Genomic DNA Extraction from various samples using GENTi[™] Advanced Genomic DNA Extraction Kit on GENTi[™] Advanced

Experimental Conditions

Materials Required

- GENTi[™] Advanced Genomic DNA Extraction Kit (901-048A/901-096A)
- GENTi™ Advanced Automatic Extraction Equipment (GTI032A)
- 1X PBS (Phosphate-buffered saline), pH 7.4 (SM-P04-100)
- · Pipette & sterile pipette tips
- Suitable protector (e.g., lab coat, disposable gloves, goggles, etc.)

Sample Information

· Extraction conditions

| Sample | Amount | Elutoin volume | |
|-------------------|-------------------------|----------------|--|
| K562 cell | 1x10 ⁶ cells | - - 80 μl | |
| Human whole blood | 200 μΙ | | |
| Buccal swab | 1 stick | | |
| Urine | 200 μΙ | | |

Sample Preparation

- K562 cell
- 1. Transfer the harvested cells into a 1.5 ml microcentrifuge tube and centrifuge at 14,000 x g for 1 min.
- 2. Discard the supernatant and resuspend the cell pellet with 400 μl of 1X PBS.
- Follow the Protocol of GENTi™ Advanced Genomic DNA Extraction Kit.

· Human whole blood

- 1. Transfer 200 μ l of human whole blood in an EDTA tube or other anticoagulant mixture.
- 2. Apply the human whole blood contained in EDTA or other anticoagulant mixture to cartridge right now.
- 3. Follow the **Protocol of GENTi™ Advanced Genomic DNA Extraction Kit.**

· Buccal swab

- 1. Collect the oral epithelial cells using sterilized swab and cut off the head of swab using sterilized scissors.
- 2. Transfer the head of swab into a 2 ml microcentrifuge tube with 400–500 µl of 1X PBS and vortex vigorously.
- 3. Follow the **Protocol of GENTi™ Advanced Genomic DNA Extraction Kit.**

Urine

- 1. Transfer 200 μ l of urine into a 5 ml conical tube and centrifuge for 2 min at 6,000 x g above.
- 2. Discard the supernatant and resuspend with 3 ml of 1X PBS.
- 3. Follow the **Protocol of GENTi™ Advanced Genomic DNA Extraction Kit.**

Protocol

GENTi™ Advanced Genomic DNA Extraction Kit Protocol

- * For more details, please refer to the manual of GENTi™ Advanced Genomic DNA Extraction Kit.
- 1. Peel back the seal of pre-filled with reagents cartridge.
- 2. Dispense 20 μ l of dissolved proteinase K solution into the 1st (7th) well.
- 3. Dispense 10 µl of RNase A solution into the 3rd (9th) well.
- 4. Dispense 200 μ l of prepared samples into the 1st (7th) well.
- 5. Load the plate onto the tray of GENTi™ 32 Advanced System.
- 6. Insert Magnetic Rod Cover to the end to bracket.

Result

| Sample | No. | Yield (μg) | A _{260/280} | A _{260/230} |
|-------------------|-----|------------|----------------------|----------------------|
| K562 cell | 1 | 19.2 | 2.11 | 2.08 |
| | 2 | 21.4 | 2.11 | 2.06 |
| | 3 | 19.4 | 2.18 | 2.21 |
| Human whole blood | 1 | 2.6 | 1.96 | 1.31 |
| | 2 | 2.2 | 1.85 | 1.24 |
| | 3 | 1.9 | 1.88 | 1.25 |
| Buccal swab | 1 | 0.5 | 2.54 | 0.42 |
| | 2 | 0.5 | 2.28 | 0.6 |
| Urine | 1 | 0.8 | 1.65 | 0.68 |
| | 2 | 0.5 | 1.12 | 0.7 |

Figure 1. Yield and purity analysis of DNA

DNA was extracted following the standard protocols for cell, blood, buccal swab, and urine using GENTi™ Advanced Genomic DNA Extraction kit. O.D. ratio and yield of extracted genomic DNA calculated using a NanoDrop™ 2000/2000C (supplier T).

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Result

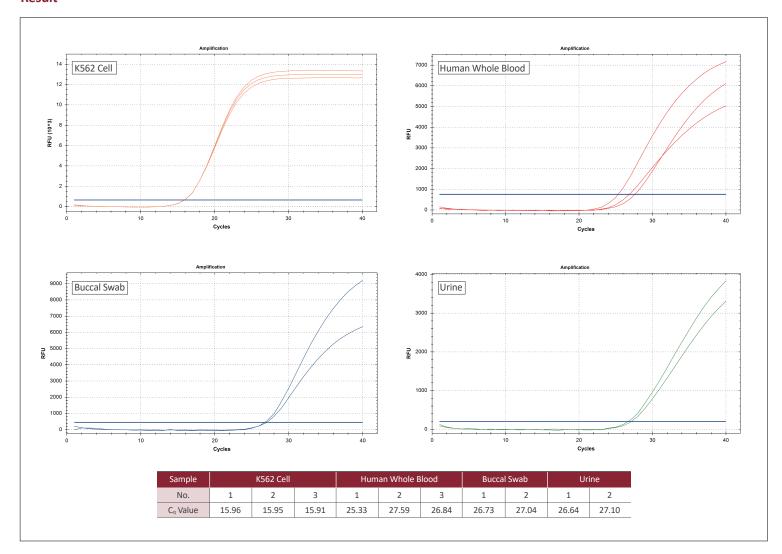


Figure 2. Real-time PCR of Genomic DNA Extraction using GENTi™ Advanced Genomic DNA Extraction Kit.
Genomic DNA extracted from cell, blood, buccal swab and urine using GENTi™ Advanced Genomic DNA Extraction Kit were used as templates for real-time PCR amplification of the GAPDH gene.

- PCR primer Human GAPDH
- Real-time PCR system and qPCR kit Real-time PCR system: CFX96™ System (1855201, supplier B) qPCR kit: Probe qPCR Mix (RR391AT)