Bacterial Genomic DNA Purification Kit

Product	Catalog #	Purifications
Bacterial Genomic DNA Purification Kit	85171	50

Description

The Bacterial Genomic DNA Purification Kit provides for rapid isolation of genomic DNA from gram-positive and gram-negative bacteria utilizing one simple protocol. In just 30 minutes you can extract high quality genomic DNA from bacterial cultures. Specially formulated Advamax 2 Beads ensure that the kit efficiently removes protein and other cellular contaminants from genomic DNA.

Benefits

- High Quality DNA – DNA is suitable for most molecular biology downstream applications
- Reproducible DNA Purification - Consistently provides high quality DNA
- Fast Processing – Only 30 minutes from start to finish
- High DNA Yield – Average yield is 40µg of DNA from a 2 ml culture

Kit Components	Part No.	Amount
Spheroplast Buffer	4050028	25 ml
Lysis 1	4050031	6 ml
Lysis 2	4050030	6 ml
Advamax 2 Beads	4050237	6 ml
Extraction Buffer	4050029	6 ml

Equipment and Materials Required

1. Microcentrifuge capable of reaching ≥10,000 x *g*
2. 2-ml tube
3. Isopropanol
4. 70% ethanol

Storage and Stability Conditions

Spheroplast Buffer must be stored at -20ºC and thawed before use. The other components of the Bacterial Genomic DNA Purification Kit should be stored at 4ºC. The kit is stable for up to one year under these conditions.

Quality Control

E. coli strain EB5 Alpha was cultured in Terrific Broth overnight at 37°C. Two milliliters of the culture were removed and genomic DNA was isolated according to the recommended protocol. The final product was analyzed for purity and integrity by agarose gel electrophoresis.

Recommended Protocol

1. Centrifuge 2 ml of bacterial culture with an OD_{600} value between 2.0 and 3.0 to obtain pellet. Discard supernatant.
2. Add 400 µl of Spheroplast Buffer and vortex at the highest speed to resuspend pellet.
3. Incubate 10 minutes at 37ºC.
4. Add 100 µl of Lysis 1.
5. Add 100 µl of Lysis 2, mix and incubate 5 minutes at 65ºC.
6. Add 100 µl of Extraction Buffer.
7. Shake 2–3 minutes or vortex for 10 seconds.
8. Centrifuge in a microcentrifuge at the highest speed for 3 minutes.
9. Add 100 µl of Advamax 2 Beads.
10. Gently invert 10 times to mix.
11. Centrifuge in a microcentrifuge at the highest speed for 3 minutes.
12. Transfer supernatant to a clean 2-ml tube.
13. Add an equal volume of isopropanol and invert several times to mix.
14. Pellet DNA by centrifugation at the highest speed for 2 minutes.
15. Carefully remove supernatant by decanting or with a pipet without disturbing the DNA pellet.
16. Wash DNA by adding 750 µl of 70% ethanol, invert 2-3 times and centrifuge at the highest speed for 2 minutes.
17. Carefully remove supernatant by decanting or with a pipet without disturbing the DNA pellet.
18. Allow samples to air dry for 20-30 minutes or until no ethanol is left.
19. Re-suspend DNA in 100 µl of 10 mM Tris·HCl or dH_2O.