EcoPURE PCR/Gel Purification Kit

50 rxns

Cat No: E5003

Shipping : Ship at ambient temperature.
Storage : Store kit between 15°C and 25°C.

General Information

EcoPURE PCR/Gel Purification Kit combines 2 kits in 1. It is designed for effective and fast purification of polymerase chain reaction (PCR) products. Using this kit, primer dimers, free nucleotides in the reaction, salts, and Taq polymerase can be easily removed. This kit is also suitable for purification of nucleic acids from reactions including restriction digestion, alkaline phosphatase treatment, or kinase reactions.

Kit Contents

EcoPURE Binding Buffer	(25 ml)
EcoPURE Wash Buffer*	(8 ml concentrate)
EcoPURE Elution Buffer	(10 ml)
EcoPURE Columns	(50)
EcoPURE Collection Tubes	(50)

*Add 32 ml absolute ethanol

Additional Equipment & Reagents Required

96–100% ethanol
100% isopropanol
Tabletop microcentrifuge achieving >12,000 rpm
1.5 ml, sterile microcentrifuge tubes

Protocol for PCR purification

Each isolation procedure is suitable for purification of 50 µl PCR product. If the volume of PCR sample is less than 50 µl, adjust total volume for each PCR tube to 50 µl. If the volume of PCR sample is larger than 50 µl, either increase the amount of Binding Buffer (Step 1) proportionally, or divide the sample into 50 µl aliquots.

1. Add 250 µl *EcoPURE* Binding Buffer to each 50 µl PCR product.

2. Add 100 µl isopropanol to the mixture from step 1 and mix well.

Important: If the PCR mixture contains primer-dimers, purification might be performed without isopropanol, however, the yield of the target DNA fragment will be lower.

3. Insert an *EcoPURE* Column into a Collection Tube and transfer the sample from step 2 to the *EcoPURE* Column.

4. Centrifuge at maximum speed in a tabletop microcentrifuge 30 seconds at room temperature.

5. Discard the flowthrough and add 700 µl *EcoPURE* Wash Buffer to the *EcoPURE* Column.

6. Centrifuge at maximum speed in a tabletop microcentrifuge 30 seconds at room temperature.

7. Discard the flowthrough and centrifuge the empty *EcoPURE* Column at maximum speed for additional 1 minute to completely remove any residual wash buffer.

8. Transfer the *EcoPURE* Column to a clean 1.5 mL microcentrifuge tube (not included).

9. Add 30-50 µL of *EcoPURE* Elution Buffer to the center of the *EcoPURE* Column membrane and incubate the column at room temperature for 1 minute.

10. Centrifuge at maximum speed in a tabletop microcentrifuge 30 seconds at room temperature.

11. Discard the *EcoPURE* Column and store the purified DNA at -20°C.

For further information;
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Protocol for GEL Purification

Each isolation procedure in the following protocol is described from 100 mg gel containing DNA products. If the weight of gel slice is larger than 100 mg, increase the amount of Binding Buffer (Step 1) proportionally.

1. Load PCR reaction mixture on a 0.8 – 2% agarose gel and run your sample in a 1x TAE or 1x TBE running buffer. Electrophoresis until DNA band of interest is separated from adjacent contaminating fragments

2. Cut desired DNA band from gel using an ethanol-cleaned scalpel or razor blade.

3. Determine the mass of a sterile 1.5 ml microcentrifuge tube and place excised agarose gel slice in it. Measure the gel mass by re-weighting the tube with the excised gel slice.

4. Add 300 µl *EcoPURE* Binding Buffer to each 100 mg agarose gel slice in the microcentrifuge tube.

5. To dissolve agarose gel slice, incubate the suspension for 10 minutes at 55°C. Vortex the tube briefly every 2 – 3 minutes during incubation in order to release the DNA.

6. After the agarose gel slice is completely dissolved, add 150 µl isopropanol for every 100 mg agarose gel slice to the mixture from step 5 and mix well.

7. Insert an *EcoPURE* Column into a Collection Tube and transfer the sample from step 6 to the *EcoPURE* Column.

8. Centrifuge at maximum speed in a tabletop microcentrifuge 30 seconds at room temperature.

9. Discard the flowthrough and add 700 µl *EcoPURE* Wash Buffer to the *EcoPURE* Column.

10. Centrifuge at maximum speed in a tabletop microcentrifuge 30 seconds at room temperature.

11. Discard the flowthrough and centrifuge the empty *EcoPURE* Column at maximum speed for additional 1 minute to completely remove any residual wash buffer.

12. Transfer the *EcoPURE* Column to a clean 1.5 mL microcentrifuge tube (not included).

13. Add 30-50 µL of *EcoPURE* Elution Buffer to the center of the *EcoPURE* Column membrane and incubate the column at room temperature for 1 minute.

14. Centrifuge at maximum speed in a tabletop microcentrifuge 30 seconds at room temperature.

15. Discard the *EcoPURE* Column and store the purified DNA at -20°C.

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