

References : ELECCRDG, ELECCRDGSG



"Restriction map and genetic screening"
Practical and theoretical objectives:

- DNA deposits in the wells of an agarose gel. Start of electrophoresis and gel staining
- Going further into the principle of DNA electrophoresis.
- Creation of a restriction map after electrophoresis (with the help of the instructions to establish the size of the fragments in base pairs).
- Identification of the one-off mutation on lambda phage DNA mutated by restriction enzyme digestion
- Going further into the principle of genetic screening.

Proposed activity:

Perform electrophoresis on 8-well agarose gel of phage lambda DNA (the products supplied are digested). Comparison of restriction fragments obtained to identify a one-off mutation on a mutated lambda phage DNA.

Composition :

To make 4 gels of 25 ml

- 1.6 g of agarose

- 100ml of TAE buffer 10 x dose for 1L
 Lambda phage, lambda phage lambda digested with EcoRI, digested with HindIII and digested twice with EcoRI and HindIII (75µL of each)
 Mutated lambda phage DNA, EcoRI digested lambda phage, HindIII digested and EcoRI and HindIII digested (75µL of each) Technical and educational instructions available on our website.

Necessary material:

DNA electrophoresis tank (for immersed gel)

70-110 V, 20my generator

Water Bath or Microwave

- Gloves
 Micropipettes from 5 to 50µl and adapted cones
 Conservation: 3 months
 DNA: in the freezer
 Colour (protected from light), TAE buffer, sterile water: in the refrigerator

Reference	Tank	Dye
ELECCRDG	standard	Azure A
ELECCRDGSG	BlueGel	SafeGreen