

# UYS CEE Gel Preparation and Run.

# Protocols

# **Agarose Gel Preparation and Run**

### Materials for the gel

For 60ml of 1% w/v agarose we need

- 60ml 1X TAE (diluted from 50X TAE stock)
- 0,6gr agarose
- 2,1µl EtBr

### **Gel Preparation**

- 1. Measure the appropriate volume of 1X TAE (falcon tube for the measurement) and 0,6gr of dry agarose.
- 2. Mix TAE and agarose in a flask
- 3. Microwave for 2-3 mins (until it boils)
- 4. After microwaving, mix it thoroughly under running water (don't smell it) until it cools down.
- 5. When in the right temperature (not too hot), add 2,1µl EtBr. Pipette up and down and mix the flask to spread the EtBr everywhere.
- 6. Prepare the gel box, χτενάκια κλπ
- 7. Calibrate the scaffold until it is completely balanced (adjust the two knobs)
- 8. Add the liquid gel slowly into the gel box. Avoid the bubbles like the devil avoids incense.
- 9. Remove bubbles with a tip if needed (or make a so-called tip with paper). If you can't remove them, move them in a place where they won't affect your DNA.

- 10.Let the gel solidify for 10-15min
- 11.After 10-15mins, untighten the scaffold and remove the gel box with the gel still on
- 12.Put the gel in the "electrode box", filled with 1X TAE. If not full, add as TAE needed until it covers the gel.
- 13.Load the samples in the gel wells.
- 14.Add the lid and electrodes. Connect the other ends of the electrodes on the machine and press RUN (figure than runs) at 110V until it runs the whole gel (more than 10 mins, depends on gel agarose % and length).
- 15. Put the gel under UV and take a photo to see what you did.
- 16. Analyze the bands on the gel.
- 17.Celebrate or cry

## Sample Preparation for Gel Run

- From the miniprep stock (or whatever you want to run) stored at -20 degrees Celsius, take 2µl of plasmid.
- Add 2µl on a PCR Eppendorf tube (0.2ml)
- Add 8µl ddH<sub>2</sub>O
- Add 4µl loading dye (6X)
  Final volume will be 14µl
- After everything is in the tube, spin down (10s run at centrifuge)
- For the gel ladder, add 4μl (μικρό εργαστήριο, 4 degrees Celsius)

