

Protocol for Gel electrophoresis

- >Prepare gel rig and mold. *There are four gel rigs available for each section. Thus, two groups may share a gel rig by placing the combs in appropriate spots. Use masking tapes around the mold to prevent the gel from leaking.*
- >Weigh 1.25g of agarose powder on the white weighing bowl using the weighing scale (Beside the hood)
- >Transfer into an Erlenmeyer flask (four of these are hung on the rack close to the wash sink).
- >Add 100ml of TBE buffer (under the hood) to the flask containing the agarose powder and swirl.
- >Add 10ul of the SYBR Safe to the solution and swirl.
- >Microwave every 30secs and swirl. Repeat this steps until the powder completely dissolves in the buffer.
- >Allow gel to cool by: cooling under a running tap water
keeping the flask in the refrigerator.
- >Keep eye on the gel and prevent it from solidifying. When you can touch the flask with your bare hand without enduring the pain, then you may proceed to the next step.
- >Carefully pour the gel on the mold and avoid bubbles. (Ensure the combs are appropriately placed)
- >When the gel completely solidifies, carefully remove the combs.
- >Remove the masking tapes and place the gel mold containing the gel in the rig. (*Note the appropriate orientation*).
- >Pour running buffer in the gel rig until it completely covers the surface of the gel.
- >Obtain ladder from the freezer (found on the white rack; return after use). TAs should demonstrate how to load DNA using the ladder.
- >Add 10ul of ladder for reference. Thereafter, add 10ul of DNA to the well next to the ladder. (*Students should note which well their DNA is loaded. One ladder is okay per row*).
- >Run gel for 40mins at 100volts. (*You should see bubbles coming up*). TAs may use this time to explain expected results.
- >Remove the gel and view. (Use the fluorescent filter box or the transilluminator).
- >Interpret results