

FAVORGEN

Package Insert

Version 2 .2008.

FavorPrep™

96-Well Genomic DNA Kit

Sample:

200 µl of whole blood

20 mg of animal tissue

10^6 - 10^7 cultured cells

10^9 bacterial cultures

Yield: up to 50 µg of genomic DNA

Operation time: < 60 min.

Elution volume: 50-100 µl

Cat. No. FADWE 001 (4 plates)

FADWE 002 (10plates)

Store at room temperature (15 - 25 °C)

Introduction

96-Well Genomic DNA Extraction Kit is designed for high-throughput purification of total DNA (including genomic, mitochondrial and viral DNA) from whole blood and a variety of animal tissues or cells. The method use proteinase K and a chaotropic salt, guanidine hydrochloride to lyse cells and degrade protein, then DNA in chaotropic salt is bonded to glass fiber matrix of plate. After washing off the contaminants, the purified DNA is eluted by low salt elution buffer or water. The entire procedure can be completed in one hour without phenol/ chloroform extraction and alcohol precipitation. The kits can be used for manual filtration or with robotic handling systems and purified DNA with approximately 20-30 kb is suitable for PCR or other enzymatic reactions.

Quality Control

The quality of 96-Well Genomic DNA Kit is tested on a lot-to-lot basis. The purified DNA is checked by agarose gel analysis and quantified with spectrophotometer.

Caution

GB Buffer and W1 Buffer contain guanidine hydrochloride, which is harmful, and irritant agent. During operation, always wear a lab coat, disposable gloves, and protective goggles.

Introduction

Name	FADWE 001 4 plates	FADWE 002 10 plates
FATG1 Buffer	120 ml	240 ml
FATG2 Buffer	120 ml	240 ml
W1 Buffer	95 ml	95 ml x 3
Wash Buffer (concentrated)*	50 ml	50 ml x 3
Elution Buffer	60ml	60 ml x 2
Proteinase K**	90 mg	225 mg
96-Well DNA binding plate	4 pcs	10 pcs
96-Well 350 µl collection plate	4 pcs	10 pcs
Adhesive film	8 pcs	20 pcs

* Add 200 ml of ethanol (96-100%) to Wash Buffer prior to the initial use.

** For Cat. No.: FADWE 001, add 9 ml ddH₂O to the bottle and mix by vortexing. For Cat. No.: FADWE 002, add 22.5 ml ddH₂O to the bottle and mix by vortexing. Store prepared Proteinase K (10 mg/ ml) at 4 °C.

Blood Protocol

Step 1

Cell lysis

- Add **200 µl FATG2 Buffer and 20 µl Proteinase K (10 mg/ ml)** to each well of a 96-Well 2 ml plate.
- Apply **200 µl of** blood sample to each well and mix by shaking. Seal with Adhesive Film.
- Incubate at 60°C for 20 minutes until the sample lysate is clear.
- Preheat required Elution Buffer (100 µl per sample) at 60°C. (For **Step 4 DNA elution**)

Step 2

DNA Binding

- Place a **96-Well DNA Binding Plate** on top of the vacuum manifold.
- Add **200 µl of ethanol** to each sample lysate in 96-Well 2ml plate from previous step. Mix immediately by pipetting 5-10 times.
- Transfer the lysate mixture to **96-Well DNA Binding Plate**.
- Apply vacuum at 10 inches Hg for 30 seconds until wells have emptied.

Step 3

Wash

- Add **300 μ l of W1 Buffer** to each well of the **96-Well DNA Binding Plate**.
- Apply vacuum at 10 inches Hg for 30 seconds until wells have emptied.
- Add **600 μ l of Wash Buffer (ethanol added)** to each well of the **96-well DNA Binding Plate** to wash again.
- Apply vacuum at 10 inches Hg for 30 seconds until wells have emptied.
- Apply vacuum for additional 10 min (or incubate at 60°C) to remove ethanol residue.

Step 4

DNA Elution

Standard elution volume is 70 μ l. If less sample is to be used, reduce the elution volume (50-70 μ l) to increase DNA concentration.

- Transfer the DNA Binding Plate on a clean **96-Well 350 μ l collection plate**.
- Add **70 μ l of preheated Elution Buffer** in the center of each well of DNA Binding Plate.
- Stand for 3 minutes until Elution Buffer or water absorbed by the matrix.
- Place the combined plates in a rotor bucket and centrifuge at 3,500 rpm for 5 min to elute purified DNA into the 96-Well 350 μ l collection plate. Seal with Adhesive Film and store 4°C or -20°C.

Tissue Protocol

Step 1

Cell lysis

- Add **200 µl FATG1 Buffer and 20 µl Proteinase K (10 mg/ ml)** to each well of a 96-Well 2 ml plate (not provided).
- Cut up to 20 mg of animal tissues (or 0.5 cm of mouse tail) and transfer into each well of 96-Well 2 ml plate. Seal with Adhesive Film.
- Incubate the plate with shaking at 60°C for 1-2 hours to lyse the sample.
- If RNA-free genomic DNA is required, add **5 µl of RNase A (50 mg/ ml, not provided)** to each well and incubate at room temperature for 4 minutes.
- Add **200 µl FATG2 Buffer to each well** and mix by shaking (seal with Adhesive Film.).
- Incubate at 70°C for 20 minutes until the sample lysate is clear.
- Preheat required Elution Buffer (50 µl per sample) at 70°C. **(For Step 4 DNA elution)**
- If there are insoluble material present following incubation, centrifuge the plate for 5 minutes at full speed and transfer the supernatants to a new 96-Well 2 ml plate (not provided).

Step 2

DNA Binding

- Place a **96-Well DNA Binding Plate** on top of the vacuum manifold.
- Add **200 µl of ethanol** to each sample lysate in 96-Well 2 ml plate from previous step. Mix immediately by pipetting 5-10 times.
- Transfer the lysate mixture to **96-Well DNA Binding Plate**.
- Apply vacuum at 10 inches Hg for 30 seconds until wells have emptied.

Step 3

Wash

- Add **300 µl of W1 Buffer** to each well of the **96-Well DNA Binding Plate**.
- Apply vacuum at 10 inches Hg for 30 seconds until wells have emptied.
- Add **600 µl of Wash Buffer (ethanol added)** to each well of the **96-Well DNA Binding Plate** to wash again.
- Apply vacuum at 10 inches Hg for 30 seconds until wells have emptied.
- Apply vacuum for additional 10 min (or incubate at 60°C) to remove ethanol residue.

Step 4

DNA Elution

Standard elution volume is 70 µl. If less sample is to be used, reduce the elution volume (50-70 µl) to increase DNA concentration.

- Transfer the DNA Binding Plate on a clean **96-Well 350µl collection plate**.
- Add **70 µl of preheated Elution Buffer** in the center of each well of DNA Binding Plate.
- Stand for 3 minutes until Elution Buffer or water absorbed by the matrix.
- Place the combined plates in a rotor bucket and centrifuge at 3,500 rpm for 5 min to elute purified DNA into the 96-Well 350µl collection plate. Seal with Adhesive Film and store 4°C or -20°C.