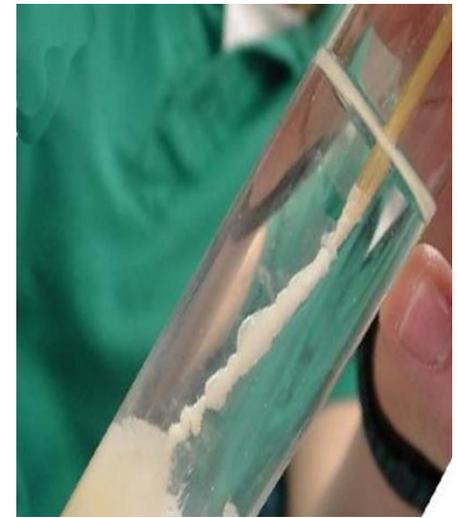


Molecular Biology

DNA Extraction

From Animal and Plant tissues

First: Opening of different cells:



a) Animal Tissues

1. Wash animal tissue with (40%) formaldehyde.
2. Use blender- glass mortar homogenizer to homogenize the tissue into tiny fragments.
3. Add (0.25%) trypsin, incubate (5 min.) at 37C.
4. Add STET buffer.
 - **Formaldehyde:** is disinfectant used to kill most of bacteria & fungi.
 - **Trypsin:** helps in damaging and opening the cell by stripping cell surface proteins and kill the cell.



b) Plant Tissues

- 1- Plant tissue must be ground into tiny fragments by used **liquid-N₂** with **metal homogenizer**.
 - 2- Add extraction solution (**CTAB**) (cetyl- trimethyl ammonium bromide)
 - **Liquid N₂:** helps in making ice crystals that play a role in scratching and opening the cell.
 - **CTAB:** as a cationic detergent makes complex with polysaccharides.
- Facilitates the separation of polysaccharides from DNA during purification process therefore it used in plant DNA extraction because of high polysaccharides content in plant tissues.



Molecular Biology

Second: Isolation of DNA from other components:

1) Add [Phenol / chloroform / Isoamylalcohol]

It is an extraction solution that used to remove proteins and other contaminants from nucleic acid sample.

- Nucleic acids are remained in **the aqueous phase**.
- Proteins are denature and separated in the **inter phase by phenol**.
- Most of lipids & polysaccharides are separated in the **lower organic phase by chloroform**.
- **Isoamylalcohol** acts as an anti-foam.

2) Centrifuge the mixture in (10000 rpm for 10 min. at 4 C). Three layers are separated; the first aqueous layer contains DNA.

Centrifugation at 4 C is **to prevent** the generation of heat through centrifugation that lead to denature the DNA and **to increase** the precipitation.

3) Transfer the upper layer to another **plastic tube** (because the DNA has ability to bind to glass tube therefore plastic tube is used), then add twice volume of **cold absolute ethanol**, which makes dehydration and pull out the water molecule from the DNA hence precipitate the DNA.

4) Mix by inversion; after 3 min., DNA will precipitate like spool DNA on a glass rod.

5) Dissolve the DNA in **by using TE buffer**.

6) Do the Electrophoresis.