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Making glycerol stocks of bacteria

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Protocol status: Working

We use this protocol and it's working

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Abstract

Freezing down a bacterial culture for long-term storage.

Materials

MATERIALS

- ✕ Liquid LB medium
- ✕ DEPC water
- ✕ Glycerol 99 % Certified AR for Analysis Fisher Chemical **Fisher Scientific Catalog #G/0650/17**
- ✕ b-broth
- ✕ fresh bacterial culture
- ✕ cryovial
- ✕ -80 freezer

Troubleshooting

Safety warnings

- ⚠ Depending on the kind of bacteria you are working with, you will have to adapt your working conditions.

Before start

Make sure you have a fresh bacterial culture.



- 1 Make sure you have autoclaved glycerol, cryovials and a fresh bacterial culture in liquid b-broth or LB. If you don't have a fresh bacterial culture, you can grow them in b-broth or LB medium at 180 rpm in flasks. 5m
- 2 Prepare a sterile 50% glycerol/50% DEPC water solution. 5m
- 3 Add 1 ml 50/50 solution to as many cryovials as you want, e.g. 6 5m
- 4 Add 1 ml of bacterial culture to the cryovials, close them with the cap and shake them. 5m
- 5 Store the cryovials in the -80 ° freezer. 2m