FavorPrep™ Stool DNA Isolation HE Mini Kit

■ Kit Contents

Cat. No.	FAST1030 (4 Preps)	FAST1033 (50 Preps)	FAST1034 (100 Preps)
SDE1 Buffer	1.8 ml	20 ml	40 ml
SDE2 Buffer	1 ml	10 ml	20 ml
SDE3 Buffer	1 ml	4 ml	8 ml
SDE4 Buffer (Concentrate)▲	1.5 ml	20 ml	40 ml
Wash Buffer (Concentrate)■	3 ml	25 ml	45 ml
Elution Buffer	0.5 ml	5 ml	7 ml
Proteinase K (Liquid)	100 µl	1050 µl	1050 µl x 2
Bead Tubes	4 pcs	50 pcs	100 pcs
FAST HE Columns	4 pcs	50 pcs	50 pcs x 2
Collection Tubes	8 pcs	100 pcs	100 pcs x 2
Elution Tubes	4 pcs	50 pcs	100 pcs
User Manual	1	1	1
Preparation of **SDE4 Buffer** and **Wash Buffer** by adding 96~100% ethanol.			
Volume of Ethanol for SDE4 Buffer▲	1.5 ml	20 ml	40 ml
Volume of Ethanol for Wash Buffer■	12 ml	100 ml	180 ml

All kit components are shipped at room temperature and should be stored at room temperatures between 15~25˚C.

■ Specification

Format/Principle	Spin Column (silica matrix)
Binding Capacity	≤125 µg DNA/Column
Operation Time	<45 mins
Sample Size	≤50 mg (solid stool)/≤200 mg (loose stool)
DNA yield	≤15 µg
Elution Volume	30 µl

■ Procedure Overview

Stool sample

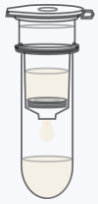
Centrifuge
18,000 xg, 1 min

- Weigh stool sample and add 320 µl SDE1-PK mixture in a bead tube
- Vortex for 5 mins
- Incubate at 60˚C for 10 mins

Centrifuge
18,000 xg, 3 mins

- Mix all the supernatant with 150 µl SDE2 Buffer in a 1.5 ml tube.
- Add 50 µl SDE3 Buffer and (Optional) 6 µl RNase A into the tube
- Incubate on ice for 5 mins

FAST HE Column



Centrifuge
18,000 xg, 1 min

- Mix 330 µl supernatant with 660 µl SDE4 Buffer (ethanol contained) in a 1.5 ml tube
- Transfer all the mixture into FAST HE Column

Centrifuge
18,000 xg, 1 min

- Add 900 µl Wash Buffer (ethanol contained)
- Repeat this step

Centrifuge
18,000 xg, 2 mins

- Drying the column membrane

Centrifuge
18,000 xg, 1 min

- Elution (add 30 µl Elution Buffer)
- Stand the column for 5 mins
- Obtain purified genomic DNA

Preparation Before Starting

1. Buffers provided in this system contain irritants. Wear gloves and lab coat when handling these buffers.
2. Additional materials: 96~100% ethanol, RNase A (Optional).
3. Set up a water bath or dry bath at 60°C and preheat the Elution Buffer to 60°C for elution step.
4. Check **SDE1 Buffer** before use. If precipitates are observed, warm-up SDE1 Buffer at 60°C until precipitates are completely dissolved.
5. Vortex **SDE3 Buffer** evenly before use.
6. Fresh preparation of **SDE1-PK mixture**, premix 300 µl of SDE1 Buffer and 20 µl of Proteinase K per sample before executing DNA extraction.
7. (Optional) If RNA-free genomic DNA is required) premix 50 µl **SDE3 Buffer** with 6 µl RNase A (50 mg/ml) per sample before executing DNA extraction.
8. Add indicated volume of ethanol (96~100%) into **SDE4 Buffer** and **Wash Buffer**, mix well and store at room temperature.
9. Recommended amount of sample:

Stool sample	Recommended amount
Loose stool (Higher than 80% water content) e.g., fish, cattle, human diarrhea	50~200 mg (50~200 µl)
Solid stool (50~80% water content) e.g., human, cat, dog	30~50 mg
Dry stool (low water content) e.g., mouse, reptile	10~30 mg

General Protocol

- **Note:** All centrifugation steps should be performed at **18,000 xg** at room temperature.
- **Note**: Avoid disturbing the pellet or debris while transferring the supernatant.

1. Weigh appropriate stool sample (see the **Recommended amount table**) into a bead tube. Add 320 µl **SDE1-PK mixture** to the tube.
2. Vortex using horizontal agitation (Horizontal Tube adapter) or a homogenizer to grind the fresh stool for 5 mins. Mix thoroughly and spin down.
   - **Note:** If the sample is frozen, reduce the grinding time from 5 mins to 1 min.
3. Incubate mixture at 60°C for 10 mins until the stool is lysed completely. Vortex occasionally during incubation.

General Protocol

4. Centrifuge for 1 min, then transfer all supernatant carefully into a 1.5 ml tube (not provided).
   - **Note:** If supernatant is less than 200 µl, the amount of sample is exceeded. Please decrease the sample amount or separate sample in multiple preparation.
5. Add 150 µl **SDE2 Buffer** and mix thoroughly by pulse-vortexing.
6. Add 50 µl **SDE3 Buffer** and **(Optional)** RNase A. Mix thoroughly by pulse-vortexing and incubate sample on ice for 5 mins.
7. Centrifuge for 3 mins, then transfer all supernatant carefully into a 1.5 ml tube (not provided).
8. Add 660 µl **SDE4 Buffer** (ethanol contained) to the sample mixture, mix thoroughly by pipetting.
9. Placed a **FAST HE Column** in a **Collection Tube**, then transfer all mixture carefully into the FAST HE Column.
10. Centrifuge for 1 min. Discard flow-through and place the FAST HE Column in a new Collection Tube.
11. Add 900 µl **Wash Buffer** (ethanol contained) to the FAST HE Column. Centrifuge for 1 min then discard flow-through.
12. Repeat step 12.
13. Centrifuge for 2 mins to dry the membrane. Discard flow-through and collection tube.
14. Place the **FAST HE Column** in an **Elution Tube**, then add 30 µl prewarmed **Elution Buffer** or ddH_2O (pH 7.5~9.0) directly onto the membrane. Stand the FAST HE Column for 5 mins.
    - **Important step!** For effective elution, ensure that the elution solution is dispensed onto the membrane center and absorbed completely.
15. Centrifuge for 1 min to elute DNA.