

High-quality and Highly Scalable Cell DNA Extraction using GenEx™ Cell, the Solution-type Genomic DNA Extraction Kit

Experimental Conditions

Materials Required

- GenEx™ Cell Sx (100 preps: 221-101 / 500 preps: 221-105)
- 200 U of lyticase or 20 U of zymolase (for yeast cell lysis)
- 30 mg/ml of lysozyme or 300 µl/ml of lysostaphin (for gram-positive cell lysis)
- Ice (for incubation or maintaining the normal state of the enzyme solution and proteinase K solution)
- Microcentrifuge tube
- Microcentrifuge (≤15,000 x g)
- Vortex mixer
- Heating block
- 70% ethanol
- Isopropanol (≥99.5%, C₃H₈O, CAS No.: 67-63-0)
- Pipette & sterile pipette tips
- Suitable protector (e.g., lab coat, disposable gloves, goggles, etc.)

Sample Information

- 5 x 10⁶ of K562 (human erythroleukemia cell line)
- 2 x 10⁹ of DH5α (gram-negative bacteria)
- 2 x 10⁹ of *Lactobacillus* (gram-positive bacteria)
- 5 x 10⁷ of yeast

Protocol

Enzymatic pre-treatment of **lysozyme** or **lysostaphin** is required for gram-positive bacteria and yeast cell DNA extraction.

- Gram-positive bacteria: Steps 1–2 of Protocol L (page 38) in the Exgene™ Cell SV mini manual were followed.
- Yeast: Steps 1–4 of Protocol M (page 40) in the Exgene™ Cell SV mini manual were followed.

* For more details, please refer to the handbook of Exgene™ Cell SV mini and GenEx™ Cell Sx.

Sample Preparation and Protocol

K562

Protocol E (pages 24–25) in the handbook was referred to.

DH5α

Protocol G (pages 28–29) in the handbook was referred to.

Lactobacillus

Following enzymatic pre-treatment, the handbook was consulted starting from Step 3 of Protocol G (page 28).

Total yeast

Following enzymatic pre-treatment, the handbook was consulted starting from Step 2 of Protocol G (page 28).

Result

Kit	GenEx™ Cell Sx			
	Yield (µg)	A _{260/280}	A _{260/230}	CV (%)
K562	15.62	1.98	2.15	1.10
	15.51	2.01	2.21	
	15.29	1.98	2.18	
DH5α	14.92	1.98	2.19	0.85
	15.11	1.99	2.20	
	14.86	2.01	1.99	
<i>Lactobacillus</i>	12.26	1.98	2.19	3.78
	12.68	2.02	2.18	
	13.21	1.98	2.20	
Yeast	6.11	2.01	2.11	2.77
	6.35	1.98	2.19	
	6.02	2.03	2.20	

Table 1. Result of yield, purity and CV (coefficient of variation) of DNA extracted from 4 different samples using GenEx™ Cell Sx.

The DNA were extracted from four different samples using GenEx™ Cell. All eluates were analyzed with a absorbance using NanoDrop™ 2000. The absorbance was performed in triplicated. The yield and CV values were calculated based on the measured absorbance values.

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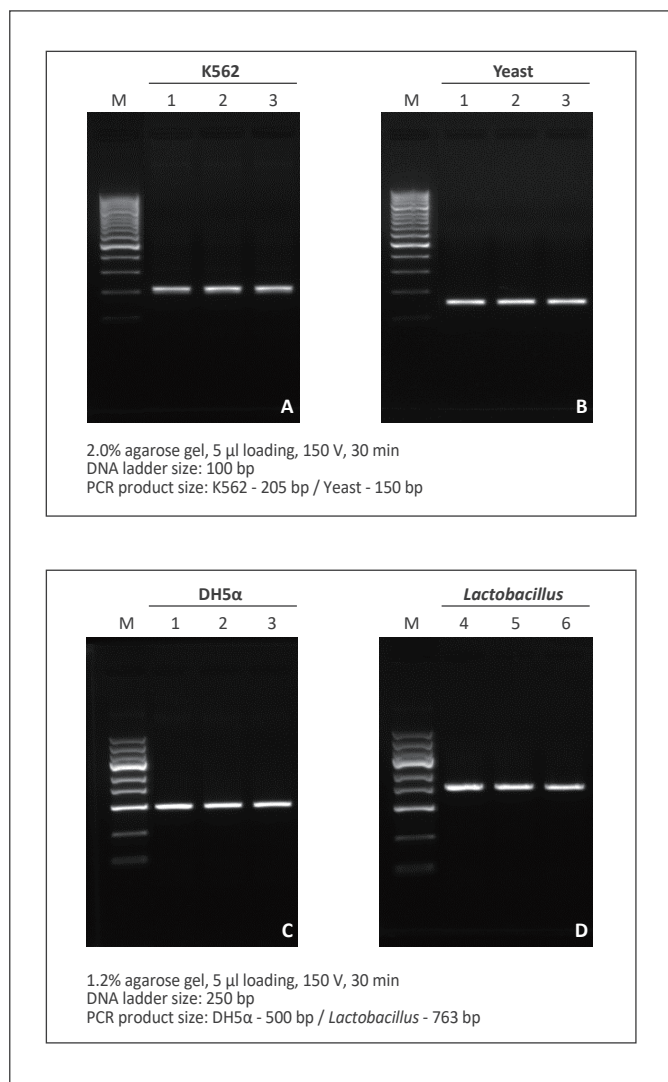


Figure 1. PCR amplification of DNA extracted from four different samples using GenEx™ Cell Sx.

PCR reaction was performed in triplicate with extracted DNA from four different samples using GenEx™ Cell Sx. Eluted PCR products were analyzed with gel electrophoresis using ethidium bromide staining.

- **PCR primer**

- A: K562 cells (human GAPDH primer)
- B: Total yeast (Scer primer)
- C: DH5α (bacteria universal primer)
- D: *Lactobacillus* (uvrC primer)

- **DNA ladder**

- Lane M: DNA ladder
- A, B: GENESTA™ 100 bp DNA ladder (GA-010)
- C, D: GENESTA™ 250 bp DNA ladder (GA-025)

- **PCR system and kit**

- MultiGene™ Optimax Thermal Cycler (TC9610, supplier L)
- 2X Taq PCR Master Mix (TAQ-OV-500R, supplier M)