TOOLS Bacterial and Fungal DNA and RNA Extraction Kit

Simple centrifugation procedures for Bacterial and Fungal DNA & RNA extraction and purification

Contents

Introduction

The TOOLS Bacterial and Fungal DNA and RNA Extraction Kit is a buffer-based system for bacterial and fungal DNA and RNA extraction and purification. The kit involves simple centrifugation procedures that facilitate the complete removal of contaminants and enzyme inhibitors. It affords rapid, simple, and cost-effective extraction and purification processes, and the purified DNA and RNA are suitable for downstream applications, such as polymerase chain reaction, blotting, and sequencing.

Materials not provided

1. 1-Bromo-3-chloropropane (BCP) (CAS Number: 109-70-6).
2. TOOLS Proteinase K (Cat No. RTT-BD03) or other compatible reagents and lysozymes.

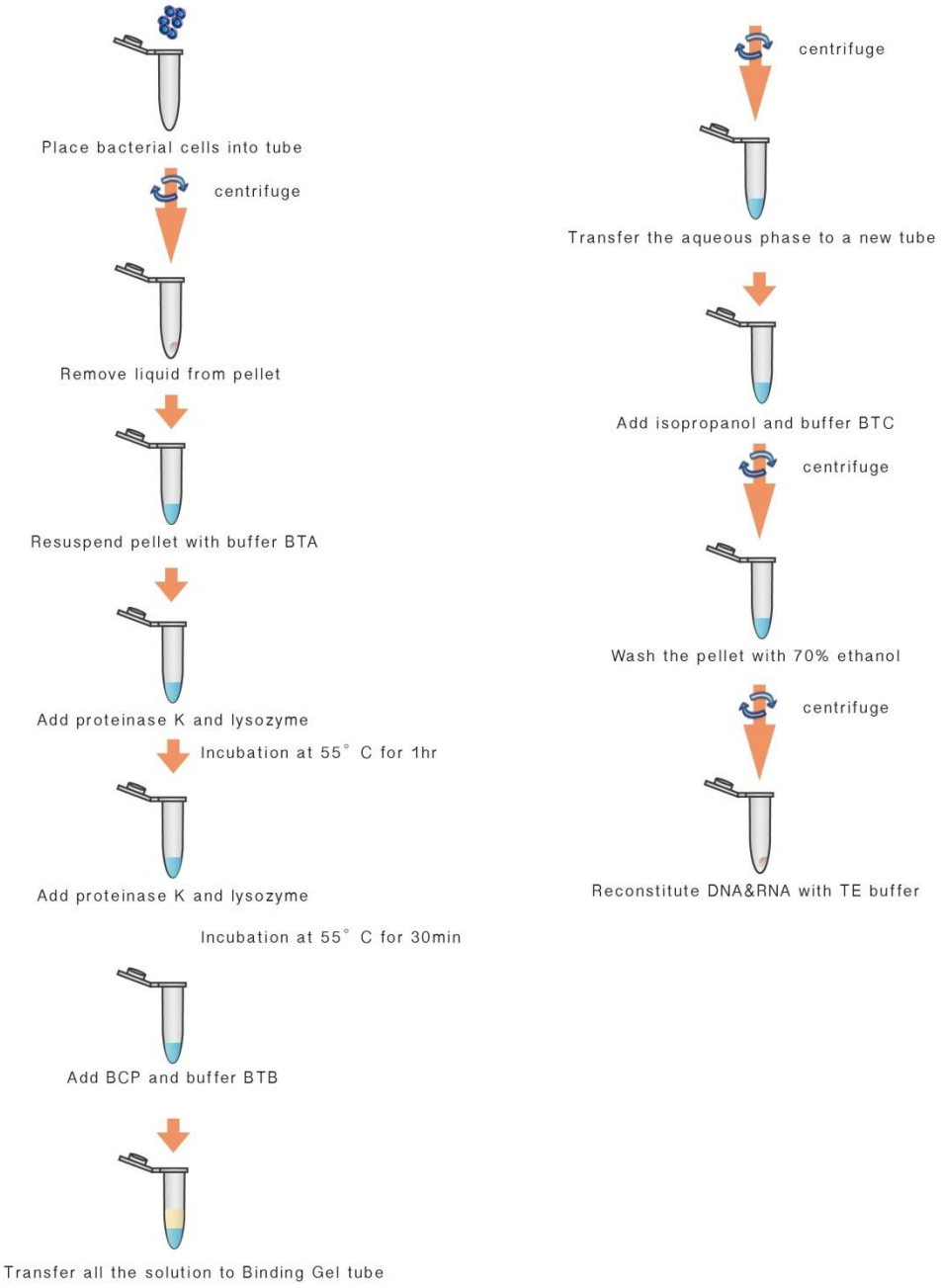
Kit Contents

Contents	TX-BFT01 (50 preps)
Buffer BTA	30 ml
Buffer BTB	12 ml
Buffer BTC	300 μl
Binding Gel	50 tubes

Storage

The TOOLS Bacterial and Fungal DNA and RNA Extraction Kit can be stored at room temperature for up to 24 months.

Workflow


Place bacterial cells into tube
centrifuge
Remove liquid from pellet
Resuspend pellet with buffer BTA
Add proteinase K and lysozyme
Incubation at 55° C for 1hr
Add proteinase K and lysozyme
Incubation at 55° C for 30min
Add BCP and buffer BTB
Transfer all the solution to Binding Gel tube
centrifuge
Transfer the aqueous phase to a new tube
Add isopropanol and buffer BTC
centrifuge
Wash the pellet with 70% ethanol
centrifuge
Reconstitute DNA&RNA with TE buffer

Protocol

Sample preparation

1. Place 1 mL of bacterial cells in a 2-mL microcentrifuge tube (RNase free).
2. Centrifuge for 5 min at 14,000 rpm, and remove a sufficient amount of supernatant.
3. Add 500 µL of BTA buffer to the tube and vortex for 60 s to resuspend the pellet thoroughly.
4. Add 5 µL of proteinase K (20 mg/mL) and 5 µL of lysozyme (100 mg/mL) to the tube and vortex for 60 s and then incubate at 55 °C for 1 h. Vortex the tube again for 20 s to mix thoroughly.

DNA and RNA extraction

1. Add 5 µL of proteinase K and 5 µL of lysozyme to the tube again.
   Vortex for 60 s and then incubate at 55 °C for 30 min.
2. Add 200 µL of BCP and 200 µL of BTB buffer to the tube, and mix the sample by inverting the tube three times. Transfer the entire solution to the Binding Gel tube (centrifuge at 13000 × g for 30 s before use).
3. Invert the Binding Gel tube three times (do not vortex) and then centrifuge at 12,000–16,000 × g for 5 min.
4. Transfer the supernatant to a new microcentrifuge tube, and add 5 µL of BTC buffer and 550 µL of isopropanol to the tube.
5. Invert the tube three times and incubate at 37 °C for 10 min.
6. Centrifuge at 14,000 rpm for 5 min. Remove the supernatant.
7. Add 600 µL of 70% ethanol. Centrifuge at 14,000 rpm for 5 min. Remove the supernatant.
8. Air dry the pellet, and rehydrate the pellet with 20–50 µL of TE buffer or double-distilled water (ddH_2O) (adjust the buffer volume according to the pellet size).

The product is for research only, not for diagnostic or clinical use.