2. Genomic DNA Extraction from Tissue of Animal

DA-b-1

Genomic DNA Extraction from Animal tissue (Rapid Method)

Protocol

2 ml micro tube

← Tissue 5~10 mg
(by scissors or hammer make it into small blocks)

← 4M TNES-UREA *1 : 200 µl

← EDT : 10 µl

Slowly shake until completely dissolved
(55°C × 2 hours use shaker)

← LDT : 180 µl

Vortex (maximum speed) : 15 sec

Flash spin down

Incubation at 70°C : 2 min

← >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

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 : 200 µl)

*1 <4M TNES-UREA>
10mM Tris-HCl, pH7.5
125mM NaCl
10mM EDTA
1% SDS
4M Urea
If the sample is difficult to dissolve, use 8M.

Results

No Data

Common protocol is usable for the following

No Data

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).

QuickGene

QuickGene DNA tissue kit S (DT-S)
QuickGene SP kit DNA tissue (SP-DT)

DA-b-2

Genomic DNA Extraction from Beef Fat

Protocol

2 ml micro tube

← Beef Fat : 5 mg *1
← MDT : 180 µl (Mix well)
← EDT : 10 µl

Tapping or pipetting 5 times

↓

Incubation at 55°C : 2 days (vortexing sometimes)

↻ 10,000 rpm, 10 min, 4°C *2

Transfer water layer to a new 1.5 ml tube, evading upper solid fat content.

← <Option> RNase A treatment *3
← LDT : 180 µl (Mix well)
← >99% ethanol : 240 µl

Vortex (maximum speed) : 15 sec (to make homogeneous solution)

Flash spin down.

↓

[Lysate]

↓

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 : 200 µl)]

*1 (Possibility up to 250 mg confirmed) Too much quantity causes clogging of cartridge or inadequate solution. Make sure to weigh until conditions have been settled.

*2 Alternate :
Centrifuge (10,000 rpm, room temp, 3 min)
Transfer water layer to a new 1.5 ml tube.
*only for 5 mg fat case

*3 Option :

↓ ← RNase A (100 mg/ml) : 20 µl

Tapping 5 times (Confirm enzyme solution is mixed.) Flash spin down and collect liquid on wall.

↓

React at RT for 2 min.

Results

The yield of genomic DNA

Starting tissue amount	Yield (µg)
250 mg	1.82
5 mg	0.47

Common protocol is usable for the following

No Data

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).

KURABO

QuickGene

QuickGene DNA tissue kit S (DT-S)
QuickGene SP kit DNA tissue (SP-DT)

DA-b-3

Genomic DNA Extraction from Kidney of Mouse

Protocol

2 ml micro tube

← Slice of mouse kidney : 5 mg
← MDT : 180 µl
← EDT : 20 µl

Incubate for over night on rotary shaker at 55°C, and dissolve the tissue completely

10,000 rpm, 3 min, RT

Transfer the supernatant to a 1.5 ml micro tube

← <Option> RNase A treatment *1
← LDT : 180 µl

Mix thoroughly by vortexing at maximum speed : 15 sec *2

Flash spin down

Incubate at 70°C for 10 min

Flash spin down.

← >99% ethanol : 240 µl

Mix thoroughly by vortexing at maximum speed : 15 sec *2

Flash spin down

Lysate

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 : 200 µl)

*1 Optioal steps
RNaseA : 20 µl
Tap the tube to mix the solution.
Flash spin down.
Set it down at room temperature for 2 min.

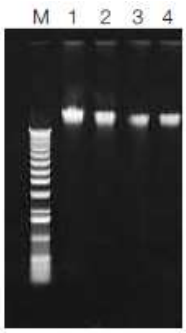
*2 Mix completely by vortexing at the maximum speed.
If the mixing is not enough by vortexing, use the tapping, pipetting or inverting.

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).

