

HiYield Gel/PCR DNA **Fragments Extraction Kit 2.0**

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Cat. No. SYG206-001 FYG206-100 FYG206-300

HiYield Gel/PCR DNA Fragments Extraction Kit 2.0

(SYG206-001/FYG206-100/FYG206-300) Ver. Q0308

Sample: 100 µl PCR Product, 300 mg of Agarose Gel

Store at RT/4°C Yield: Up to 50 μg

Contents

	SYG206-001	FYG206-100	FYG206-300
EZ Buffer	2 ml	60 ml	80 ml x2
W1 Buffer	2 ml	45 ml	125 ml
W2 Buffer	300 μl x2	15 ml	25 ml x2
Elution Buffer	1 ml	10 ml	30 ml
EZ Column	4 pcs	100 pcs	300 pcs
Collection Tube	4 pcs	100 pcs	300 pcs
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Buffer Preparation

• Add ethanol (96-100%) to the W2 Buffer prior to first use

	SYG206-001	FYG206-100	FYG206-300
W2 Buffer	300 μl x2	15 ml	25 ml x2
Ethanol (96 ~ 100%)	1.2 ml x2	60 ml	100 ml x2

Important Notes

- 1. Buffers provided in this system contain irritants. Wear gloves and lab coat when handling these buffers.
- 2. Add ethanol (96- 100 %) to W2 Buffer when first open.
- 3. Prepare dry bath or water bath before the operation.
- 4. Resolve any precipitate by warming at 37°C.

Purification Protocols

I. Gel Extraction:

Step 1. Gel Dissociating

- a. Excise the agarose gel slice containing relevant DNA Fragments.
- b. Transfer up to 300 mg (**do not over 300 mg**) of the gel slice into a microcentrifuge tube (not provided).
- c. Add 500 µl EZ Buffer to the sample and mix by vortexing.
- d. Incubate at 60 °C for 10 minutes until the gel slice has been completely dissolved. During incubation, invert the tube every 2-3 minutes.
- e. Cool down the dissolved sample mixture to room temperature slowly.

Step 2. DNA Binding

- a. Place a EZ Column in a Collection Tube.
- b. Apply 800 µl of the sample mixture into the EZ Column.
- c. Centrifuge at 14,000 x g for 30 seconds.
- d. Discard the flow-through and place the EZ Column back in the Collection Tube.

Note: If the sample mixture is more than 800 µl, repeat this DNA Binding Step.

Step 3-1. Wash

- a. Add 400 µl W1 Buffer to EZ Column.
- b. Centrifuge at 14,000 x g for 30 seconds.
- c. Discard the flow-through and place the EZ Column back into the Collection Tube.

Step 3-2. Wash

- a. Add 600 µl W2 Buffer (ethanol added) into the EZ Column.
- b. Centrifuge at 14,000 x g for 30 seconds. Discard the flow-through and place EZ Column back in the Collection Tube.

Step 4. Dry

a. Centrifuge again for 2 minutes at 14,000 x g to dry the column matrix.

Step 5. Elution

- a. Transfer dried EZ Column into a new microcentrifuge tube (not provided).
- b. Add 50 90 μ l Elution Buffer into the center of the column matrix. (If DNA is larger than 5 kb, use preheated 60 °C Elution Buffer to improve the elution efficiency.)
- c. Stand for 2 minutes until Elution Buffer is absorbed by the matrix.
- d. Centrifuge for 2 minutes at full speed to elute purified DNA.

Step 6. Store DNA

a. Store DNA at 4 °C or -20 °C.

II. PCR Clean Up:

Step 1. Sample preparation

a. Add 500 µl EZ Buffer to 100 µl PCR product and mix by vortexing.

Step 2. DNA Binding

- a. Place a EZ Column in a Collection Tube.
- b. Apply 800 μl of the sample mixture into the EZ Column.
- c. Centrifuge at 14,000 x g for 30 seconds.
- d. Discard the flow-through and place the EZ Column back in the Collection Tube.

Note: If the sample mixture is more than 800 µl, repeat this DNA Binding Step.

Step 3-1. Wash

- a. Add 400 µl W1 Buffer to EZ Column.
- b. Centrifuge at 14,000 x g for 30 seconds.
- c. Discard the flow-through and place the EZ Column back into the Collection Tube.

Step 3-2. Wash

- a. Add 600 µl W2 Buffer (ethanol added) into the EZ Column.
- b. Centrifuge at 14,000 x g for 30 seconds. Discard the flow-through and place EZ Column back in the Collection Tube.

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Step 6. Store DNA

a. Store DNA at 4 °C or -20 °C.

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