FAVORGEN

For research use only

FavorPrep™ 96-Well GEL/ PCR Clean UP Kit

User Manual

Version 2 .2008.

Cat. No. FAPKE 001 (4 plates)

FAPKE 002 (10plates)

Store at room temperature (15 - 25℃)

Introduction

FavorPrep™ 96-well GEL/ PCR Clean Up kit is designed for rapid purification of fragment DNA from agarose gel , PCR or other enzymatic reaction. The procedure uses chaotropic salt to dissolve agarose and denature enzyme. With the suitable binding condition provided by this system, the DNA in the sample mixture binds to glass fiber matrix in the 96-well DNA Binding Plate. The contaminants are washed with an ethanol-contained wash buffer and finally, the purified DNA is eluted by low salt Elution buffer or water. The protocol does not require DNA phenol extraction and alcohol precipitation. The entire procedure can be done within 30~40 minutes and the purified DNA is ready for restriction digestion, ligation, labeling, PCR, and sequencing reaction.

Specification:

Sample Size: up to 50μ l PCR or other enzymatic reaction mixture
Up to 100 mg agarose gel slice

Binding Capacity: up to 10μ l/ well

DNA Size range: 70 bp~12Kb

Operation: centrifuge/ Vacuum manifold

Handling Time: about 30 minutes for PCR clean up
about 40 minutes for Gel DNA extraction

Recovery: 90~95% PCR clean up
70~85%for Gel DNA extraction

Application: Fluorescent or radioactive sequencing
Restriction digestion
Library screening
Ligation
Labeling
Transformation

Kit Contents:

Name	FAPWE 001 (4 Plates)	FAPWE 002 (10Plates)
Binding Buffer D1	135 ml	340 ml
Wash Buffer (concentrated)*	50 ml	50 ml x 3
Elution Buffer	30 ml	60 ml
96-Well DNA Binding plate	4 pcs	10 pcs
96-Well 0.35 ml Collection plate	4 pcs	10 pcs
Adhesive Film	8 pcs	20 pcs

* Add 200 ml of ethanol to each Wash Buffer before first open.

Important Notes:

1.Buffer provided in this kit contain irritants. Wear gloves and lab coat when handling these buffers.

2. When excising the agarose gel, remove the extra gel to minimize the size of the gel.

3. Preheat a water bath to 50 ℃ for Gel DNA Extraction Protocol.

4. Add 200 ml of ethanol (96~100%) to each Wash Buffer before first open.

PCR Clean Up Protocol (Centrifuge)

Step1

- Transfer **up to 50μ l** of PCR or enzymatic product to each well of a clean 96-Well Storage Plate (1~2 ml)(not provided).
- Add **250μ l** of **Binding Buffer D1 to each well**. Mix well by Pipetting

Step 2: DNA Binding

- Place a **96-Well DNA Binding Plate** on top of a 96-Well 2 ml Plate (not provided).
- Transfer the sample mixture from **step 1** to the **96-Well DNA Binding Plate**. (about 300 μl)
- Place the assembly plates in a rotor bucket and centrifuge at 3,500 rpm for 5 min.
- Discard the flow-through and return the **96-Well DNA Binding Plate** to the 96-Well 2 ml Plate.

Step3: Wash

- Add **650 μl** of **Wash Buffer (ethanol added)** into each well of the **96-Well Plasmid Plate**.
- Place the assembly plates in a rotor bucket and centrifuge at 3,500 rpm for 5 min. ·
- Discard the flow-through and return the **96-Well DNA Binding Plate** back to the 96-Well 2 ml Plate.
- Place the assembly plates in a rotor bucket and centrifuge at 3,500 rpm for an additional 10 minutes (or incubate at 60 °C for 10 minutes) to remove residual ethanol.

Step 4: DNA Elution

- Transfer the **96-Well DNA Binding Plate** on a clean **96-Well 350μl collection plate (provided)**.
- Add **50 μl** of **Elution Buffer** or ddH_2O (pH8.0-8.5) into the membrane center of the **96-Well DNA Binding Plate**.
- Stand the **96-Well DNA Binding Plate** for 2 minutes until Elution Buffer or ddH_2O has been absorbed completely by the membrane.
- Place the assembly 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.

PCR Clean Up Protocol (Vacuum)

Step1

- Transfer **up to 50µ l** of PCR or enzymatic product to each well of a clean 96-Well Storage Plate (1~2 ml)(not provided).
- Add **250µ l** of **Binding Buffer D1 to each well**. Mix well by Pipetting

Step 2: DNA Binding

- Place a **96-Well DNA Binding Plate** on top of the vacuum manifold.
- Transfer the sample mixture from **step 1** to each well of the **96-Well DNA Binding Plate**. (about 300 µl)
- Apply vacuum at 10 inches Hg for 5 minutes until wells have emptied.

