

Glycerol Stocks for Long-term Storage

Aim

Creating Glycerol Stocks with the goal to store and freeze bacterial or DNA samples for long-term Storage.

Procedure

1. Follow the steps for Inoculating an Overnight Liquid Culture.
2. After you have bacterial growth, add 500 μL of the overnight culture to 500 μL of 50% glycerol in a 2 mL screw top tube or cryovial and gently mix.
3. Note: Make the 50% glycerol solution by diluting 100% glycerol in dH₂O.
4. Note: Snap top tubes are not recommended as they can open unexpectedly at -80°C .
5. Freeze the glycerol stock tube at -80°C . The stock is now stable for years, as long as it is kept at -80°C . Subsequent freeze and thaw cycles reduce shelf life.
6. To recover bacteria from your glycerol stock, open the tube and use a sterile loop, toothpick or pipette tip to scrape some of the frozen bacteria off of the top. Do not let the glycerol stock unthaw! Streak the bacteria onto an LB agar plate.
7. Grow your bacteria overnight at the appropriate temperature.
8. The next day you will be able to start an overnight culture for plasmid
9. DNA prep the following day.

Lab protocol

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