

PureHelix™ *Genomic DNA Prep Kit [Yeast]* (Column Type) - Ver. 3.0

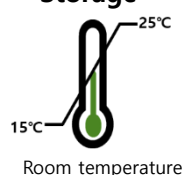
Kit contents

PureHelix™ <i>Genomic DNA Prep Kit [Yeast]</i> – Ver. 3.0			
Cat. No.	GCY50 (50preps)	GCY100 (100preps)	GCY200 (200preps)
H1	5ml	10ml	20ml
NGD1	15ml	30ml	60ml
NPS	15ml	30ml	60ml
EB	5ml	10ml	20ml
WB	11ml (Add 44ml ethanol)	22ml (Add 88ml ethanol)	44ml (Add 176ml ethanol)
MaxBinder™ Solution	5ml	10ml	20ml
Lyticase Suspension Solution	0.5ml	0.5ml	0.5ml
Lyticase (2.5unit/μl)	70μl (Dry)	70μl (Dry) x 2ea	70μl (Dry) x 4ea
RNase A (50mg/ml)	0.15ml (Dry)	0.15ml (Dry) x 2ea	0.15ml (Dry) x 4ea
Proteinase K (10mg/ml)	0.5ml (Dry)	0.5ml (Dry) x 2ea	0.5ml (Dry) x 4ea
Column set (with cap, 50ea/ Blue Box)	1box	2box	4box
Instructions for Use	1ea	1ea	1ea

Description

PureHelix™ *Genomic DNA Prep Kit [Yeast]* is designed for rapid and high purity isolation of genomic DNA from yeast cells. The spin column based method completely removes PCR inhibitors such as divalent cations and proteins resulting in a high purity preparation of genomic DNA. There is no use of phenol and chloroform, handling is safe and does not produce any harmful waste. DNA purified with this kit is suitable for a variety of applications, including PCR amplification, digestion with restriction endonucleases and genome sequencing.

Storage



Shelf life



Quality Control

Each lot of **PureHelix™ Genomic DNA Prep Kit [Yeast]** was tested against predetermined specifications to ensure consistent product quality.

Protocol

<Yeast>

It is essential to use the correct amount of starting material in order to obtain optimal DNA yield and purify. Amount of 2×10^7 yeast cells can generally be processed. Collect cultured cells and place on ice until use. Cultured cells can be used either fresh or frozen.

Important things to do before starting

- Before using **WB**, add **absolute ethanol** according to the bottle label to obtain a working solution. You may use 80% ethanol, instead of WB. Ethanol does not supplied in this kit.
- Add **70 µl** of **Lyticase suspension solution** into the **Lyticase** tube to made **2.5U/µl** concentration, and then store at -20°C.
- Add **0.5 ml** of **distilled water** to the provided **Proteinase K** tube for making **10mg/ml** concentration, and then store at -20°C.
- Add **0.15 ml** of **distilled water** to the provided **RNase A** tube for making **50mg/ml** concentration, then store at -20°C.

1. Cell Lysis

- 1) Harvest 500 µl of cultured yeast cells (**containing $1 \sim 2 \times 10^7$**) by centrifuge at 10,000 rpm for 1min. Discard the supernatant.
- 2) Add **100 µl of H1** and **1 µl of Lyticase (2.5U/µl)** to the cell pellet. Vortex vigorously for 10 sec.
- 3) Incubate for 15 min at 37°C and centrifuge at 12,000 rpm for 1 min. Discard the supernatant.
- 4) Add **300 µl of NGD1** and **2 µl of RNase A (50mg/ml)** to the cell pellet. Vortex vigorously for 10 sec.
- 5) Add **8 µl of Proteinase K (10mg/ml)** and mix by pipetting. Incubate at 60°C for 10 min, and then cool the tube on ice for 5 min.

- 6) Add **300 µl of NPS**. Vortex briefly.
- 7) Place the tube on ice for 5 min, and centrifuge for 5 min at 12,000 rpm.
- 8) Transfer **700 µl of the supernatant** into a clean 1.5ml tube.
- 9) Add **200 µl of absolute ethanol** and vortex vigorously.

2. Column Activation [Optional]

※ **These steps are required for the best yield.**

- 1) Place a Spin Column into a 2 ml collection tube.
- 2) Add **100 µl of MaxBinder™ Solution** into the Spin Column.
- 3) Centrifuge at 12,000 rpm for 30 sec and immediately proceed to next step.
You need not discard the flow-through from the collection tube.

3. Loading

- 1) Pipet **400 µl of the mixture** from step 1 (Cell Lysis) into a spin column sitting in a 2 ml collection tube.
- 2) Centrifuge for 30 sec at 12,000 rpm. Discard the flow-through.
- 3) Pipet **the remains of the mixture** from step 1 into the spin column.
- 4) Centrifuge for 30 sec at 12,000 rpm. Discard the flow-through.

4. Washing

- 1) Add **500 µl of WB** (80% ethanol) into the spin column.
- 2) Centrifuge for 30 sec at 12,000 rpm. Discard the flow-through.
※ **Repeat these steps for high-purity DNA preparation.**
- 3) Centrifuge at 12,000 rpm for 2 min to remove residual ethanol.

5. Elution

- 1) Discard the 2 ml collection tube and carefully place the spin column in a clean 1.5 ml tube.
Add **40-50 µl of EB or distilled water** into the center of the column.
- 2) Centrifuge at 12,000 rpm for 2 min. Discard the spin column.
Store the eluted DNA at 4°C or -20°C.

Products

Cat. No.	Products	Size
GCY50	PureHelix™ <i>Genomic</i> DNA Prep Kit [Yeast] - Ver.3.0 (Column type)	50preps
GCY100	PureHelix™ <i>Genomic</i> DNA Prep Kit [Yeast] - Ver.3.0 (Column type)	100preps
GCY200	PureHelix™ <i>Genomic</i> DNA Prep Kit [Yeast] - Ver.3.0 (Column type)	200preps