

## Making bacterial glycerol stocks

### Overview

Whenever you successfully transform a bacterial culture with a plasmid or whenever you obtain a new bacterial strain it is a good idea to make a long-term stock of that bacteria. Bacteria can be stored for years if they are stored at  $-80^{\circ}\text{C}$  and in a high percentage of glycerol.

### Materials

Overnight bacterial culture

Cryogenic vials

50% Glycerol solution

**Note:** Make the 50% glycerol solution by diluting 100% glycerol in dH<sub>2</sub>O.

### Procedure

1. Inoculate liquid LB culture with a single colony and incubate overnight.
2. Add 500  $\mu\text{L}$  of the overnight culture to 500 $\mu\text{L}$  of 50% glycerol (sterile) in a 2 mL screw top tube or cryovial and gently mix by vortexing or pipetting.

**Note:** Snap top tubes are not recommended as they can open unexpectedly at  $-80^{\circ}\text{C}$ .

3. Write on the vial all relevant information - vector, strain, date, researcher, etc.
4. Freeze the glycerol stock tube at  $-80^{\circ}\text{C}$ . The stock is now stable for years, as long as it is kept at  $-80^{\circ}\text{C}$ . Subsequent freeze and thaw cycles reduce shelf life.

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