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# FavorPrep<sup>™</sup> MicroElute Gel / PCR Purification Kit

 For extraction of DNA fragments from agarose gel
 For purification of PCR products or reaction mixtures (concentration and desalination of reaction mixtures) Cat. No.: FAEPK 000 FAEPK 001 FAEPK 001-1 FAEPK 001-2 (For Research Use Only)

### **Kit Contents:**

| Cat. No:   | FAEPK 000        | FAEPK 001  | FAEPK 001-1 | FAEPK 001-2 |  |
|--|------------------|------------|-------------|-------------|--|
|  | (4 preps_sample) | (50 preps) | (200 preps) | (300 preps) |  |
| MF Buffer  | 3 ml             | 30 ml      | 115 ml      | 175 ml      |  |
| Wash Buffer (concentrate) <sup>a</sup>                   | 1 ml             | 12.5 ml    | 35 ml       | 50 ml       |  |
| Elution Buffer   | 0.5 ml           | 5 ml       | 5 ml        | 5 ml        |  |
| MF Column  | 4 pcs            | 50 pcs     | 200 pcs     | 300 pcs     |  |
| Collection Tube  | 4 pcs            | 50 pcs     | 200 pcs     | 300 pcs     |  |
| User Manual  | 1                | 1          | 1           | 1           |  |
| Preparation of Wash Buffer by adding ethanol (96 ~ 100%) |                  |            |             |             |  |
| Ethanol volume for Wash Buffer <sup>a</sup>              | 4 ml             | 50 ml      | 140 ml      | 200 ml      |  |

### **Specification:**

Principle: spin column (silica matrix)

DNA Binding capacity of spin column: 5 µg

Sample size: up to 200 mg of agarose gel up to 100 µl of reaction solution

DNA size: 65 bp ~ 10 kbp

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

Operation time: 10 ~ 20 min

Elution volume: 10 ~12 µl

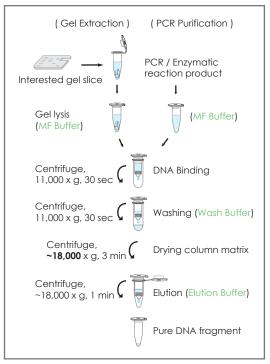
### **Important Notes:**

- 1. Buffer provided in this kit contain irritants. Wear gloves and lab coat when handling these buffer.
- Add the required volume of ethanol (96~100%) to Wash Buffer before use.
- 3. For gel DNA extraction, excising the extra agarose gel to minimize the size of the gel (up to 200 mg).
- For concentraction or purification of DNA fragments from enzymatic reactions, the maximum sample volume is 100 µl and the maximum amount of DNA fragments is 5 µg.
- 5. Centrifugation steps are done by a microcentrifuge capable of the speed at 11,000 ~1,8000 x g.

#### Gel Extraction Protocol: For extraction of DNA fragments from agarose gel Please Read Important Notes Before Starting Following Steps. HINT: Prepare a 55 °C dry bath or water bath for step 4.

- 1. Excise the agarose gel with a clean scalpel.
- Remove the extra agarose gel to minimize the size of the gel slice.
- 2. Transfer up to 200 mg of the gel slice into a microcentrifuge tube. (not provided).
- The maximum volume of the gel slice is 200 mg.
- 3. Add 500 µl of MF Buffer to the sample and mix by vortexing.
- For > 2% agarose gels, add 1000  $\mu$ l of MF Buffer.
- 4. Incubate at 55 °C for 5 ~10 minutes and vortex the tube every 2 ~ 3 minutes until the gel slice dissolved completely.
  During incubation, interval vortexing can accelerate the gel dissolved.
  - Make sure that the gel slice has been dissolved completely before proceed the next step.
- 5. Cool down the sample mixture to room temperature. And place a MF Column into a Collection Tube.
- 6. Transfer 700 µl of the sample mixture to the MF Column. Centrifuge at 11,000 x g for 30 seconds, then discard the flow-through.
- If the sample mixture is more than 700  $\mu I$ , repeat this step for the rest of the sample mixture.
- 7. Add 600 µl of Wash Buffer (ethanol added) to the MF Column. Centrifuge at 11,000 x g for 30 seconds, then discard the flow-through.
  - Make sure that ethanol (96-100 %) has been added into Wash Buffer when first use.

# Brief procedure:



8. Centrifuge again at full speed (~ 18,000 x g) for an additional 3 minutes to dry the column matrix.

• Important step ! The residual liquid should be removed thoroughly on this step.

- 9. Place the MF Column to a new microcentrifuge tube (not provided).
- 10. Add 10 ~ 12 µl of Elution Buffer or ddH2O to the membrane center of the MF Column. Stand the MF Column for 1 min.
  - Important step ! For effective elution, make sure that the elution solution is dispensed onto the membrane center and is absorbed completely. • Important : Do not elute the DNA using less than suggested volume (10 µl). It will lower the final yield.
- The average eluate volume is 10 µl from 12 µl elution buffer volume.
- 11. Centrifuge at full speed (~ 18,000 x g) for 1 min to elute DNA.

