

Genomic DNA Extraction kit (Cultured Cell) 2.0

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Ver. Q0307

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Cat. No. SYG117-001 FYG117-100 FYG117-300

Genomic DNA Extraction kit (Cultured Cell) 2.0

SYG117-001/FYG117-100/FYG117-300

Store at RT/4 °C Ver. Q0307

Sample : 10^7 Cultured Cell Yield: Up to 50 μg

Contents

	SYG117-001	FYG117-100	FYG117-300
N1 Buffer	4 ml	100 ml	100 ml x3
N2 Buffer	1.5 ml	35 ml	95 ml
N3 Buffer	2 ml	45 ml	125 ml
W1 Buffer	2 ml	45 ml	125 ml
W2 Buffer	300 ul x2	15 ml	25 ml x2
Elution Buffer	1 ml	10 ml	30 ml
GZ 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

	SYG117-001	FYG117-100	FYG117-300
W2 Buffer	300 ul x2	15 ml	25 ml x2
Ethanol (96 ~ 100%)	1.2 ml x2	60 ml	100 ml x2

Additional Requirements

- 1. Ethanol (96-100%)
- 2. RNase A(Optional)
- 3. Microcentrifuge tubes

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 Protocol:

Step 1. Sample Preparation

- a. Transfer up to 10⁷ cultured cells to a microcentrifuge tube (not provided).
- b. Centrifuge 1 minute at 6,000 x g. Remove the supernatant completely.
- c. Resuspend the cell with 50 µl N1 Buffer by pipetting.

Step 2. Cell Lysis

- a. Add 300 μ l N2 Buffer to the sample and mix thoroughly by vortexing.
- b. Incubate at 60 °C for 10 mins until the sample lysate is clear.
- **c.** Invert the tube every 3 minutes during incubation.

Optional Step.

If RNA-free genomic DNA is required, perform this optional step.

Add 5 µl of RNase A (10 mg/ ml, not provided) to sample lysate and mix by vortexing. Incubate at room temperature for 5 mins.

Step 3. Protein Removal

- a. Add 400 μl N3 Buffer to the sample and shake vigorously.
- b. Centrifuge 1 minute at 12,000 x g.

Step 4. DNA Binding

- a. Place a GZ column in Collection Tube.
- b. Transfer the supernatant to GZ column and centrifuge 30 seconds at 14,000 x g.
- c. Discard the flow-through and place GZ Column back in the Collection Tube.

Step 5. Washing

- a. Add 400 µl of W1 Buffer into the GZ Column.
- b. Centrifuge at full speed (14,000 x g) for 1 minute.
- c. Discard the flow-through and place the **GZ Column** back in the **Collection Tube**.
- d. Add 600 µl W2 Buffer (Ethanol added) in the GZ Column.
- e. Centrifuge at full speed (14,000 x g) for 1 minute.
- f. Discard the flow-through and place the **GZ Column** back in the **Collection Tube**.

Step 6. Dry column

a. Centrifuge at full speed $(14,000 \times g)$ for 3 minutes to dry the column.

Step 7. Elution

- a. Place GZ Column to a clean 1.5 ml microcentrifuge tube.
- b. Add **50-90 µl of preheated Elution Buffer** (70°C) into the center of the column matrix.
- c. Stand at room temperature for 3 minutes.
- d. Centrifuge at full speed (14,000 x g) for 2 minutes to elute purified DNA.

Step 8. Store DNA

a. Store the DNA fragment at 4°C or -20°C.

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