FavorPrep™ 96-well Gel/ PCR Clean-Up DNA Kit

(For Research Use Only)

Kit Contents/ Cat. No.:

	FAPKE96001 1 plate	FAPKE96002 2 plates	FAPKE96004 4 plates
Binding Buffer D1	60 ml	135 ml	135 ml x 2
Wash Buffer (concentrated)	35 ml †	35 ml x 2 †	35 ml x 4†
Elution Buffer	20 ml	50 ml	50 ml x 2
Filter Plate (96-Well DNA binding plate)	1 plate	2 plates	4 plates
96-Well 2 ml Plate	3 plate	6 plates	12 plates
96-Well PCR plate	1 plate	2 plates	4 plates
Adhesive Film	2 pcs	4 pcs	8 pcs

† Add 140 ml ethanol (96~100%)to each Wash Buffer when first open.

Quality Control

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

Specification:

Sample Size: up to 200 mg agarose gel slice
up to 100 µl PCR or other enzymatic reaction mixture
Binding Capacity: up to 20 µg/ well
DNA Size range: 70 bp ~12Kb
Minimum elution volume: 50 µl
Operation: centrifuge/ vacuum manifold
Handling Time: about 40 minutes for gel DNA extraction
about 30 minutes for PCR clean up
Recovery: 70~85 % for gel DNA extraction
90~95 % PCR clean up
Downstream application: Fluorescent or radioactive sequencing, Restriction digestion, Library screening, Ligation, Labeling, Transformation

Important Notes:

1. Buffers provided in this system contain irritants. Wear gloves and lab coat when handling these buffers.
2. The maxium sample size is described on specification, do not use the sample more than the limitation.
3. When excising the agarose gel, remove the extra gel to minimize the size of the gel.
4. Add ethanol (96~100 %) to Wash Buffer when first open.
5. Prepare bath to 55 °C before the operation for Gel DNA Extraction Protocol.
6. Preheat the Elution Buffer to 60 °C for DNA elution to increase the recovery of DNA fragment >1,000 bp.

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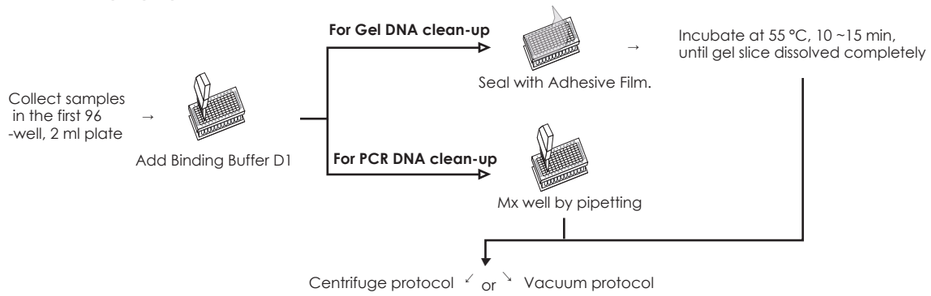
Brief procedure:

Material to be provided by user for a 96-well preparation

1. Centrifuge equiment with a swing -bucket rotor and adaptor for 96-well plate, capable of at least 5,600 ~ 6,000 X g.
2. Vacuun manifold for 96-well plate and Vaccum source for 10 inches Hg.
3. 96 ~100 % ethanol for preparation of the Wash Buffer when first use.
4. For Gel DNA Extraction, a 55 °C incubator for dissolving agarose gel.
5. For increasing the recovery of DNA fragment (>1,000 bp), a 60 °C incumator for pre-heating Elution Buffer or ddH2O.

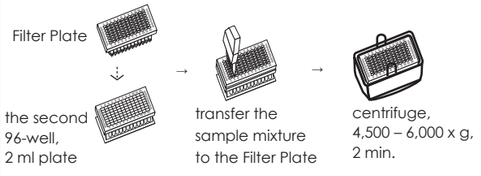
• STEP 1. Sample preparation



• STEP 2. Bind DNA to Filter Plate:

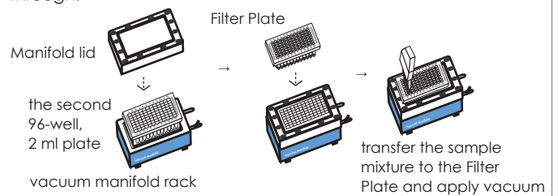
centrifuge protocol

· Transfer the sample mixture to combined plate. (Filter Plate + the second 96-well, 2 ml Plate)
· Centrifuge and discard the flow-through.



