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Making tetracycline LB agar plates

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We use this protocol and it's working

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Abstract

Making LB agar plates containing tetracycline for selecting strains carrying the pJC8 cosmid. Plates can be stored at 4 °C wrapped in aluminium foil for up to a month.



Making tetracycline LB agar plates

- 1 Autoclave or microwave LB agar from media room

Note

Contents of LB Agar:

37 g pre-mixed powder consisting of:

- 5 g peptone
 - 10 g peptone from casein
 - 10 g sodium chloride
 - 10 g agar-agar
- 1 L Sterile H₂O

- 2 Make up 15 mg/ml tetracycline hydrochloride in sterile water
 - 2.1 Retrieve tetracycline hydrochloride from -20 °C freezer
 - 2.2 Weigh an empty Eppendorf
 - 2.3 In the fume hood, add tetracycline hydrochloride to the Eppendorf
 - 2.4 Weigh the Eppendorf, and calculate the difference to determine the weight (mg) of compound
 - 2.5 To calculate sterile water (uL) to add = (compound mg / 15 mg/mL) x 1000 uL
- 3 Let LB agar cool to 55 °C
- 4 Dilute tetracycline hydrochloride 1/1000 dilution in LB agar (final conc: 15 ug/mL).
- 5 Using a serological pipette, dispense 35 mL agar into 90 mm Petri dishes. Leave to set.

