

Gel Electrophoresis

1. Use the designated bench (on the right).
2. Prepare a new 1xTAE buffer: **490ml dH₂O + 10 ml 50xTAE**
3. Make 1% agarose solution in 150-200 ml flask; mix these reagents in the following order:

75 ml 1xTAE
7.5 µl SYBR green
0.75 g agarose

4. Heat to boiling in microwave (~1.5 min) swirling each 20 sec.
5. Place flask on the bench to cool down to ~56°C.
6. Prepare a small gel box (in the drawer, labeled "Lamp lab").
7. Prepare a gel tray. Attach the rubber walls, make sure they are tight. Place a comb in the desired place.
8. Pour gel solution into the gel tray. Let set about 30-60 min.
9. When ready, carefully take the comb out (one single movement), remove the rubber walls, and place the gel tray in the gel box to run the gel ("RUN TO THE RED").
10. Fill the gel box with the prepared 1x TAE solution until the gel is completely submerged (covered with the buffer for ~3-4mm).
11. Take the PCR samples, SYBR green, the ladder and the loading buffer out from the freezer. Let it thaw.
12. (If needed) Load **5 µl** of the ladder into the 1st well.
13. On parafilm, mix **1 µl SYBR green + 9 µl 6x loading buffer**
14. On parafilm, mix **1 µl SYBR/buffer mixture (from step 13) + 5 µl PCR product**
15. Load **5 µl** of each PCR sample/SYBR mixture (from step 14) into the rest of the wells.
16. Cover the gel box with the lid.
17. Plug into the power pack and run at constant voltage approx. 130V until bands have migrated to approximately 1 inch from the end (~15-20 min).
18. When it is done, turn off the power pack and remove the gel.
19. Place gel onto UV illuminator to see the results.