MQ PCR/Gel Product Purification Kit

Cat. No. PGP-M-001-100 Size: 100 Preparations

MOLEQULE-ON[®]

Kit Components	Volume
Binding Buffer GP1	80 ml
Wash Buffer GP2 concentrate	25 ml
Elution Buffer GP3	10 ml
GP Column	100
2 ml Collection Tube	100
Gel Band Cutter (trial pack)	5

Preparation

Add 100ml of 96-100% of Ethanol in Was Buffer GP2 concentrate, mix well and use in further procedure.

Materials Required But Not Supplied

96-100% Ethanol, 1.5 ml microcentrifuge tube, Disposable pipette tips, Micropipettes, Centrifuge for microcentrifuge tubes, Heating block, Vortex mixer, Personal protection equipment (lab coat, gloves, goggles)

Storage

MQ PCR/Gel Product Purification Kit should be stored dry at 15°C - 25°C. It can be stored for up to 24 months without showing any reduction in performance and quality.

Features

MQ PCR/Gel Product Purification Kit are designed for extraction of DNA from agarose gels and for purification of PCR products.

Parameter	Gel Extraction	PCR Clean-up
Maximum Sample Volume	300 mg gel slice	100 µl PCR solution
Gel	< 2,5% TAE or TBE	
Typical Recovery	70-80%	80-90%
Binding Capacity	10 µg	10 µg
DNA Fragment Length	50 bp – 10 kbp	50 bp – 10 kbp
Effective Primer Removal		< 25 bp
Elution Volume	20-50 µl	20-50 µl
Operation Time	20 minutes	20 minutes
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Procedure for PCR Product Clean-up

Before starting the preparation please check if Wash Buffer GP2 was prepared with ethanol as mentioned in Preparation Section. Unless otherwise noted the centrifugation steps are carried out at 13,000 rpm (\sim 11,000 – 18,000 x g) in a conventional, table-top microcentrifuge.

1. Sample Preparation

Mix 1 volume of sample (up to 100 μl) with 5 volumes of Binding Buffer GP1 by vortexing (e.g. mix 40 μl PCR reaction and 200 μl Binding Buffer GP1).

Note:

For sample volumes less than 40 μ l adjust the volume of the sample to 40 μ l using Binding Buffer GP1 or PCR grade water first. Then add 200 μ l Binding Buffer GP1 and mix by vortexing. If the volume of the used PCR reaction tube is not sufficient, please transfer the PCR sample into a microcentrifuge tube before adding Binding Buffer GP1.

2. DNA Binding

- Place a GP Column into a Collection Tube.
- Apply the sample mixture from previous step into the GP Column and centrifuge at 13,000 rpm for 30 seconds.
- Discard the flow-through and place the GP Column back into the Collection Tube.

<u>3. Wash</u>

- Add 600 µl Wash Buffer GP2 to the GP Column and centrifuge at 13,000 rpm for 30 seconds.
- Discard the flow-through and place the GP Column back into the Collection Tube.

<u>4. Dry</u>

• Centrifuge again for 2 minutes at 13,000 rpm to dry the column matrix.

5. DNA Elution

- Transfer GP Column into a new microcentrifuge tube (not provided).
- Add 20-50 µl Elution Buffer GP3 to the center of the column matrix.
- Allow to stand for 2 minutes until Elution Buffer is absorbed by the matrix.
- Centrifuge at 13,000 rpm for 2 minutes to elute purified DNA.

Note:

Yield of larger fragments (>5 kbp) can be increase by using pre-warmed (70°C) Elution buffer.

Procedure for DNA extraction from agarose gels

Before starting the preparation please check if Wash Buffer GP2 was prepared with ethanol as mentioned in Preparation Section.

Unless otherwise noted the centrifugation steps are carried out at 13,000 rpm (\sim 11,000 – 18,000 x g) in a conventional, table-top microcentrifuge.

1. Gel Dissociation

- Take a Gel Band Cutter or a clean scalpel to excise the DNA fragment from an agarose gel (< 2.5% agarose concentration recommended). Remove extra agarose to minimize gel slice.
- Transfer up to 300 mg of gel slice into a microcentrifuge tube (not provided).
- Add 500 µl of Binding Buffer GP1 to the sample and mix by vortexing.
- Incubate at 55°C for 10-15 minutes until the gel slice has been completely dissolved. During incubation, invert the tube every 2-3 minutes.

2. DNA Binding

- Place a GP Column into a Collection Tube.
- Apply up to 800 µl of the sample mixture from previous step into the GP Column and centrifuge at 13,000 rpm for 30 seconds.
- Discard the flow-through and place the GP Column back into the Collection Tube.

Note:

If the sample mixture is more than 800 μ l, repeat the DNA biding steps.

<u>3. Wash</u>

- Add 600 µl Wash Buffer GP2 to the GP Column and centrifuge at 13,000 rpm for 30 seconds.
- Discard the flow-through and place the GP Column back into the Collection Tube.

Note:

For TAE gels proceed to the next step. For TBE gels we recommend to repeat Wash steps to remove Boric Acid completely.

<u>4. Dry</u>

• Centrifuge again for 2 minutes at 13,000 rpm to dry the column matrix.

5. DNA Elution

- Transfer GP Column into a new microcentrifuge tube (not provided).
- Add 20-50 µl Elution Buffer GP3 to the center of the column matrix.
- Allow to stand for 2 minutes until Elution Buffer is absorbed by the matrix.
- Centrifuge at 13,000 rpm for 2 minutes to elute purified DNA.

Note:

Yield of larger fragments (>5 kbp) can be increase by using pre-warmed (70°C) Elution buffer.

For Laboratory Use Only