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# 🌐 Creating Bacterial Glycerol Stocks for Long-term Storage

🔗 Forked from [Creating Bacterial Glycerol Stocks for Long-term Storage](#)

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Behavioural Genomics



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**Protocol status:** Working  
We use this protocol and it's working

**Created:** Mar 04, 2024

## ABSTRACT

Bacterial glycerol stocks are important for long-term storage. Bacteria on LB agar plate can be stored at 4°C for a few weeks. However, if you want to store bacteria for a longer time, you will need to establish glycerol stocks. The addition of glycerol stabilizes the frozen bacteria, preventing damage to the cell membranes and keeping the cells alive. A glycerol stock of bacteria can be stored stably at -80°C for many years.

## SAFETY WARNINGS



- Snap top tubes are not recommended as they can open unexpectedly at -80°C
- Be sure to label both the lid and the tube of a glycerol stock before you place the sample at -80°C. Frozen tubes are hard to write on and samples stored for long periods at -80°C can lose labels stuck to tube!
- Try not to freeze/thaw your glycerol stock too many times as this will reduce the shelf life. Placing the glycerol stock on dry ice while streaking onto LB agar will prevent it from thawing completely.

## BEFORE START INSTRUCTIONS

Make the 50% glycerol solution by diluting 100% glycerol in dH2O and autoclaving.

## Steps

- 1 Grow an overnight culture of the selected strain

### Protocol



NAME

### Inoculating a Liquid Bacterial Culture

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Priota Islam

PREVIEW

- 2 Add 500  $\mu$ L of the overnight culture to 500  $\mu$ L of 50% glycerol in a 2 mL screw top tube or cryotube and gently mix

- 3 Shake the glycerol 5-6 times before freezing.


### Expected result

One uniform solution, no layers present

- 4 Freeze the glycerol stock tube at -80°C.

- 5 To recover bacteria from your glycerol stock, open the tube and use a sterile loop or pipette tip to scrape some of the frozen bacteria off of the top. Do not let the glycerol stock thaw!

Protocol



NAME

Streaking and Isolating Bacteria on a LB Agar Plate

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Priota Islam

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