Comparing Yield and Quality of Genomic DNA extracted from Four Different Cell Types

Experimental Conditions

Materials Required

- Exgene[™] Cell SV mini (106-101)
- Manual DNA extraction kit (Supplier : A)
- 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 (maintaining the normal state of the enzyme solution and Proteinase K solution)
- Microcentrifuge tube (for sample preparation)
- Microcentrifuge (≤15,000 x g)
- Vortex mixer
- Heating block
- Absolute ethanol (≥95.0%, C₂H₅OH, CAS No. 64-17-5)
- Pipette & sterile pipette tips
- Suitable protector (e.g., lab coat, disposable gloves, goggles, etc.)

Sample Information

- Sample type:
 - K562 (5 x 10⁶ cells), cultured cells
 - DH5α (2 x 109 cells), gram negative bacteria
 - Lactobacillus (2 x 109 cells), gram positive bacteria
 - Total yeast (5 x 10⁷ cells)
- Extraction conditions
 - Sample amount: according to each protocol
 - Elution volume : 50 μl

- Protocol

K562: The protocol is according to <u>A. Protocol for Blood and Body Fluid/Cultured Cells using Microcentrifuge</u> (page 18~20).

DH5 α : The protocol is according to <u>K. Protocol for Gram Negative Bacteria</u> (page 37) and then the next step is according to <u>G. Protocol for Animal Tissue</u> (step 3 on page 31).

Lactobacillus: The protocol is according to <u>L. Protocol for Gram Positive Bacteria</u> (page 38~39) and then the next step is according to <u>G. Protocol for Animal Tissue</u> (step 5 on page 31).

Total yeast: The protocol is according to M. Protocol for Yeast (page 40~41) and then the next step is according to G. Protocol for Animal <u>Tissue</u> (step 3 on page 31).

* For more details and methods, please refer to the handbook of Exgene™ Blood/ Clinic/Cell SV mini protocol.

Result

Kit	K562 (A)			
	Yield (ng/μl)	A260/280	A260/230	CV (%)
Exgene™ Cell SV	18.49	1.99	2.09	1.84
Supplier A	17.02	1.98	2.11	1.71

Kit	DH5α (B)			
	Yield (ng/μl)	A260/280	A260/230	CV (%)
Exgene™ Cell SV	17.19	1.99	2.08	3.19
Supplier A	15.93	1.98	2.06	1.98

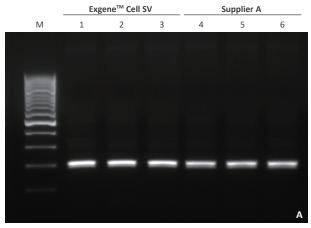
Kit	Lactobacillus (C)			
	Yield (ng/μl)	A260/280	A260/230	CV (%)
Exgene [™] Cell SV	15.04	1.99	2.11	0.68
Supplier A	13.57	1.98	2.14	2.58

Kit	Yeast (D)			
	Yield (ng/μl)	A260/280	A260/230	CV (%)
Exgene [™] Cell SV	11.14	2.01	2.07	2.09
Supplier A	9.95	2.00	2.07	1.03

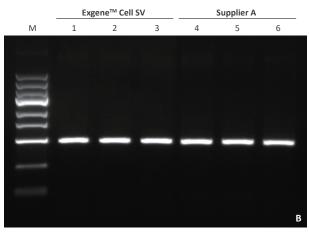
Table 1. Comparison of average yield, purity and CV (coefficient of variation) values of DNA extracted from 4 samples using each DNA extraction kit

The DNA were extracted from four samples using Exgene™ Cell SV mini and Supplier A's DNA extraction kit. All eluates were analyzed with a absorbance using NanoDrop™ 2000. The absorbance was performed in triplicated and then the results were averaged. The yield and CV values were calculated based on the measured absorbance values.

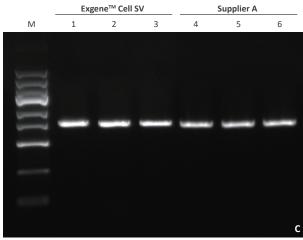
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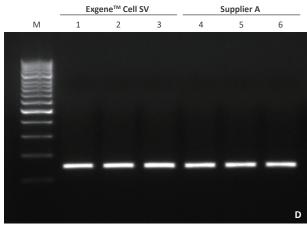
2.0% agarose gel, 5 μ l loading, 150 V, 30 min DNA ladder size : 100 bp PCR product size : 205 bp



1.2% agarose gel, 5 µl loading, 150 V, 30 min DNA ladder size : 250 bp PCR product size : 500 bp



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2.0% agarose gel, 5 μl loading, 150 V, 30 min DNA ladder size : 100 bp PCR product size : 205 bp

Figure 1. Comparison of gel-electrophoresis of DNA extracted from four samples using each DNA extraction kit

After DNA templates were extracted from four samples with Exgene™ Cell SV mini and Supplier A's DNA extraction kit, All DNA templates were performed in triplicate with conventional PCR methods. Eluted PCR products were analyzed with gel-electrophoresis using ethidium bromide staining.

Figure and PCR primer information

Figure A : K562 cells, human GAPDH primer Figure B : DH5 α , bacteria universal primer Figure C : *Lactobacillus*, uvrC primer Figure D : Total yeast, Scer primer

Lane information

Lane M : GENESTATM 100 bp DNA ladder (GA-010) or GENESTATM 250 bp DNA ladder (GA-025)

Lane 1~3 : Exgene™ Cell SV mini

Lane 4~6: DNA Extraction kit from Supplier A

• PCR instrument and kit information

 $\label{eq:MultiGenet} \begin{tabular}{ll} MultiGene & Mostimax Thermal Cycler (TC9610, Supplier : L) \\ 2X Taq PCR Master Mix (TAQ-OV-500R, Supplier : M) \\ \end{tabular}$