

## Product information

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# Rapid Bacteria Genomic DNA Isolation Kit

**Catalog #:** BS8225  
**Size:** 50 preps  
**Storage:** 18 to 25°C (1 year)

### Product Description:

The kit is designed for rapid small-scale extraction of high quality genomic DNA from a variety of Gram-negative or Gram-positive bacteria. Purified DNA can be used for many downstream applications such as PCR, restriction enzyme digestion, hybridization and other applications.

### Contents:

Components	BS8225 50 Preps
Universal Digestion Buffer	25 ml
Buffer PB	12 ml
TE Buffer	10 ml
Protocol	1

### Features:

1. Rapid and Simple.
2. High Quality of DNA. OD260/OD280 of purified DNA is generally 1.8~1.9.
3. No Toxic Substance. The kit does not contain toxic reagents.
4. Easy to Scale Up.

### Procedures:

#### 1. Sample Preparation.

##### A. Gram-negative bacterias (E. coli, streptococcus, pneumococcus, etc.).

- a. Transfer 1 ml overnight culture (about  $2 \times 10^9$  cells) into a centrifuge tube and centrifuge at 10,000 x g for 30 seconds, discard supernatant.
- b. Add 400  $\mu$ l Universal Buffer Digestion into the pellets, vortex and incubate at 65 °C until cells are lysed thoroughly.

**Note 1:** Usually incubation is 30~60 minutes. If RNA-free DNA is needed, add 20  $\mu$ l RNase A (20 mg/ml; **NOT supplied in the kit**) and incubate at room temperature for 5 minutes before step 3.

**Note 2:** Buffer Digestion may form precipitates during long-term storage. Warm the bottle at 65 °C to dissolve the precipitates.

## Procedures (contd.):

### B. Gram-positive bacteria (staphylococcus, Corynebacterium diphtheriae, etc.).

- a. Transfer 1 ml overnight culture (about  $2 \times 10^9$  cells) into a centrifuge tube and centrifuge at 10,000 x g for 30 seconds, discard supernatant.
- b. Add 180  $\mu$ l lysozyme solutions (20 mg/ml lysozyme, 20 mM Tris-HCl, pH 8.0, 2.5 mM EDTA, 1% Triton X-100; **NOT supplied in the kit**). Mix thoroughly and incubate at 37 °C for 30-60 minutes. Centrifuge at 10,000 x g for 1 minute, discard the supernatant.
- c. Add 400  $\mu$ l Universal Buffer Digestion into the pellets, vortex and incubate at 65 °C until cells are lysed thoroughly.

**Note 1:** Usually incubation is 30~60 minutes. If RNA-free DNA is needed, add 20  $\mu$ l RNase A (20 mg/ml. NOT supplied in the kit) and incubate at room temperature for 5 minutes before step 3.

**Note 2:** Buffer Digestion may form precipitates during long-term storage, warm the bottle at 65 °C to dissolve the pellet.

3. Add 200  $\mu$ l Buffer PB, mix by inverting. Incubate at -20 °C for 5 minutes.
4. Centrifuge at 12,000 x g for 5 minutes at room temperature. Transfer the supernatant to a new 1.5 ml tube.
5. (*Optional*) Add 0.2 ml of chloroform to the supernatant, mix well by inverting 10 times. Centrifuge at 12,000 x g for 2 minutes. Carefully transfer the supernatant to a clean 1.5 ml tube.
6. Add equal volume of isopropanol (approx 0.3~0.5 ml) to the solution, mix well by inverting 5 times. Incubate at room temperature for 2~5 minutes. Centrifuge at 12,000 x g for 5 minutes, discard the supernatant carefully.
7. Add 1 ml of pre-cooled 75% ethanol to the pellet, mix well by inverting 10 times. Centrifuge at 12,000 x g for 1 minute, discard the supernatant.
8. Repeat Step 7.
9. Air-dry the pellet at room temperature with the lid open for 2~5 minutes.

**Note:** Don't over dry.

10. Add 50~200  $\mu$ l of TE (10 mmol/L Tris, 1 mmol/L EDTA, pH 8.0) buffer to dissolve DNA pellet. Keep at 4 °C for a couple hours until DNA pellet is completely dissolved. The purified DNA is ready for use. Or keep at -20 °C for long term storage.



PRODUCTS ARE INTENDED FOR BASIC SCIENTIFIC RESEARCH ONLY.  
NOT INTENDED FOR HUMAN OR ANIMAL USE.