Step 3: Wash

- Add **650 µl** of **Wash Buffer (ethanol added)** to each well of the **96-Well DNA Binding Plate**.
- Apply vacuum at 10 inches Hg for 5 minutes until wells have emptied.
- Apply vacuum at 10 inches Hg for an additional 10 min (or incubate at 60°C for 10 min) to remove residual ethanol.

Step 4*: *DNA Elution

- Transfer the **96-Well DNA Binding Plate** on a clean **96-Well 0.35 ml collection plate (provided)**.
- Add **50 µl** of **Elution Buffer** or ddH_2O (pH8.0-8.5) into the membrane center of the **96-Well DNA Binding Plate**.
- Stand the **96-Well DNA Binding Plate** for 2 minutes until Elution Buffer or ddH_2O has been absorbed completely by the membrane.
- Place the assembly 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.

Gel DNA Extraction Protocol (Centrifuge)

Step1

- Transfer **up to 100 mg** of agarose gel (containing relevant DNA fragment) to each well of a clean 96-Well Storage Plate (1~2 ml)(not provided).
- Add **300μ l** of **Binding Buffer D1 to each well**. Incubate at 50 ℃ for 10~15 minutes until the gel slice dissolved completely.

Step 2: DNA Binding

- Place a **96-Well DNA Binding Plate** on top of a 96-Well 2 ml Plate (not provided).
- Transfer the sample mixture from **step 1** to the **96-Well DNA Binding Plate**. (about 300 μl)
- Place the assembly plates in a rotor bucket and centrifuge at 3,500 rpm for 5 min.
- Discard the flow-through and return the **96-Well DNA Binding Plate** to the 96-Well 2 ml Plate.

Step 6: Wash

- Add **650 μl** of **Wash Buffer (ethanol added)** into each well of the **96-Well Plasmid Plate**.
- Place the assembly plates in a rotor bucket and centrifuge at 3,500 rpm for 5 min. ·
- Discard the flow-through and return the **96-Well DNA Binding Plate** back to the 96-Well 2 ml Plate.
- Place the assembly plates in a rotor bucket and centrifuge at 3,500 rpm for an additional 10 minutes (or incubate at 60 °C for 10 minutes) to remove residual ethanol.

Step 7: DNA Elution

- Transfer the **96-Well DNA Binding Plate** on a clean **96-Well 350μl collection plate (provided)**.
- Add **50 μl** of **Elution Buffer** or ddH_2O (pH8.0-8.5) into the membrane center of the **96-Well DNA Binding Plate**.
- Stand the **96-Well DNA Binding Plate** for 2 minutes until Elution Buffer or ddH_2O has been absorbed completely by the membrane.
- Place the assembly 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.

Gel DNA Extraction Protocol (Vacuum)

Step1

- Transfer **up to 100 mg** of agarose gel (containing relevant DNA fragment) to each well of a clean 96-Well Storage Plate (1~2 ml)(not provided).
- Add **300μ l** of **Binding Buffer D1 to each well**. Incubate at 50 ℃ for 10~15 minutes until the gel slice dissolved completely.

Step 2: DNA Binding

- Place a **96-Well DNA Binding Plate** on top of the vacuum manifold.
- Transfer the sample mixture from **step 1** to each well of the **96-Well DNA Binding Plate**. (about 300 μl)
- Apply vacuum at 10 inches Hg for 5 minutes until wells have emptied.

Step 3: Wash

- Add **650 μl** of **Wash Buffer (ethanol added)** to each well of the **96-Well DNA Binding Plate**.
- Apply vacuum at 10 inches Hg for 5 minutes until wells have emptied.
- Apply vacuum at 10 inches Hg for an additional 10 min (or incubate at 60°C for 10 min) to remove residual ethanol.

Step 4: ***DNA Elution***

- Transfer the **96-Well DNA Binding Plate** on a clean **96-Well 0.35 ml collection plate (provided)**.
- Add **50 μl** of **Elution Buffer** or ddH_2O (pH8.0-8.5) into the membrane center of the **96-Well DNA Binding Plate**.
- Stand the **96-Well DNA Binding Plate** for 2 minutes until Elution Buffer or ddH_2O has been absorbed completely by the membrane.
- Place the assembly 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.