00_07 Preparation of LB Medium

MITTWOCH, 14.7.2021

Goal-Setting

- Preparation of LB-medium / LB-agar

Terms / abbreviations

- ddH_2O = MilliQ water
- LB = Lysogeny broth

Risk areas

- None

Required materials and / or information

- Chemicals:
  - Bacto Trypton, BD Biosciences
  - MilliQ water, Sartorius arium pro VF
  - Sodium Chloride, Carl Roth
  - Yeast Extract, Carl Roth
- Materials:
  - Schottflask(s)
  - Stirring fish

Templates, devices, software

- Analysis balance, Kern ABJ 220-4NM
- pH meter, Knick Digital pH-Meter 646

Preliminary work

- Calculate the needed masses for specific volumina

Operation

- pH meter (ready to use in the chemical room)
  - If it is used for the first time, ask an experienced person around how to use it
  - Regularly calibrate the pH meter according to manufacturer
  - User guide:
    - pH meter should measure about 6.5 when the solution is stored (small Falcon Tube)
    - Take it out and unlock it (small switch at the top)
    - Wash it carefully with some MilliQ water
    - Hold into the solution and measure
    - Wash again after use, turn off the switch and put it back into the small Falcon Tube

<u>The following recipe is standardized to 1 L</u>

1. Weigh the following components into a schottflask
   a. Bacto Trypton: 10 g
   b. Sodium Chloride: 5 g
   c. Yeast Extract: 5 g
2. Fill up to 1 L with ddH_2O

3. Mix by stirring/shaking/inverting
4. Use NaOH to adjust pH to 7
5. Add 15 g Agar (optional; if plates should be casted)
6. Mix by stirring/shaking/inverting
7. Autoclave
    a. Note: Do not autoclave full bottles! Fill 500 mL bottles only up to 400 mL and 1 L bottles only up to 800 mL!
8. Store at 60 °C or directly cast plates (only with agar)
9. Store liquid medium at RT or in fridge (never open bottle in unsterile environment!)

Disposal

- Dispose the solid agar in S1 waste

Troubleshooting

- None

Follow-up work

- 05_01 Casting Agar Plates