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We use this protocol and it's working

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Gel Electrophoresis

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ABSTRACT

2023 NUS-Singapore iGEM team followed this protocol for gel electrophoresis to isolate the DNA fragments from the PCR products.

MATERIALS

List of DNA Ladder Used by 2023 NUS-Singapore iGEM Team:
1. Quick-Load® Purple 1 kb DNA Ladder
2. Quick-Load® Purple Low Molecular Weight DNA Ladder

SAFETY WARNINGS

Proper lab PPE must be worn at all times.

PROTOCOL integer ID:
88415

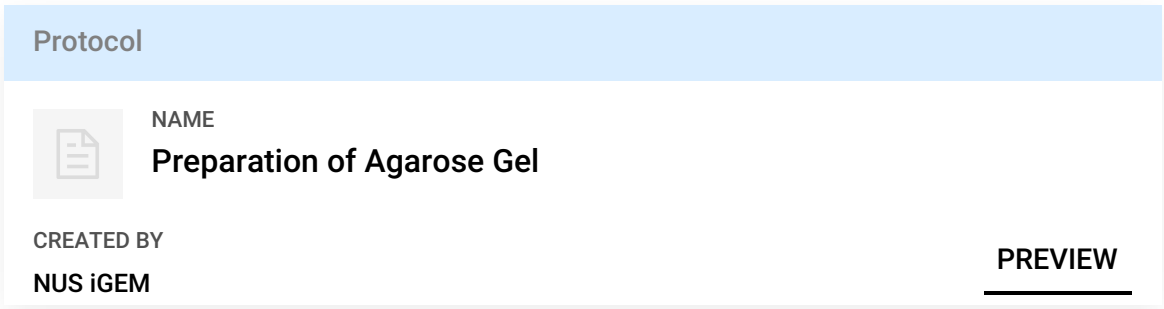
Keywords: Gel, Gel Electrophoresis

1 Set up the required apparatus:

1.1 Secure the gel tray tightly onto the gel caster.

1.2 Place an 8-well or 15-well comb into the gel tray.

2 Make an agarose gel by following the steps in the "Preparation of Agarose Gel" protocol. Usually, a 1% agarose gel is used.



2.1 For a 1% agarose gel, mix the following in a conical flask:

- 5 g of Agarose powder.
- 50 mL of 1x TAE buffer (Tris-Acetate-EDTA Buffer).

2.2 Swirl the agarose solution to mix it well.

2.3 Heat the agarose solution in a microwave until it boils.

2.4 Take out the conical flask from the microwave and swirl the conical flask until the agarose solution is clear and without undissolved agarose powder or lumps.

> **Safety information**
>
> Wear thermal gloves when handling the hot conical flask from the microwave to prevent burns.

2.5 Add 5 µL of Ultra GelRed gel stain into the agarose solution and swirl the conical flask until the colour becomes uniform.

2.6 Secure a gel tray tightly onto a gel caster and place an 8-well comb or 15-well comb into the tray.

2.7 Pour the agarose solution into the tray, ensuring that there are no bubbles.

2.8 Cool down the agarose solution for at least 00:30:00 to get a solidified agarose gel. 30m

3 After the agarose gel has solidified in the gel tray, remove the gel tray from the gel caster and place it in a buffer tank with pre-filled 1x TAE buffer.

4 Add the correct DNA ladder and the sample(s) into the wells.

5 Close the buffer tank with the tank lid, ensuring that the colour of the electrodes on the tank lid is the same as the one on the buffer tank.

6 Run the gel electrophoresis at 110V for 40 minutes, making sure that bubbling occurs when the gel electrophoresis begins.

7 Upon finishing gel electrophoresis, put the agarose gel onto a UV Sample Tray, next, put it into the Gel Doc EZ System, and then run the gel imaging program on the desktop.

8 Save the gel image.