

## **TOOLS Fecal DNA Extraction Kit**



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### Introduction

The TOOLS Fecal DNA Extraction Kit is a buffer-based system for cell DNA 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 is suitable for downstream applications, such as polymerase chain reaction, Southern blotting, genomic DNA library screening, and sequencing.

### Materials not provided

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

### **Kit Contents**

Contents	TX-STD01 (50 preps)
Buffer STA	30 ml
Buffer STB	12 ml
Buffer STC	300 µl
Binding Gel	50 tubes

### Storage

TOOLS Fecal DNA Extraction Kit can be stored at room temperature for up to 24 months.

### Protocol

### Sample preparation

1. Place 180–220 mg of stool in a 2-mL microcentrifuge tube and place the tube on ice.

Note: If the sample is in liquid form, add 200 µL of the sample into the microcentrifuge tube.

- 2. Add 500  $\mu$ L of STA buffer to the tube and vortex for 60 s to resuspend the pellet thoroughly.
- 3. Add 5 μL of proteinase K (20 mg/mL) and 5 μL of lysozyme (100 mg/mL) to the tube, vortex it for 60 s, and incubate it for 1 h at 55 °C. Vortex the tube again for 20 s to mix thoroughly.

### **DNA** extraction

- 1. Add 5  $\mu$ L of proteinase K and 5  $\mu$ L of lysozyme to the tube again. Vortex it for 60 s, and then incubate it at 55 °C for 30 min.
- 2. Add 200  $\mu$ L of BCP and 200  $\mu$ L of STB 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 three times (do not vortex), and then centrifuge it at  $12,000-16,000 \times g$  for 5 min.
- 4. Transfer the supernatant to a new microcentrifuge tube and add 5  $\mu$ L of STC buffer and 550  $\mu$ L of isopropanol to the tube.
- 5. Invert the tube three times and incubate at 37  $^{\circ}$ C for 10 min.
- 6. Centrifuge the tube at 14,000 rpm for 5 min. Remove the supernatant.
- 7. Add 600 µL of 70% ethanol. Centrifuge it at 14,000 rpm for 5 min. Remove the supernatant.
- Air dry the pellet and rehydrate the pellet with 20–50 μL of TE buffer or ddH<sub>2</sub>O (adjust the buffer volume according to the pellet size).

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

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