DNA Experiment

OBJECTIVE: To extract DNA from different plant samples.

FUNDAMENTALS:

1st - DNA extraction requires a series of basic steps: Firstly, the cell wall and plasma membrane must be broken in order to access the nucleus of the cell. Next, the nuclear membrane must also be ruptured to release the DNA. The soaps used as dishwashing detergents emulsify the lipids in the cell membranes and break them down.

2nd - Salt prevents the joinment of proteins to DNA.

3rd - To isolate DNA, it must be precipitated in alcohol. DNA is soluble in water, but in alcohol it unwinds and precipitates at the interface between alcohol and water. In addition to allowing us to see the DNA, the alcohol separates the DNA from other cellular components, which are left in the alcohol.

Explanatory video: <u>https://youtu.be/xbnovZaX8OU</u> MATERIALS:

1.- Vegetable sample: This can be onion, tomato, banana, wheat germ or a handful of peas.

- 2.- Distilled or mineral water
- 3.- Table salt
- 4.- Dishwasher detergent
- 5.- 96 proof or very cold alcohol
- 6.- Glass rod
- 7.-. Test tube
- 8.- Blender and a knife.
- 9.- Beaker
- 10.- Pipette

PROCEDURE:

PRACTICAL DNA EXTRACTION

1.- In the small beaker pour 3 teaspoons of dishwashing detergent.

2.- Add a spoonful of salt.

3.- Add 25 millilitres of distilled water. It is necessary to use the pipette to extract the right amount of water.

4.- Keep this solution in an ice bath.

5.- Cut the central area of the chosen vegetable into squares.

6.- Grind the pieces of the vegetable (in the large beaker) with a little water in the blender (the mixture of cells and water should be opaque), operating the blades twice in 10-second pulses. This will break up many cells.

7.- In the large container, mix the cell triturate with the initial solution in the beaker.

8.- Shake the mixture vigorously for at least 5 minutes, without forming foam.

9.- Strain the liquid obtained through a sieve.

10.- Remove 5 ml of the mixture to a test tube and add 5 ml of alcohol cooled to 0° C with a pipette. The alcohol should be allowed to drip slowly down the inside of the test tube, with the tube at an inclination. The alcohol will float on top of the mixture.

11.- Leave to stand for 5 minutes until a cloudy area forms between the 2 layers.

(While waiting this time, you can collect and clean all the containers used previously).

12.- Insert a rod just below the alcohol (the cloudy area between the 2 layers).

Stir the rod back and forth and little by little the larger fragments of DNA will roll up.

13.- After one minute, remove the rod through the alcohol layer, so that the DNA will be stuck to the end of the rod.

13.- After one minute, remove the rod through the layer of alcohol and the DNA will stick to the end of the rod, looking like wet cotton wool.

RESULTS:

The filamentous product obtained from the extraction is not pure DNA, since, intermixed with it, therenare fragments of RNA.

RNA fragments are intermixed with it. A "professional" extraction is carried out by adding enzymes that fragment the RNA molecules and prevent them from binding to the DNA.

ACTIVITIES:

1.- What the purpose of exposing cells to a strong detergent is.

- 2.- Draw a picture of the action of the detergent on the cells.
- 3.- Research and briefly explain what electrophoresis consists of.