

Gel Electrophoresis

Running a gel electrophoresis

Estimated bench time: 135 min

Estimated total time: 3 hours

MATERIALS

- Microwave
- Agarose gel electrophoresis tray
- Autoclave tape
- Centrifuge
- Comb with large teeth
- Running chamber and power source
- Gel imager
- Blue light plate and filter glasses
- Scalpel
- Digested Inserts and Plasmid
- 6x Purple gel loading dye
- 1X TAE buffer
- Agarose
- Sybr Safe nucleic acid dye
- 10 kb DNA ladder

SETUP & PROTOCOL

Gel electrophoresis

1. Weigh 400 mg agarose (for 0.8 % TAE gel) or 500 mg agarose (for 1% TAE gel) in 250 mL Erlenmeyer flask and add 50 mL 1X TAE buffer. Carefully bring to a boil in the microwave. Keep boiling until all agarose has dissolved completely.
2. Prepare the gel trays by closing the open ends with autoclave tape. Put in a comb with large teeth at the end of the tray.
3. Add 2 µL SybrSafe dye to the agarose solution. Mix well by swirling and pour the mixture into the tray. Let the gel solidify for ~30 min.
4. Add 8 µL 6X Purple Gel Loading Dye to the digested inserts and plasmid. Mix well by carefully pipetting up and down.
5. Once the gel has solidified, take off the tape and put the gel in a running chamber with comb facing the black electrode. Fill the chamber with 1X TAE buffer until the gel is completely submerged. Carefully remove the comb.
6. Load 25 µL of the digested inserts and plasmid on the gel. Load 10 µL of the 10 kb marker in a separate slot. Furthermore, load a negative sample of the non-digested plasmid on the gel.
7. Put the cover on the chamber and attach the plugs to the power supply. Run the gel at 120 V for 45 minutes.
8. Take a picture of the gel using the gel imager.