

Gel Electrophoresis of Agarose

Gel electrophoresis is one of the techniques scientists use to look at the DNA they have. This technique separates DNA molecules by size. First a gel is prepared. Gels are made of agarose. This consistency offers resistance to the pieces of DNA as they try to move through the gel. The gel is prepared with wells at one end so that DNA samples can be loaded into the gel. Once the DNA samples are loaded onto the gel, an electric current is applied to the gel. DNA is negatively charged due to all the phosphate groups in the backbone of DNA. Thus, DNA will move towards the positive electrode. In many analyses, polymerase chain reaction (PCR) is used to amplify specific regions of DNA that are known to vary among individuals. Apart from DNA molecules, proteins can also be run on gels. Most commonly proteins are run on gels made of polyacrylamide in the presence of SDS. In SDS PAGE (SDS polyacrylamide gel electrophoresis) all proteins are coated with SDS and are thus negatively charged.

5 x TBE Buffer stock solution

For 1 litre Buffer :

1. 54 gr Tris Base
2. 27,5 gr Boric Acid
3. 20 ml 0.5M EDTA (pH 8)
4. dH₂O to 1 litre

Mix!!!

Buffer electrophoresis :

- 5 x TBE stock solution → 0,5 x TBE Buffer
Add 900 ml dH₂O for every 100 ml 5 x TBE stock solution.
- Add 50 – 60 µl Ethidium Bromide in every litre 0.5 x TBE

Ethidium Bromide (EtBr) 10 mg/ml :

- 1gr EtBr for 100 ml dH₂O
- Stir with magnet
- Cover with a foil
- Store in a dark place at 4 °C

Gel agarose 1.5 %

- 200 ml Buffer
- 300 mg Agarose

Every PCR and NIRCA product is loaded on 1.5% gel agarose.

Gel agarose 1 %

- 200 ml Buffer
- 200 mg Agarose

Only DNA products are loaded on 1% gel agarose.

Gel agarose 2 %

- 200 ml Buffer
- 400 mg Agarose

Only FVL digestion products are loaded on 2% gel agarose.