

# Genomic DNA Extraction from Diverse Forensic Samples using GENTi™ Advanced Blood DNA Extraction Kit on GENTi™ 32 Advanced Automatic Extraction System

## Experimental Conditions

### Materials Required

- ♦ GENTi™ 32 Advanced Automatic Extraction System (GTI032A)
- ♦ GENTi™ Advanced Blood DNA Extraction Kit (903-048A/903-096A)
- ♦ Buffer CL (lysis for cigarette butt and fingerprint, 106-921)
- ♦ Proteinase K, 120 mg (392-103)
- ♦ PK Storage Buffer, 7 ml (392-903)
- ♦ Microcentrifuge & vortex mixer
- ♦ Thermomixer (e.g., HCM100-Pro, 5032103100, supplier: D)
- ♦ Pipette & sterile pipette tips
- ♦ Suitable protector (e.g., lab coat, disposable gloves, goggles, etc.)

### Sample Information

- ♦ Sample type and appropriate starting amount

Sample	Amount
Dried blood spot (DBS)	2 dots
Blood swab	1 ea
Cigarette butt	12 pieces of 2 cm <sup>2</sup>
Fingerprint	12 pieces of 2 cm <sup>2</sup>

- ♦ Extraction conditions
  - Extraction protocol: Blood\_High Protocol (operation time: 37' 07")
  - Elution volume: 70 µl

### Sample Preparation

#### • Dried blood spot (DBS)

1. Prepare human blood on the indicating FTA cards (DBS) collected within 3 days.
2. Collect the 5 mm diameter DBS samples using sterilized punching machine.
3. Transfer the two dots into 1st/7th well of the cartridge of GENTi™ Advanced Blood DNA Extraction Kit.

#### • Blood swab

1. Prepare human blood swab samples taken within 1 day.
2. Cut only the cotton part of swab and transfer it into 1st/7th well of the cartridge of GENTi™ Advanced Blood DNA Extraction Kit.

#### • Cigarette butt

1. Cut the outer paper of cigarette butt filter part into 12 equal parts of 2 cm<sup>2</sup> in size.
2. Transfer 12 pieces to a 2 ml microcentrifuge tube and add 300 µl of Buffer CL and 20 µl of Proteinase K solution (20 mg/ml).
3. Transfer the tube to a thermomixer and incubate at 1,500 rpm for 20 min at 56 °C.
4. Centrifuge at full speed for 1 min at room temperature.
5. Carefully collect 300 µl of the supernatant and transfer it into 1st/7th well of the cartridge of GENTi™ Advanced Blood DNA Extraction Kit.

#### • Fingerprint

1. Cut the fingerprinting into 12 equal parts of 2 cm<sup>2</sup> in size.
2. Transfer 12 pieces to a 2 ml microcentrifuge tube and add 300 µl of Buffer CL and 20 µl of Proteinase K solution (20 mg/ml).
3. Transfer the tube to a thermomixer and incubate at 1,500 rpm for 20 min at 56 °C.
4. Centrifuge at full speed for 1 min at room temperature.
5. Carefully collect 300 µl of the supernatant and transfer it into 1st/7th well of the cartridge of GENTi™ Advanced Blood DNA Extraction Kit.

\* All prepared samples are according to GENTi™ Advanced Blood DNA Extraction Kit protocol.

## Protocol

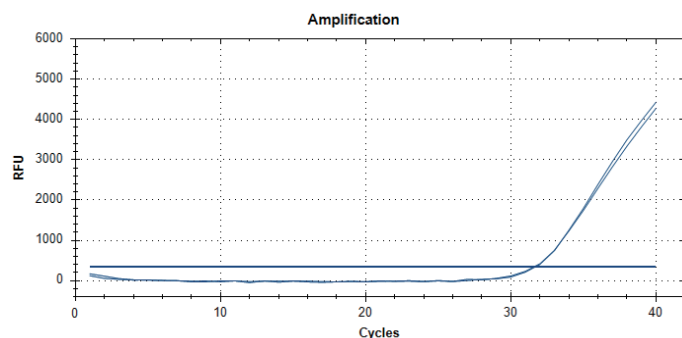
### GENTi™ Advanced Blood DNA Extraction Kit Protocol

\* For more details and methods, please refer to the handbook of GENTi™ Advanced Blood DNA Extraction Kit.

1. Add each of samples to 1st/7th well of the cartridge of GENTi™ Advanced Blood DNA Extraction Kit.
2. Add 15 µl of dissolved Proteinase K solution (2 µg/µl, not provided) to 1st/7th well.
3. Load the cartridge on the tray of GENTi™ 32 Advanced Automatic Extraction System.
4. Insert the magnetic rod cover to the end to strip bracket.
5. Select the correct extraction protocol (Blood\_High Protocol) and operate the extraction system.

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## Result

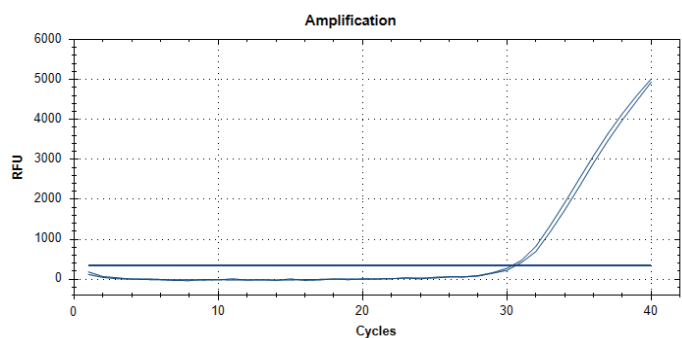


DBS	Yield (ng/μl)	A <sub>260/280</sub>	A <sub>260/230</sub>	C <sub>q</sub> Value
#1	3.8	1.81	1.13	31.72
#2	2.9	1.81	0.43	32.62

**Figure 1. Result of the yield and purity of genomic DNA extracted from dried blood spots and real-time PCR analysis using it as a template.**

Blood genomic DNA were extracted from dried blood spot with GENTi™ Advanced Blood DNA Extraction Kit on GTENTI™ 32 Automatic Extraction System. The quality and yield of eluted DNA were checked by measuring the absorbance and determining the A<sub>260/280</sub> and A<sub>260/230</sub> ratio using a spectrophotometer. Subsequently, DNA quantitation was performed in duplicate using a TaqMan-based real-time PCR assay (on CFX-96).

