

LB (Lysogeny Broth) Recipe

a common nutrient-rich media for growing bacteria

Ingredients:

- 10g Tryptone
- 5g yeast extract (available at natural food stores)
- 50g (for 5%) up to 200g (for 20%) NaCl (can substitute non-iodized or sea salt)
- 15g MoorAgar Inc. Gelidium agar
- Purified H₂O - Bring the volume up to 1L
- Optional: some recipes suggest adjusting the pH to 7.0-7.5 with N NaOH

Materials/Equipment/Supplies:

- Erlenmeyer Flask, Beaker, Glass container that will hold at least twice the volume of your media
- Graduated Cylinder for measuring water
- Hot Plate, Stove top, Microwave...
- Foil for covering media, Plastic wrap if using a microwave
- Sterile Petri Dishes (plastic or glass)*
- Heat Resistant Hand Protection
- 10% Bleach or wipes for cleaning work area
- Optional: Glass rod for stirring hot liquid
- Scale for weighing solid ingredients
- Tools for handling solid ingredients - spoons, scoops, weigh boats, etc.

* Note: keep sterile petri dishes closed until ready to pour the agar into them

Procedure:

1. Measure recipe ingredients; agar and purified water into clean flask, beaker, or other glass container.
2. Cover container with foil or plastic wrap if using a microwave.
3. While wearing heat resistant hand protection, hold the flask or beaker over the flame. Swish or stir the mixture constantly while heating. Or place on a hot plate or in a microwave.
4. Boil the mixture until the agar is completely dissolved -in media with 15% salt dissolve may be slow -15+ minutes. Remove from heat. (Note: incompletely dissolved agar will leave the media fragile or squishy)
5. Swirl the media vigorously to mix.
6. Cool the media until it is just cool enough to handle, about 20 to 30 minutes You should be able to hold your hand against the container reasonably comfortably for a few seconds. (If the media is too cool, it will start to solidify in the container. If it is too hot, it will leave excess condensation on the lids.)
7. Pour enough melted agar into each sterile petri dish to cover the bottom - about 1/8" to 1/4" deep. Replace the lid immediately.
8. Place agar plates on a counter top to cool and set. Agar medium will set like stiff gelatin at room temperature.
9. Once cool - the agar medium is ready for storage or use.

Storage:

Stack agar plates upside down in the refrigerator. Do Not Freeze! The purpose of placing the plates upside down is to prevent condensation from dripping down onto the agar surface which could then facilitate movement of organisms between colonies. Plates will keep refrigerated for 4-6 weeks.