## Overview and Considerations:

**Note 1:** ~25mls/plate.

## Reagents

1. Tryptone
2. Yeast Extract
3. Sodium Chloride (NaCl)
4. Bacto-Agar
5. Kanamycin or Amphicillan
6. Water

## Protocol

Adjust weights according to volume needed.

1. Make LB media in a 1 L volumetric flask by adding:
   * 10 g tryptone,
   * 5 g yeast extract,
   * 10 g NaCl,
   * and 15 g bacto-agar if you are making plates
   * 800 ml water.
2. Check the pH is 7. Bring the volume up to 1 L.
3. Autoclave at 45oC for 30 mins (Setting 4). Also autoclave a stirring bar in foil.
4. For LB Media
   * Cool media
   * Close up lids
   * Store at 4oC until required
5. For plates
   * Store at 55oC (this will cool the media, but not let it set)
   * Slide the stir bar in gently
   * Add either kanamycin sulfate (25 ug/ml final concentration) or ampicillan (100 ug/ml final concentration) to LB-Agar.
6. Pour solution into 15 mm petri dishes, just enough to cover the surface area and let stand at room temperature (with the lid on) for ~20 mins or until the agar is set.
7. Draw a single vertical line for plates containing ampicillan and two vertical lines for plates containing kanamycin.
8. Store at 4oC for up to a month.