KURABO

Results

AGE of extracted genomic DNA from Mouse Tissue

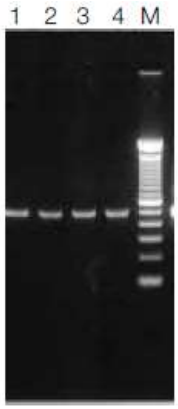


M : Size marker
1 : Lung tissue sample
2 : Kidney tissue sample
3 : Tail tissue sample
4 : Liver tissue sample

Other

・PCR

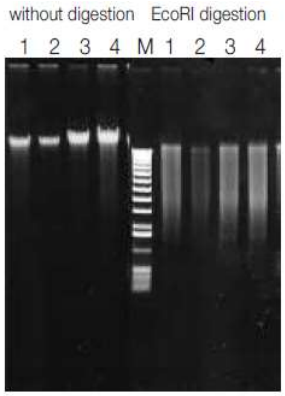
AGE of G3PDH PCR fragments amplified by genomic DNA extracted from mouse tissue using QuickGene isolation system and reagents



M : 100bp ladder marker
1 : Lung tissue sample
2 : Kidney tissue sample
3 : Tail tissue sample
4 : Liver tissue sample

・Restriction Enzyme Digestion

AGE of *Eco*RI restriction enzyme digestion fragments with genomic DNA extracted from mouse tissue using QuickGene isolation system and reagents



M : Size marker
1 : Tail tissue sample
2 : Liver tissue sample
3 : Lung tissue sample
4 : Kidney tissue sample

Common protocol is usable for the following

Mouse Lung, Mouse Liver

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).

DA-b-4

Genomic DNA Extraction from Liver of Mouse

Protocol

2 ml micro tube

← Slice of mouse liver : 5 mg
← MDT : 180 µl
← EDT : 20 µl

Incubate for over night on rotary shaker at 55°C, and dissolve the tissue completely

10,000 rpm, 3 min, RT

Transfer the supernatant to a 1.5 ml micro tube

← <Option> RNase A treatment *1
← LDT : 180 µl

Mix thoroughly by vortexing at maximum speed : 15 sec *2

Flash spin down

Incubate at 70°C for 10min

Flash spin down.

← >99% ethanol : 240 µl

Mix thoroughly by vortexing at maximum speed : 15 sec *2

Flash spin down

Lysate

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 : 200 µl)

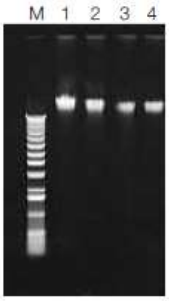
*1 Optioal steps
RNaseA : 20 µl
Tap the tube to mix the solution.
Flash spin down.
Set it down at room temperature for 2 min.

*2 Mix completely by vortexing at the maximum speed.
If the mixing is not enough by vortexing, use the tapping, pipetting or inverting.

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).

Results

AGE of extracted genomic DNA from Mouse Tissue

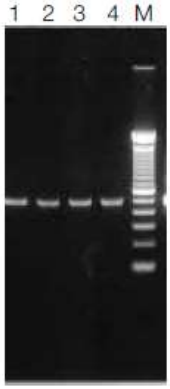


M : Size marker
1 : Lung tissue sample
2 : Kidney tissue sample
3 : Tail tissue sample
4 : Liver tissue sample

Other

・PCR

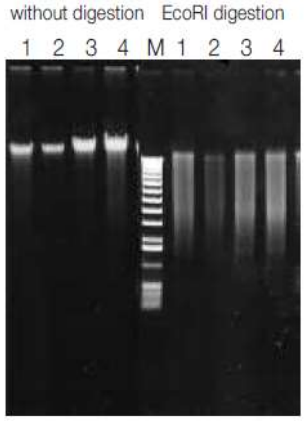
AGE of G3PDH PCR fragments amplified by genomic DNA extracted from mouse tissue using QuickGene isolation system and reagents



M : 100bp ladder marker
1 : Lung tissue sample
2 : Kidney tissue sample
3 : Tail tissue sample
4 : Liver tissue sample

・Restriction Enzyme Digestion

AGE of *Eco*RI restriction enzyme digestion fragments with genomic DNA extracted from mouse tissue using QuickGene isolation system and reagents



M : Size marker
1 : Tail tissue sample
2 : Liver tissue sample
3 : Lung tissue sample
4 : Kidney tissue sample

Common protocol is usable for the following

Mouse Lung, Mouse Kidney

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).

