with milliliter (mL)

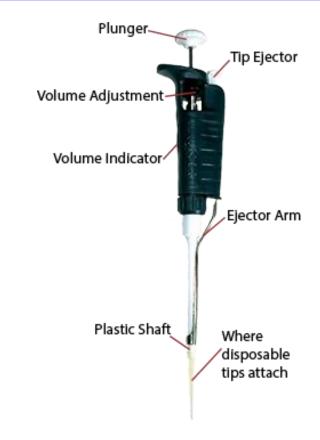
Pipette pump: provides safe and accurate pipetting and dispensing of liquids from serological pipettes. Glass or plastic pipettes are inserted into the tapered collar and the thumbwheel is rotated for precision filling or dispensing

Transfer (Teat) pipets (droppers) are disposable plastic pipets used to transfer small volumes of liquids.



How to work micropipette?

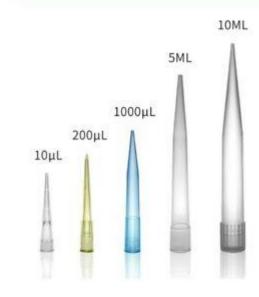
Micropipette: It has two stops. First, It can be transferred to the correct volume of solution Second, transferring too much liquid. The second stop is only used to expel the last drop of liquid from the tip.



Pipette Tip

Pipette tips are used with pipettes and pipettors to speed processing and reduce **cross-contamination**.

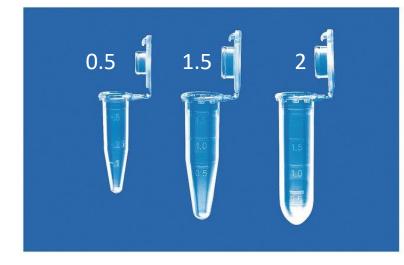
They are available in a variety of materials and styles. Popular pipette styles used are: **Universal pipette tips**, **Filter pipette tips** and **Low retention pipette tips**.



Microcentrifuge tube

Are single-use tubes made from **polypropylene** for preparing, mixing, centrifuging, transporting and storing solid and liquid samples and reagents.

The product can be used for training, routine and research laboratories in the areas of life sciences, industry or chemistry



Microcentrifuge and Vortex

Microcentrifuge: is an important piece of equipment for any laboratory, as it is used for **spinning a variety of samples at high speed**, enabling, for example, the pelleting of nucleic acids or proteins from solution, microfiltration of small aqueous samples and gathering liquid into the bottom of a tube to avoid.



Eppendorf MiniSpin Microcentrifuge, 12 × 1.5/2.0 mL

-Always Balance

- Always secure sample with lid

Vortex: is a simple device used commonly in laboratories to mix small vials of liquid



Vortex mixer