



Description

BacPlaque Agarose

30 g

70034-3

Novagen's BacPlaque Agarose is pre-qualified to ensure consistent agarose overlay performance for plaque assays and is suitable for preparative gel electrophoresis. Many commercial "DNA grade" agaroses contain contaminants that are toxic to insect cells or lack the physical characteristics required to obtain good overlays. BacPlaque Agarose is optimized for consistent plaque formation in the BacVector® and TriEx™ Systems with Novagen's Sf9 Insect Cells.

Preparation of a 3% BacPlaque Agarose Stock

For plaque assays it is convenient to prepare sterile stocks of 3% w/v BacPlaque Agarose in water.

1. For a 20 ml stock, add 0.6g agarose to 20 ml of sterile deionized water in a 100 ml Pyrex media bottle, cap loosely and autoclave.
2. Mix well as soon as the autoclave cycle has finished (being careful to avoid splashing of the hot liquid). Allow to cool, tighten the cap and store at room temperature.

Using the 3% BacPlaque Agarose Stock

For plaque assay protocols that use BacPlaque Agarose, see Technical Bulletin 216.

1. Loosen the cap of the 20 ml 3% BacPlaque™ Agarose stock and microwave until the agarose is completely melted and there are no remaining lumps.

Note:

The agarose stock should contain no unmelted agarose.

2. Place the bottle in a 37°C water bath.
3. Cool the 20 ml stock to less than 50°C and add prewarmed (37°C) BacVector Insect Cell Medium containing 5% v/v fetal bovine serum to a total volume of 60 ml. Use gentle swirling to mix. The final agarose concentration is 1% (w/v) agarose at 2/3 strength medium containing serum. Although antibiotics can be added for plaque assays they must be avoided during Direct Plaquing Transfection (see Technical Bulletin 216).
4. The overlay solution can be used immediately or kept at 37°C for at least 1 day without solidifying.

Preparative Gel Electrophoresis

Due to its excellent physical properties, absence of nucleases and other contaminants that interfere with ligation, and low electroendosmosis rating, BacPlaque Agarose is qualified for use as a low melting point agarose for electrophoresis and recovery of DNA from the gel matrix. BacPlaque Agarose has a low melting point of 65°C and solidifying temperature of < 37°C. Gels will solidify faster when incubated at 4°C. Gels can be run at room temperature, but care should be taken to avoid excess heating during electrophoresis. Use standard protocols for recovery of DNA from low melting point agarose gels.

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