

Quick Start Guide

GenElute™ HP Endotoxin-Free Plasmid Maxiprep Kit

NA0300S, NA0300, NA0310

Reagents to Prepare

- Spin RNase A Solution tube briefly and add 750 µL (4 and 10 preps) or 1.9 mL (25 preps) of RNase A Solution to the Resuspension Solution. Store at 4 °C.
- Add 120 mL (4 and 10 preps) or 300 mL (25 preps) of 95-100% ethanol to Wash Solution 2. After each use, tightly cap the diluted Wash Solution to prevent ethanol evaporation.

Protocol

Vacuum format.

Harvest Bacteria

1. Pellet cells from 150 mL overnight culture by spinning for 10 min at 5,000 x g. Discard supernatant.

Resuspend and Lyse Bacteria

2. Resuspend cells in 12 mL Resuspension Solution. Pipet or vortex to mix.
3. Add 12 mL Lysis Solution. Gently invert 6-8 times to mix. Do not vortex. Allow to clear for 3-5 min.

Prepare Cleared Lysate

4. Remove plunger from Filter Syringe and place the barrel in an upright position.
5. Add 12 mL Neutralization Solution (chilled prior to use) to lysed cells and gently invert 6-8 times.
6. Add 9 mL Binding Solution and gently invert 1-2 times. Immediately add the mixture to the barrel of the filter syringe and let sit for 5 min.

Prepare Column

7. Place Column onto vacuum manifold and apply vacuum.
8. Add 4 mL Column Preparation Solution to the column and allow it to pass through.

Bind DNA to Column

9. Hold Filter Syringe over the column and gently insert the plunger to expel the cleared lysate. Allow the lysate to pass through the column.

Wash to Remove Contaminants

10. Add 12 mL Wash Solution 1 to column and allow it to pass through.
11. Add 12 mL Wash Solution 2 to column and allow it to pass through.
12. Maintain vacuum until the column is dry (10 min). If more than 6 columns are on the manifold, dry for at least 20 min.

Elute Purified DNA

13. Transfer column to a collection tube.
14. Add 3 mL Elution Solution to the column.
15. Spin in a swinging bucket rotor at 3,000 x g for 5 min for maximum plasmid DNA yield or 1,000 x g for 5 min for maximum plasmid DNA concentration.

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