

DA-c-8

Genomic DNA Extraction from Paraffin-embedded Samples

Protocol 1 (using Xylene for deparaffinization)

Microtubes (1.5 ml)

sliced FFPE block of mouse fetus : 10 µm x 3

← Xylene : 1000 μl

Vortex (maximum speed): 15 sec

→ 15,000 rpm, 2 min, RT

Remove the supernatant

→ >99% ethanol : 1000 μl

Vortex (maximum speed): 15 sec

★ 15,000 rpm, 2 min, RT

Remove the supernatant and dry up at RT: 10 min

← 0.075M KCI : 100 μI

→ 0.5% Tween20 : 20 µl

▼ MDT : 180 μl

← EDT : 40 μl

Vortex (maximum speed): 15 sec

Flash spin down



Incubation at 50°C: 3 hrs

── 15,000 rpm, 5 min, 4°C

Transfer the supernatant into the new 1.5 ml microtubes



Incubation at 90°C: 1 hr

── 15,000 rpm, 35 min, 4°C

Transfer the supernatant into the new 1.5 ml microtubes

↓ LDT : 180 µl

Vortex (maximum speed): 15 sec

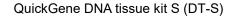
→ >99% ethanol : 240 μl

Vortex (maximum speed): 15 sec

Flash spin down









Transfer all contents of the micro tube into the cartridge of QuickGene



Refer to the extraction protocol for each device written in the kit handbook. (from the step after transferring the lysate into the cartridge)

Genomic DNA (Elution volume : 100 μl)





Protocol 2 (not using Xylene for deparaffinization)

Microtubes (1.5 ml)

■ sliced FFPE block of mouse fetus: 10 µmx 3

→ Tween20 : 50 µl

Tap the tube 5 times

Flash spin down

Incubation at 95°C: 10 min

→

Incubation at 65°C: 10 min

← MDT : 180 μl **←** EDT : 20 μl

Mix by pipetting using 1000ul micro pipet

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Incubation at 65°C: over night

── 15,000 rpm, 4 min, 4°C

Transfer the supernatant into the new 1.5 ml microtubes

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Incubation at 90°C: 1 hr

── 15,000 rpm, 35 min, 4°C

Transfer the supernatant into the new 1.5 ml microtubes

← LDT : 180 μl

Vortex (maximum speed): 15 sec

→ >99% ethanol : 240 μl

Vortex (maximum speed): 15 sec

Flash spin down

Lysate

Transfer all contents of the micro tube into the cartridge of QuickGene

and for each device written in the kit

Refer to the extraction protocol for each device written in the kit handbook. (from the step after transferring the lysate into the cartridge)

Genomic DNA (Elution volume : 100 μl)





Results

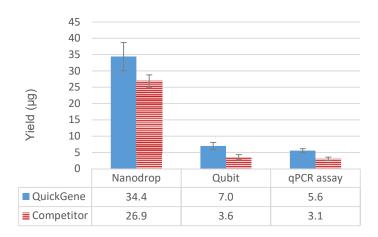
Electropherogram

No Data

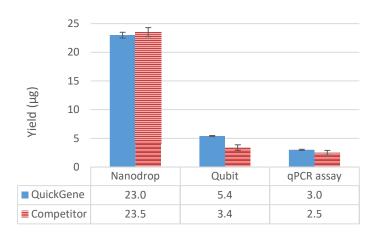
The yield of genomic DNA

The isolated DNA were quantified by nanodrop, Qubit and qPCR assay systems. qPCR assay was performed with TaqMan Gene Expression Assays.

<w/ Xylene (Protocol 1) >



<w/o Xylene (Protocol 2) >



KKURABO



Protein contamination: A260/280

The purity of isolated DNA were evaluated by nanodrop.

Sample	Protocol	Purity (A260/280)			Purity (A260/230)		
		No.1	No.2	No.3	No.1	No.2	No.3
QuickGene	1 (using Xylene)	2.01	2.02	2.02	2.22	2.24	2.25
Competitor Q	Using Xylene	2.06	2.02	2.03	2.09	2.11	2.07
QuickGene	2 (non Xylene)	2.00	1.99	1.99	2.20	2.16	2.16
Competitor Q	Non Xylene	2.04	2.02	2.02	2.06	2.09	2.12

Other

No Data

Common protocol is usable for the following

No Data

