

Genomic DNA Extraction from fecal samples using AllEx® Fecal DNA/RNA Kit

Experimental Conditions

Materials

- AllEx®64 Automated Nucleic Acid Extraction System
- AllEx® Fecal DNA/RNA Kit
- DNA extraction commercial kit for fecal (supplier A)

Sample & Extraction Information

Sample type	Feces (fecal)
Origin	Human or swine
Target	Microbial and host DNA/RNA
Sample amount	0.2 g
Elution volume	100 µl
AllEx® 64 Protocol	Fecal-P2
Operating time	20' 47"

Protocol

For more details and methods, please refer to [the manual of AllEx® Fecal DNA/RNA Kit](#).

Preparation of Proteinase K Solution

To obtain 20 mg/ml of proteinase K solution, add 2.4 ml of PK Storage Buffer to one bottle of lyophilized Proteinase K (48 mg), and gently invert to dissolve.

Sample Preparation

1. Place 0.2 g of each fecal sample to 2 ml Glass Bead Tube (provided).
2. Add 1 ml of Buffer FL and 25 µl of proteinase K solution (20 mg/ml). Vortex the mixture at least 1 min.
3. Incubate at 65°C for 5 min.
4. Centrifuge at 13,000 rpm ($\geq 10,000 \times g$) for 5 min.
5. Transfer 200 µl of the supernatant to 1st (or 7th) well.

* The standard protocol was adopted for supplier A's commercial kit.

Result

Human Fecal

Kit	AllEx® Fecal DNA/RNA		Supplier A	
	Mean	CV	Mean	CV
Yield (µg)	75.0	0.01	6.8	0.05
A ₂₆₀ /A ₂₈₀	1.99	0.01	1.85	0.01
A ₂₆₀ /A ₂₃₀	1.60	0.01	1.47	0.03

Swine Fecal

Kit	AllEx® Fecal DNA/RNA		Supplier A	
	Mean	CV	Mean	CV
Yield (µg)	30.3	0.08	4.1	0.18
A ₂₆₀ /A ₂₈₀	2.11	0.01	1.90	0.02
A ₂₆₀ /A ₂₃₀	2.07	0.01	1.20	0.14

Table 1. DNA yield and purity

Nucleic acid was extracted from human and swine fecal samples (N=4). The AllEx® Fecal DNA/RNA Kit demonstrated superior DNA yield and purity compared to the supplier A's manual kit, as measured by NanoDrop™ 2000.

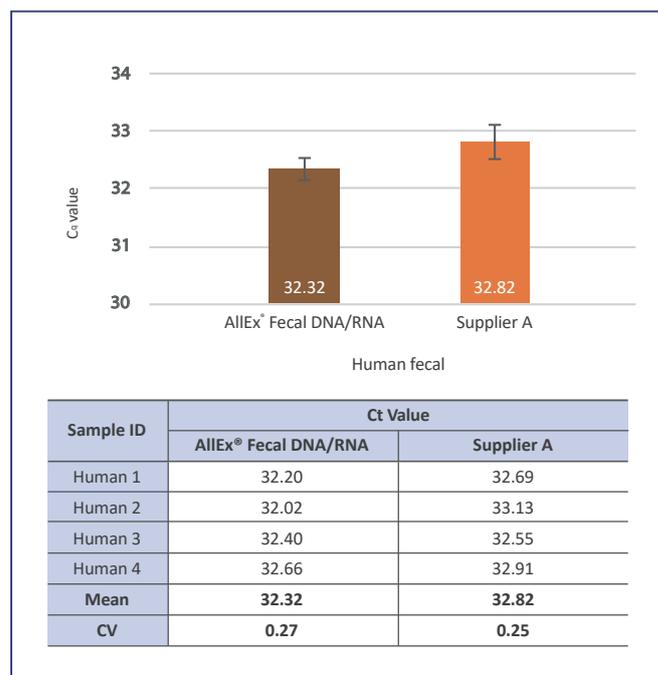


Figure 1. Real-time PCR performance with human fecal samples

qRT PCR data for human GAPDH gene (79 bp) were amplified from DNA extracts. qPCR was performed with HyperScript™ One-step RT-PCR Master Mix (602-110) on the CFX96™ System (1855201).

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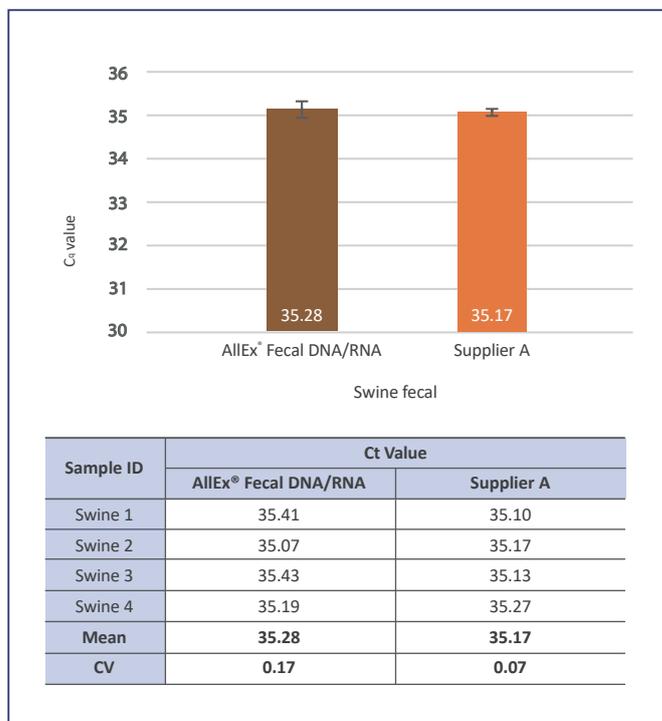


Figure 2. Real-time PCR performance with swine fecal samples
 qRT PCR data for swine GAPDH gene (150 bp) were amplified from DNA extracts. qPCR was performed with HyperScript™ One-step RT-PCR Master Mix (602-110) on the CFX96™ System (1855201).

Summary

- High-quality DNA were efficiently extracted from human and swine fecal samples using the AllEx®64 Automated Nucleic Acid Extraction System and AllEx® Fecal DNA/RNA Kit.
- Compared to supplier A's spin column-based manual kits, the AllEx® solution demonstrated equal or superior performance with the entire process, including pre-treatment, completed within 40 minutes.

Ordering Information

Cat. No.	Product	Size
AEX064	AllEx®64 Automated Nucleic Acid Extraction System	1 Unit
948-048	AllEx® Fecal DNA/RNA Kit	48 T
948-096	AllEx® Fecal DNA/RNA Kit	96 T