DA-b-5

Genomic DNA Extraction from Lung of Mouse

Protocol

2 ml micro tube

← Slice of mouse lung : 5 mg
← MDT : 180 µl
← EDT : 20 µl

Incubate for over night on rotary shaker at 55°C, and dissolve the tissue completely

10,000 rpm, 3 min, room temp

Transfer the supernatant to a 1.5 ml micro tube

← <Option> RNase A treatment *1
← LDT : 180 µl

Mix thoroughly by vortexing at maximum speed : 15 sec *2

Flash spin down

Incubate at 70°C for 10 min

Flash spin down.

← >99% ethanol : 240 µl

Mix thoroughly by vortexing at maximum speed : 15 sec *2

Flash spin down

Lysate

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 : 200 µl)

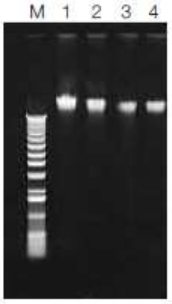
*1 Optioal steps
RNaseA : 20 µl
Tap the tube to mix the solution.
Flash spin down.
Set it down at room temperature for 2 min.

*2 Mix completely by vortexing at the maximum speed.
If the mixing is not enough by vortexing, use the tapping, pipetting or inverting.

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).

Results

AGE of extracted genomic DNA from Mouse Tissue

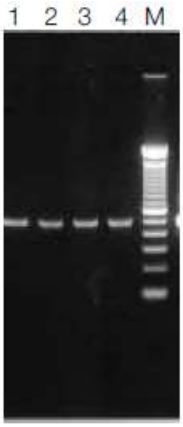


M : Size marker
1 : Lung tissue sample
2 : Kidney tissue sample
3 : Tail tissue sample
4 : Liver tissue sample

Other

・PCR

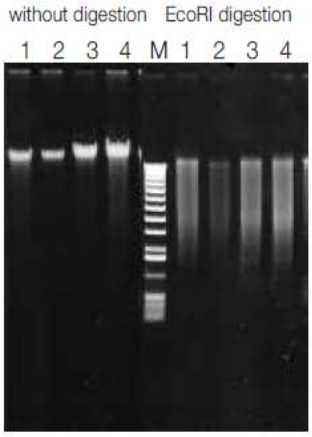
AGE of G3PDH PCR fragments amplified by genomic DNA extracted from mouse tissue using QuickGene isolation system and reagents



M : 100bp ladder marker
1 : Lung tissue sample
2 : Kidney tissue sample
3 : Tail tissue sample
4 : Liver tissue sample

・Restriction Enzyme Digestion

AGE of *Eco*RI restriction enzyme digestion fragments with genomic DNA extracted from mouse tissue using QuickGene isolation system and reagents



M : Size marker
1 : Tail tissue sample
2 : Liver tissue sample
3 : Lung tissue sample
4 : Kidney tissue sample

Common protocol is usable for the following

Mouse Kidney, Mouse Liver

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).

QuickGene

QuickGene DNA tissue kit S (DT-S)
QuickGene SP kit DNA tissue (SP-DT)

DA-b-6

Genomic DNA Extraction from Mouse Tail (Disruption Method)

Protocol

2 ml micro tube

← Mouse tail : 5 ± 0.5 mg

← 4.8 mm or 5.5 mmφ stainless ball : 2 balls

Freeze rapidly with liquid nitrogen

Disrupt with MS-100 (Tomy Seiko) *1

← MDT : 180 µl + EDT : 20 µl

Mix well

Incubate at 55°C for 15 min with mixing

Remove balls and transfer the solution to a new 1.5 ml micro tube

10,000 rpm, 1 min, RT

Leave undissolved residue,
and transfer 150 µl of supernatant to a new 1.5 ml micro tube

← LDT : 180 µl + >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

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 : 200 µl)

*1 In the case of 4.8 mmφ stainless ball :
2,700 rpm, 60 sec,
2 times

In the case of 5.5 mmφ stainless ball :
2,400 rpm, 30 sec,
2 times

Results

No Data

Common protocol is usable for the following

No Data

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).