#### PCR Clean-Up Protocol: For purification of PCR products or reaction mixtures Please Read Important Notes Before Starting Following Steps.

- 1. Transfer 10 ~ 100 µl of PCR product (excluding oil) to a microcentrifuge tube (not provided) and add 5 volumes of MF Buffer, mix well by vortexing.
  - For example, Add 250 µl of MP Buffer to 50 µl of PCR product.
  - The maximum volume of PCR product is 100 µl (excluding oil). Do not excess this limit. If PCR product is more than 100 µl, separate it into multiple tubes.
- 2. Place a MF column into a Collection Tube.
- 3. Transfer the sample mixture to the MF Column. Centrifuge at 11,000 x g for 30 seconds, then discard the flow-through.
- 4. Add 600 µl of Wash Buffer (ethanol added) to the MF Column. Centrifuge at 11,000 x g for 30 seconds, then discard the flow-through.
- Make sure that ethanol (96-100 %) has been added into Wash Buffer when first open.
- 5. Centrifuge again at full speed (~18,000 x g) for an additional 3 minutes to dry the column matrix.
- Important step ! The residual liquid should be removed thoroughly on this step.
- 6. Place the MF Column to a new microcentrifuge tube (not provided).
- 7. Add 10~12 µl of Elution Buffer or ddH2O to the membrane center of the MF Column. Stand the MF Column for 1 min.
  - Important step ! For effective elution, make sure that the elution solution is dispensed onto the membrane center and is absorbed completely.
  - Important : Do not elute the DNA using less than suggested volume (10 µl). It will lower the final yield.
  - The average eluate volume is 10 µl from 12 µl elution buffer volume.
- 8. Centrifuge at full speed (~18,000 x g) for 1 min to elute DNA.

# **Troubleshooting**

(For Gel Extraction)

| Problems   | Possible reasons                                     | Solutions  |
|--|--|--|
| The gel slice is<br>hard to<br>dissolve                    | Agarose gel of high<br>percentage (> 2 %)<br>is used | Add 5 volumes of FAGP Buffer to 1 volume of the gel slice.   |
|  | The size of the gel slice is too large               | If the gel slice is more than<br>200 mg, separate it into<br>multiple tubes.                                 |
| Low or none<br>recovery of<br>DNA fragment                 | The column is loaded with<br>too much agarose gel    | The maximum volume of the gel slice is 200 mg per column.  |
|  | Elution of DNA fragment is<br>not efficient          | Make sure the pH of Elution<br>Buffer or ddH2O is between<br>7.0- 8.5.                                       |
|  |  | Make sure that the elution<br>solution has been completely<br>absorbed by the membrane<br>before centrifuge. |
|  | The size of DNA fragment<br>is larger than 5 Kb      | Preheat the elution solution to 60 °C before use.  |
| Eluted DNA   | Contaminated scalpel                                 | Using a new or clean scalpel.  |
| contains<br>non-specific<br>DNA fragment                   | DNA fragment is dena-<br>tured                       | Incubate eluted DNA at 95 °C<br>for 2 min, then cool down<br>slowly to reanneal denatured<br>DNA.            |
| Poor perfor-<br>mance in the<br>downstream<br>applications | Salt residue remains in<br>eluted DNA fragment       | Wash the column twice with<br>Wash Buffer.   |
|  | Ethanol residue remains in eluted DNA fragment       | Do discard the flow-through<br>after washing with Wash<br>Buffer and centrifuge for an<br>additional 3 min.  |

(For PCR Clean-Up)

| Problems   | Possible reasons                                | Solutions   |
|--|---|---|
| Low or none<br>recovery of<br>DNA fragment                 | Apply more than 100 µl of<br>PCR product        | If PCR product is more than<br>100 µl, separate it into<br>multiple tubes.  |
|  | Elution of DNA fragment is<br>not efficient     | Make sure the pH of Elution<br>Buffer or ddH2O is between<br>7.0- 8.5.  |
|  |   | Make sure that the elution<br>solution has been completely<br>absorbed by the column<br>membrane before centrifuga<br>tion. |
|  | The size of DNA fragment<br>is larger than 5 Kb | Preheat the elution solution to 60 °C before use.   |
| Poor perfor-<br>mance in the<br>downstream<br>applications | Salt residue remains in<br>eluted DNA           | Wash the column twice with<br>Wash Buffer.  |
|  | Ethanol residue remains in eluted DNA           | Do discard the flow-through<br>after washing with Wash<br>Buffer and centrifuge for an<br>additional 3 min.                 |



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