33. **Agarose Gel Electrophoresis**

Agarose gel electrophoresis is a method used to separate DNA fragments. DNA molecules will be separated according to their sizes and charges. The location of DNA fragments within the gel can be determined directly by staining with appropriate dyes, and can be detected with naked eyes.

![Fig. 1 Gel electrophoresis kit](image)

**Procedure**

1. Prepare a buffer solution by dissolving premixed TBE powder with distilled water according to the instruction stated in the manual provided by the supplier. The prepared buffer solution can be stored under 4°C for several days.

2. Use agarose powder and prepared buffer solution to make a 1.5% agarose solution.
   - Add agarose powder into a conical flask containing buffer solution.
   - Heat the mixture in microwave oven for about 1 minute until the agarose powder is completely dissolved.
• Observe the process of heating closely to avoid boiling up of the mixture.
• A hot plate may be used instead of microwave oven.

3. Seal the lateral openings of the gel mould with cellotape properly.

4. Pour the agarose solution into the gel mould slowly.

5. Place the comb into the gel mould at one side.

6. Allow the agarose solution to solidify for about 30 minutes.

7. Remove the comb. A row of “wells” is formed in the gel.

8. Place the gel in the gel tank. The side with the wells should be placed at the cathode side of the tank.

9. Add buffer solution to immerse the gel completely.

10. Remove the sharp end of the syringe needle with a pair of scissors.

11. Use the syringe to load the marker solution into the well gently.

12. Rinse the syringe with buffer solution for several times before every loading.

13. Load the samples in the wells respectively.

14. Cover the gel tank properly with the lid.
15. Connect the electrodes and turn on the power supply. A 120V DC voltage is needed (Fig. 3).

16. Migration of DNA fragments (Fig. 4)
   - Allow the DNA fragments to move in the gel.
   - The tracking dye in the samples indicates the progress of electrophoresis.
   - When the tracking dye reaches about one third to half of the length of the gel, it is the time to collect.
   - In general, 30 minutes are long enough to finish the electrophoresis process.

17. Switch off the power supply. Take out the gel carefully.

18. Stain the gel in methylene blue solution (Fig. 5).

19. DNA bandings will become visible after staining (Fig. 6).

20. If the gel is over-stained, place it in distilled water to de-stain.

21. The gel with DNA bandings is available for analysis now. The gel can be stored under 4°C for several months.
Material

1. agarose powder
2. TBE powder provided by the supplier
3. DNA sample and marker solution provided by the supplier
4. methylene blue solution
5. gel electrophoresis unit
6. 1 ml syringe with sharp end cut
7. 250 ml conical flask
8. power pack
9. microwave oven