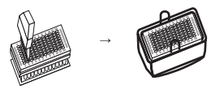
vacuum protocol

· Transfer the sample mixture to a Filter Plate assembled with manifold and the second 96-well, 2 ml Plate .
· Apply vacuum until the wells have emptied and discard the flow -through.



• STEP 3. Wash Filter Plate *twice* with Wash Buffer

1st Washing : Add Wash Buffer and centrifuge 5,600 - 6,000 x g, 2 min.
2nd Washing : Add Wash Buffer and centrifuge 5,600 - 6,000 x g, 15 min.



1st Washing : Add Wash Buffer and apply vacuumat 10 inches Hg for until the wells have emptied.
2nd Washing: add Wash Buffer and apply vacuumat 10 inches Hg for 10 min.

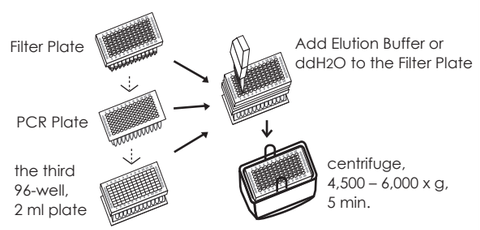
• STEP 4 . Dry the membranes of Filter Plate:

· Stand the Filter plate on a clean paper towel at room temperature 10 min.

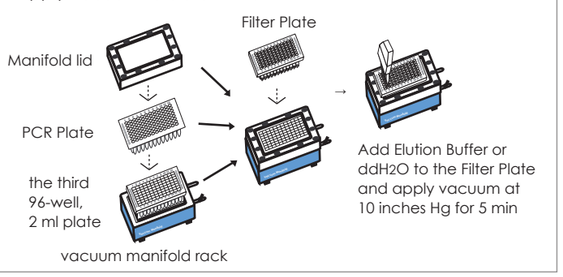
· Gently tap the tips of the Filter Plate on paper towel to remove residual liquid
· Apply vacuum at 10 inches Hg for an addition 10 min.

• STEP 5. DNA Elution:

· Add Elution Buffer or ddH2O to the Filter Plate. Stand for 3 min.
· Centrifuge to elute DNA.



· Add Elution Buffer or ddH2O to the Filter Plate. Stand for 3 min.
· Apply vacuum to elute DNA.



Protocol: (centrifugation processing)
- using centrifuging force to handle DNA binding step and washing steps.

Material to be provided by user for a 96-well preparation

1. Centrifuge equiment with a swing -bucket rotor and adaptor for 96-well plate, capable of at least 5,600 ~ 6,000 X g.
2. Preheat required Elution Buffer or ddH2O (40 ~100 µl per well) at 60 °C. (For Step 4 DNA elution)
   -- For increasing the recovery of DNA fragment (>1,000 bp).

Please Read Important Notes Before Starting The Following Steps.

STEP 1: Sample Preparation

For Gel DNA Clean-Up

- Transfer up to 200 mg of agarose gel (containing relevant DNA fragment) to each well of a 96-Well 2 ml Plate (provided, the first 96-well, 2 ml plate).
- Add 500 µl of Binding Buffer D1 to each well, sealing with a adhesive film. Incubate at 55 °C for 10~15 minutes until the gel slice dissolved completely. During the incubation, briefly shake the incubated plate for every 5 minutes to make the sample mixture mix well with Binding Buffer D1.

For PCR DNA Clean-Up

- Transfer up to 100 µl of PCR or enzymatic product to each well of a clean 96-Well 2 ml Plate. (provided, the first 96-well, 2 ml plate).
- Add 5 volume of Binding Buffer D1 to each well. Mix well by pipetting.
  - For example, Add 250 µl of Binding Buffer D1 to 50 µl of PCR or enzymatic product.

STEP 2: DNA Binding

- Place a Filter Plate (96-Well DNA Binding Plate) on top of a clean 96-well plate (provided, the second 96-Well 2 ml Plate).
- Transfer the sample mixture to the Filter Plate.
- Place the combined plate (Filter Plate combined with the second 96-Well, 2 ml plate) in a rotor bucket and centrifuge at 4,500 – 6,000 x g for 2 min.
- Discard the flow-through and return the Filter Plate back to the second 96-Well 2 ml Plate.

