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Preparation of 2% Agarose Gel for Electrophoresis

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The agarose gel consists of microscopic pores that act as a molecular sieve which separates molecules based upon the charge, size and shape. Agarose gel electrophoresis can also be used to separate other charged biomolecules such as RNA and proteins. Agarose is isolated from the seaweed genera *Gelidium* and *Gracilaria* and consists of repeated agarobiose (L- and D-galactose) subunits. The concentration of agarose in a gel depends on the sizes of the DNA fragments to be separated, with most gels ranging between 0.5%-2%.

Stephane Fadanka, Shalo Minette, Nadine Mowoh 2022. Preparation of 2% Agarose Gel for Electrophoresis. **protocols.io**
<https://protocols.io/view/preparation-of-2-agarose-gel-for-electrophoresis-b87rrzm6>



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Reagents

Agarose powder(electrophoresis grade)

1xTBE

Nucleic acid Stain(SafeView)

Materials and Equipment

Weighing boat

Electronic balance

Spatula

Electrophoretic gel tank and components(BlueGel)

Microwave

Beaker



Micropipette P-10

10ul micropipette

The entire process is generally safe but it is advisable to wear proper protective clothing before starting.




Measuring Agarose powder


22m

- 1 Accurately weigh  **0.5 g** of Agarose- electrophoresis grade (Cas 9012-36-6) into a weighing boat using a clean spatula and transfer the powder into a 50ml or 100ml beaker.
- 2 Measure  **25 mL** of 1xTBE (diluted from a 10x stock as described in the citation below)and dispense into the beaker

Nadine Mowoh, Jenny Molloy. Preparing 10x TBE Electrophoresis buffer.

<http://dx.doi.org/10.17504/protocols.io.j8nlkkok5l5r/v1>

- 3 Mix by swirling gently and transfer the beaker into the microwave at medium high to heat and boil for  **00:01:00** ^{1m}
- 4 Turn off the microwave and take out the beaker , allow it to cool for  **00:01:00** at ^{1m}
 **Room temperature** , just enough that you can conveniently hold the beaker in your palm.

- 5 Pipette  **2.5 µL** of nucleic acid gel stain (10000x) into the gel while it's still liquid and swirl gently to mix

Avoid swirling too hard as it might generate bubbles that will make your gel rough and affect migration of nucleic acid


- 6 Assemble the electrophoretic gel casting tray on a level surface and put the combs in place

- 7 Pour the gel gently into the tray to avoid bubbles

- 8 Leave gel to set for  **00:20:00** to polymerise

20m

- 9 Carefully remove the comb from the solidified gel to have the sample wells

- 10 Transfer the gel tray with the solidified gel to a horizontal electrophoretic gel tank and add about  **35 mL** of TBE buffer (depending on the size of your gel tank) to completely cover the gel. Load the nucleic acid samples into the wells and allow them run.