- Spectrophotometer: NanoDrop™ 2000/2000c (ND-2000)
- Real-time PCR system: CFX-96™ System (1855201)
- qPCR kit: Probe qPCR Mix (RR391A)
- Primer and probe: human GAPDH primer/probe

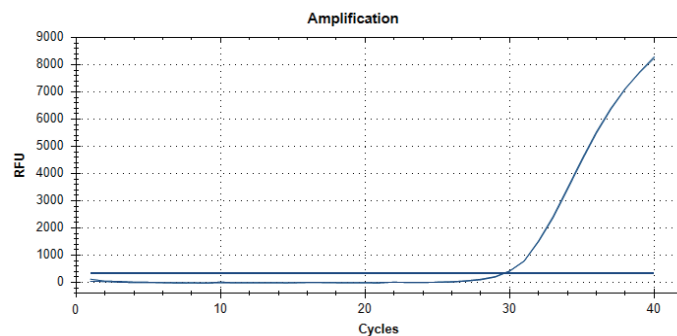


Blood swab	Yield (ng/μl)	A <sub>260/280</sub>	A <sub>260/230</sub>	C <sub>q</sub> Value
#1	5.5	2.24	0.89	30.65
#2	5.6	2.18	1.10	30.38

**Figure 2. Result of the yield and purity of genomic DNA extracted from blood swab and real-time PCR analysis using it as a template.**

Blood genomic DNA were extracted from blood swab with GENTi™ Advanced Blood DNA Extraction Kit on GTENTI™ 32 Automatic Extraction System. The quality and yield of eluted DNA were checked by measuring the absorbance and determining the A<sub>260/280</sub> and A<sub>260/230</sub> ratio using a spectrophotometer. Subsequently, DNA quantitation was performed in duplicate using a TaqMan-based real-time PCR assay (on CFX-96).

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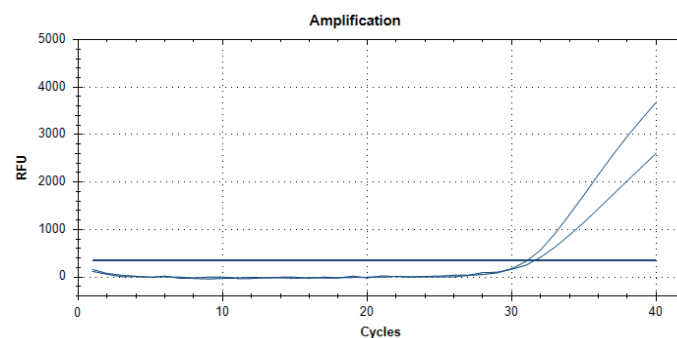


Cigarette butt	Yield (ng/μl)	A <sub>260/280</sub>	A <sub>260/230</sub>	C <sub>q</sub> Value
#1	5.3	2.05	0.92	29.67
#2	5.6	2.16	0.96	29.70

**Figure 3. Result of the yield and purity of genomic DNA extracted from cigarette butt and real-time PCR analysis using it as a template.**

Genomic DNA were extracted from lip epidermal cells present in cigarette butts with GENTi™ Advanced Blood DNA Extraction Kit on GTENTI™ 32 Automatic Extraction System. Buffer CL and Proteinase K solution were added to the samples prior to DNA extraction for optimal performance. The quality and yield of eluted DNA were checked by measuring the absorbance and determining the A<sub>260/280</sub> and A<sub>260/230</sub> ratio using a spectrophotometer. Subsequently, DNA quantitation was performed in duplicate using a TaqMan-based real-time PCR assay (on CFX-96).

- Spectrophotometer: NanoDrop™ 2000/2000c (ND-2000)
- Real-time PCR system: CFX-96™ System (1855201)
- qPCR kit: Probe qPCR Mix (RR391A)
- Primer and probe: human GAPDH primer/probe



Fingerprint	Yield (ng/μl)	A <sub>260/280</sub>	A <sub>260/230</sub>	C <sub>q</sub> Value
#1	2.9	2.14	0.94	31.07
#2	2.5	1.84	0.86	31.59

**Figure 4. Result of the yield and purity of genomic DNA extracted from fingerprint and real-time PCR analysis using it as a template.**

Genomic DNA were extracted from epidermal cells present in fingerprint with GENTi™ Advanced Blood DNA Extraction Kit on GTENTI™ 32 Automatic Extraction System. Buffer CL and Proteinase K solution were added to the samples prior to DNA extraction for optimal performance. The quality and yield of eluted DNA were checked by measuring the absorbance and determining the A<sub>260/280</sub> and A<sub>260/230</sub> ratio using a spectrophotometer. Subsequently, DNA quantitation was performed in duplicate using a TaqMan-based real-time PCR assay (on CFX-96).

- Spectrophotometer: NanoDrop™ 2000/2000c (ND-2000)
- Real-time PCR system: CFX-96™ System (1855201)
- qPCR kit: Probe qPCR Mix (RR391A)
- Primer and probe: human GAPDH primer/probe