Quick Start Guide

GenElute™ Bacterial Genomic DNA Kit

NA2100, NA2110, NA2120

Reagents to prepare:

- Dilute Wash Solution Concentrate with 10 mL (10 prep), 80 mL (70 prep) or 360 mL (350 prep) of 95-100% ethanol. After each use, tightly cap the diluted Wash Solution Concentrate to prevent ethanol evaporation.
- Prepare Lysozyme Solution (sold separately L4919):
 Prepare 2.115x10⁶ unit/mL Lysozyme Solution using the Gram-Positive Lysis Buffer as the diluent.
- Dissolve Proteinase K in water to obtain a 20 mg/mL stock solution. This solution can be stored for several days at 2–8 °C. For longer-term storage, the unused portion of the solution may be stored in aliquots at –20 °C until needed.

NOTE: All spins at $\ge 12,000 \times g$

1. Harvest & Lyse Cells

 Pellet 1.5 mL of bacterial culture by spinning for 2 min.

For Gram-Negative Bacteria:

- Resuspend pellet in 180 μL Lysis Solution T.
- Add 20 μ L Proteinase K solution to cell suspension. Vortex or pipet to mix. Incubate at 55 °C for 30 min.
- Add 200 μL Lysis Solution C, vortex or pipet to mix.
 Incubate at 55 °C for 10 min.

For Gram-Positive Bacteria:

- \bullet Resuspend pellet in 200 μL Lysozyme Solution and incubate at 37 °C for 30 min.
- Add 20 μL Proteinase K and 200 μL Lysis Solution C to cell suspension. Vortex or pipet to mix. Incubate at 55 °C for 30 min.

2. Prepare Column

 Add 500 µL of Column Preparation Solution to each Column with Collection Tube. Spin for 1 min and discard flowthrough.

3. Bind DNA to Column

- Add 200 µL ethanol to the lysed cells, vortex or invert to mix.
- Transfer EtOH mixture to Column and spin for 1 min.

4. Wash Column

- Transfer column to new Collection Tube and add 500 μL Wash Solution 1 to Column. Spin for 1 min and discard flowthrough.
- Add 500 µL Previously Diluted Wash Solution Concentrate to Column.
 Spin for 3 min to dry Column.

5. Elute DNA

 Transfer column to new Collection Tube and add 200 µL Elution Solution. Spin for 1 min. Repeat elution if desired.

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