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Lysogeny Broth (LB) medium V.1

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Protocol status: Working

I use this protocol and it's working

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Abstract

Lysogeny broth (LB) is a nutritionally rich medium which is primarily used for the growth of bacteria^[1]. LB broth is commonly used when cultivating Escherichia coli. There exist different formulations of LB and lead to the development of derivations for specialized use.

Guidelines

Follow step by step, unless stated otherwise. Equipment needed should be standard to a microbiology lab.

Materials

Analytical scale, autoclave, bottle, weight vessel, LAF bench

Protocol materials

☒ Sodium chloride **Merck MilliporeSigma (Sigma-Aldrich) Catalog #S9625** In [2 steps](#)

☒ Tryptone **Merck Millipore (EMD Millipore) Catalog #T9410** In [2 steps](#)

☒ Yeast Extract **Merck MilliporeSigma (Sigma-Aldrich) Catalog #Y0875** In [2 steps](#)

☒ Agar **Merck MilliporeSigma (Sigma-Aldrich) Catalog #A1296** Step 2.2

Safety warnings





⚠ When removing autoclaved components, be sure to take care as this can be very hot. If using antibiotics, use sufficient PPE to protect yourself, as some can be toxic to humans.

Before start

Prepare glassware by cleaning it, and ensure that scale is sufficiently calibrated



500 mL LB-Lennox (broth) medium

- 1 All compounds are measured using a high precision analytical scale from powdered compounds. Each compound is measured to within 1% of the target weight. All compounds are mixed in a Duran bottle
- 1.1 Fill the bottle with  400 mL double-distilled water
- 1.2 Measure  5000 mg Tryptone,  2500 mg Yeast extract and  2500 mg Sodium chloride


Powdered compounds:

 Tryptone **Emd Millipore Catalog #T9410**


 Yeast Extract **Emd Millipore Catalog #Y0875**


 Sodium chloride **Emd Millipore Catalog #S9625**

- 1.3 Add powdered solids into bottle, and use a magnetic mixer with a stir bar to mix for

 00:05:00

5m

- 1.4 Adjust pH while mixing to  6.7 using concentrated sodium hydroxide

- 1.5 Add distilled water to a total of  500 mL

- 1.6 Autoclave liquid at  121 °C for  00:15:00


15m





Note

Cool to 50°C and supplement with antibiotics as appropriate

500 mL LB-Lennox (agar) medium

20m

- 2 All compounds are measured using a high precision analytical scale from powdered compounds. Each compound is measured to within 1% of the target weight. All compounds are mixed in a Duran bottle
- 2.1 Fill the bottle with  400 mL double-distilled water

2.2 Measure  5000 mg Tryptone,  2500 mg Yeast extract,  2500 mg Sodium chloride and  7500 mg agar

Powdered compounds:


 Tryptone **Emd Millipore Catalog #T9410**

 Yeast Extract **Emd Millipore Catalog #Y0875**

 Sodium chloride **Emd Millipore Catalog #S9625**

 Agar **Emd Millipore Catalog #A1296**

2.3 Add powdered solids into bottle, and use a magnetic mixer with a stir bar to mix for

 00:05:00

5m

2.4 Adjust pH while mixing to  6.7 using concentrated sodium hydroxide

2.5 Add distilled water to a total of 500 mL

2.6 Autoclave liquid at  121 °C for  00:15:00

15m

Note

Cool to 50°C and supplement with antibiotics as appropriate

Agar can be stored, then reheated to 50°C to be poured