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DNA Gel Electrophoresis

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DISCLAIMER

ABSTRACT

Agarose gel electrophoresis can help determine the mass of a certain DNA fragment. This protocol will help user conduct gel electrophoresis appropriately.

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MATERIALS TEXT

6x Gel Loading Dye, agarose, TAE, 10000x GelRed, DNA ladders, DNA samples

DISCLAIMER:

1 Mix **1 g** agarose with **100 mL** electrophoresis buffer (1X TAE) in a flask, then heat and stir until agarose is completely dissolved.

Microwave can be used for heating.

2 Wait until the molten agarose cools down to around **40 °C** .

Temperature too high could wrap the plastic tray. Water bath can be used to control the temperature.

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3 While waiting for the cool down of molten agarose, set up and level the gel casting tray and the gel casting platform on a flat area. Inset the comb.

4 Add **10 µL** 10000X GelRed nucleic acid stain into molten agarose and mix well.

5 Pour the molten agarose onto the tray. Immediately rinse out the flask before any remaining agarose sets in it.

6 Wait at least **00:20:00** for gel solidification. 20m

6.1 While waiting for gel solidification, prepare the samples as follows:

Mixing the components:

6X gel loading dye	X µL
DNA sample/DNA ladder	5X µL

In order to receive a clearer ladder photograph, the recipe for DNA ladder with gel loading dye is recommended as follows:

6X gel loading dye	X µL
DNA ladder	X µL
Double distilled water	4X µL

7 Once the gel has completely solidified, gently remove the comb by pulling it up slowly and vertically.

8 Carefully place the tray in the gel tank. Add electrophoresis buffer (1X TAE) until covering the whole gel surface.

9 Pipette the prepared samples using a P20 (2µL-20µL) pipette to the wells.

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The samples should sink to the bottom of the wells and displace the buffer.
The amount for each load should be decided by multiple facters like DNA concentration and gel concentration. The DNA amount we use normally is from 10ng to 100ng, or 5µL to 10µL in volume.

10 Run the gels at 90 V, approximately 50 mA, for 90 min.

11 When the run is finished, carefully take the gel tray with gel out from gel tank. Photograph the gel under ultraviolet (UV) light.

UV light is harmful to skin and eyes. Avoid directly look into the light source and wear personal protective equipment. Use a gel imaging system if possible.

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