STEP 3: Wash Filter Plate Twice (W1 Buffer and Wash Buffer)

1st Washing

- Add 650 µl of Wash Buffer (ethanol added) to each well of the Filter Plate.
- Place the combined plate (Filter Plate combined with the second 96-Well 2 ml plate) in a rotor bucket and centrifuge at 4,500 – 6,000 xg for 2 min.
- Discard the flow-through and return the Filter Plate back to the second 96-Well 2 ml Plate.

2nd Washig

- Add 650 µl of Wash Buffer to each well of the Filter Plate.
- Place the combined plate in a rotor bucket and centrifuge at 4,500 – 6,000 xg for 15 min.
- Discard the second 96-Well 2 ml plate and the flow-through.

STEP 4: Dry the membranes of Filter Plate

- Stand the Filter plate on a clean paper towel at room temperature 10 min.

STEP 5: DNA Elution

- Place a 96-well PCR Plate (provided) on top of a clean 96-Well 2 ml Plate. (provided, the third 96-well, 2 ml plate). and place the Filter Plate on the 96-Well PCR plate.
  (top: Filter Plate, middle: 96-well PCR Plate, bottom: 96-Well 2 ml plate)
- Add 50 ~ 100 µl of Elution Buffer or ddH2O (pH8.0-8.5) to the membrane center of the Filter Plate. Stand for 3 min until Elution Buffer or ddH2O has been absorbed by the membrane completely.
- Place the combined plate in a rotor bucket and centrifuge at 4,500 – 6,000 x g for 5 min to elute DNA to the 96-well PCR plate.

Protocol: (Vacuum processing)
- using vacuum force to handle DNA binding step and washing steps.

Material to be provided by user for a 96-well preparation

1. Vacuun manifold for 96-well plate and a vaccum source for 10 inched Hg.
2. Preheat required Elution Buffer or ddH2O (40 ~100 µl per well) at 60 °C. (For Step 4 DNA elution)
   -- For increasing the recovery of DNA fragment (>1,000 bp).

Please Read Important Notes Before Starting The Following Steps.

STEP 1: Sample Preparation

For Gel DNA Clean-Up

- Transfer up to 200 mg of agarose gel (containing relevant DNA fragment) to each well of a 96-Well 2 ml Plate (provided, the first 96-well, 2 ml plate).
- Add 500 µl of Binding Buffer D1 to each well, sealing with a adhesive film. Incubate at 55 °C for 10~15 minutes until the gel slice dissolved completely. During the incubation, briefly shake the incubated plate for every 5 minutes to make the sample mixture mix well with Binding Buffer D1.

For PCR DNA Clean-Up

- Transfer up to 100 µl of PCR or enzymatic product to each well of a clean 96-Well 2 ml Plate. (provided, the first 96-well, 2 ml plate).
- Add 5 volume of Binding Buffer D1 to each well. Mix well by pipetting.
  - For example, Add 250 µl of Binding Buffer D1 to 50 µl of PCR or enzymatic product.

STEP 2: DNA Binding

- Place a clean 96-well plate (provided, the second 96-Well, 2 ml Plate) on the rack of vacuum manifold and cover the manifold lid.
- Place a Filter Plate (96-well DNA Binding Plate, provided) on top of the second 96-Well 2 ml plate.
- Transfer the sample mixture to the Filter Plate.
- Apply vacuum until the wells have emptied.

STEP 3: Wash Filter Plate Twice (W1 Buffer and Wash Buffer)

1st Washing

- Add 650 µl of Wash Buffer (ethanol added) to each well of the Filter Plate.
- Apply vacuum at 10 inches Hg until the wells have emptied.

2nd Washing

- Add 650 µl of Wash Buffer to each well of the Filter Plate.
- Apply vacuum at 10 inches Hg for 10 min.

STEP 4: Dry the membranes of Filter Plate

- Gently tap the tips of the Filter Plate on paper towel to remove residual liquid
- Apply vacuum at 10 inches Hg for an addition 10 min.

STEP 5: DNA Elution

- place a 96-well PCR Plate (provided) on top of a clean 96-well plate (provided, the third 96-Well, 2 ml Plate). Place the combined plate on the rack of vacuum manifold and cover the manifold lid.
- place the Filter Plate on top of the 96-well PCR Plate.
- Add 50 ~ 100 µl of Elution Buffer or ddH2O (pH8.0-8.5) to the membrane center of the Filter Plate. Stand for 3 min until Elution Buffer or ddH2O has been absorbed by the membrane completely.
- Apply vacuum at 10 inches Hg for 5 min to elute DNA to the 96-well PCR plate.