Luria-Bertani (LB) media liquid and agar

Materials

- NaCl;
- BactoTM tryptone;
- yeast extract;
- agar (only for for plating agar medium);
- ddH2O;
- 5 M NaOH;
- 1000x antibiotic of choice

Procedure

1. Add the following to a 1 L (non-sterile) bottle
2. NaCl to a final concentration of 0.17 M.
3. 1% (w/v) BactoTM tryptone.
4. 0.5% (w/v) yeast extract.
5. ddH2O to 600 mL.
6. 100 µL of 5 M NaOH (adjusts the pH to ~7.0).
7. Add 9 g agar. N.B! If you are not plating the medium, skip this step.
8. Shake. It is unnecessary to dissolve all solids now because autoclaving will do this.
9. Autoclave for 20 min within 2 hr. N.B! If you are not plating the medium, do this step and then store at room temperature. In that case, the protocol ends here.
10. Let it cool to ~40–50°C (touchable, so the antibiotics will not be destroyed by the high temperature).
11. Add 600 µL of 1000x antibiotic of choice (if any) and gently swirl the bottle to mix. Do not shake the bottle vigorously as this will create many bubbles that will be transferred to your plates!
12. Pour into empty petri dishes just enough to cover the surface (~20 mL per plate). Tip: you don't have to cover the whole surface when pouring - pour a little and swirl the plate to cover the whole surface. If bubbles remain in the plates, heat the plate surface carefully with a burner to burst them. But make sure not to heat the solution in the plate too much since it might degrade the antibiotic.
13. Leave the plates at room temperature to solidify (~1 hr).

14. Solidified plates should be turned upside down for a few hours at room temperature, then stored at 4°C.

References

Liljeruhm, J., Gullberg, E., & Forster, A. C. (2014). *Synthetic biology: a lab manual*.