KURABO

QuickGene

QuickGene DNA tissue kit S (DT-S)
QuickGene SP kit DNA tissue (SP-DT)

DA-b-7

Genomic DNA Extraction from slice of Mouse Tail

Protocol

2 ml micro tube

← Slice of mouse tail : 5 mg (5 ~ 6 mm)
← MDT : 180 µl
← EDT : 20 µl

Incubate for over night on rotary shaker at 55°C, and dissolve the tissue completely

10,000 rpm, 3 min, RT

Transfer the supernatant to a 1.5 ml micro tube

← <Option> RNaseA treatment *1
← LDT with >99% Ethanol : 420 µl *2

Mix thoroughly by vortexing at maximum speed : 15 sec *3

Flash spin down

Lysate

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 : 200 µl)

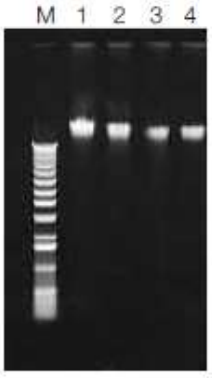
*1 Optioal steps
RNaseA : 20 µl
Tap the tube to mix the solution.
Flash spin down.
Set it down at room temperature for 2 min.

*2 Add 240 µl of >99% Ethanol into 180 µl of LDT and mix completely before using.

*3 Mix completely by vortexing at the maximum speed.
If the mixing is not enough by vortexing, use the tapping, pipetting or inverting.

Results

AGE of extracted genomic DNA from Mouse Tissue



M : Size marker
1 : Lung tissue sample
2 : Kidney tissue sample
3 : Tail tissue sample
4 : Liver tissue sample

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).

KURABO

The yield of genomic DNA (5 mg of mouse tail)

QuickGene isolation system and reagents	3.6 µl
Comparison method using spin column	3.6 µl

Protein contamination：A260/280

	#1	#2	#3	#4	#5	#6	#7	#8
QuickGene isolation system and reagents	1.95	1.94	1.95	1.94	1.95	1.97	1.96	1.96
Comparison method using spin column	1.96	1.94	1.97	2.01	1.95	1.99	2.00	1.99

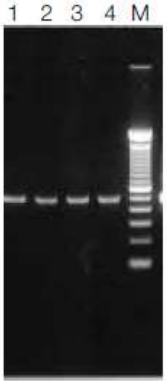
Chaotropic salt contamination：A260/230

	#1	#2	#3	#4	#5	#6	#7	#8
QuickGene isolation system and reagents	2.03	2.05	2.12	1.84	1.90	1.88	1.90	1.91
Comparison method using spin column	1.57	1.71	2.03	1.77	2.21	2.31	1.94	1.96

Other

・PCR

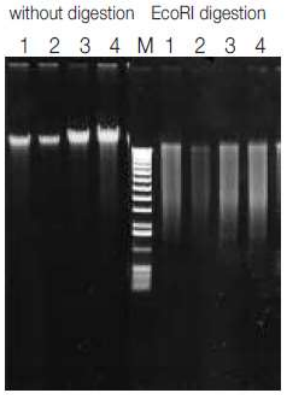
AGE of G3PDH PCR fragments amplified by genomic DNA extracted from mouse tissue using QuickGene isolation system and reagents



M : 100bp ladder marker
1 : Lung tissue sample
2 : Kidney tissue sample
3 : Tail tissue sample
4 : Liver tissue sample

・Restriction Enzyme Digestion

AGE of *Eco*RI restriction enzyme digestion fragments with genomic DNA extracted from mouse tissue using QuickGene isolation system and reagents



M : Size marker
1 : Tail tissue sample
2 : Liver tissue sample
3 : Lung tissue sample
4 : Kidney tissue sample

Common protocol is usable for the following

No Data

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).