

Isolation of genomic DNA Quick Guide

Saliva DNA with Oragene®

QuickGene DNA tissue kit L (DT-L)



In this Quick Guide, the protocol for isolation of genomic DNA from Saliva is modified based on the Handbook of QuickGene tissue kit L (DT-L) and the Operation Manual of QuickGene-Mini8L. * Before using, please read the Operation Manual.



Wear protective gloves and safety goggles during the experiments.

step1 Preparations

In order to isolate the target genomic DNA, please prepare the following items.

1 Preparations

QuickGene-Mini8L	10ml pipette or pipette aid	Tube mixer
DNA Tissue Kit L (DT-L)		
High grade ethanol (>99%)		
1.5 ml microtube (for DNA collection)		Oragene•DNA (OG-500) ® /DNA Genotek Inc. (For Saliva sample collection and preservation)
15 or 50 ml centrifuge tube (for lysate preparation)		
Micropipettes (P-1000)		
Water bath $(50^{\circ}\text{C} / 70^{\circ}\text{C})$		
Protective gloves		
Safety goggles		

2 Preparations of Reagents

◆Proteinase K (EDT)

Store at 2-8°C. This reagent is not used for the Saliva protocol.

♦ Tissue Lysis Buffer (MDT)

This reagent is not used for the Saliva protocol.

♦ Lysis Buffer (LDT)

Mix thoroughly before use. If the precipitates are formed, dissolve fully by incubating at 37° C.

♦ Wash Buffer (WDT)

Add 160 ml ethanol (>99%) into the bottle and mix well.

After adding the ethanol, close the cap and store at room temperature.

♦ Elution Buffer (CDT)

Use CDT for elution of genomic DNA.



Continue to step.2

step2 Protocol

In order to gain the target yield of DNA, please follow the protocol below.

1 Set the temperature of the water bath at 50℃

2 Prepare the Saliva sample

- 1) Collect the Saliva sample by reference to the user instructions of the Oragene®.
- 2) Mix with vortex mixer (maximum speed) for 15 seconds.
- 3) Transfer 2 ml of Oragene/Saliva sample to a new 15 ml (or 50 ml) centrifuge tube.
- 4) Incubate with the water bath at 50°C for 2 hours.

Regarding the Oragene® product, please contact the supplier or manufacturer for further information.

3 Set the temperature of the water bath at 70°C

4 Prepare Lysate

- 1) Add 2 ml of LDT.
- 2) Mix with vortex mixer (maximum speed) for 15 seconds.
- 3) Incubate with the water bath at 70° C for 10 minutes.
- 4) Add 2.4 ml of ethanol (>99%).
- 5) Mix with vortex mixer (maximum speed) for 15 seconds.

5 Complete the lysis

Perform the isolation operation quickly after completing the lysis.

QuickGene

step3 Isolation protocol with QuickGene-Mini8L

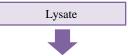
Use QuickGene-Mini8L to isolate genomic DNA.

QuickGene-Mini8L Workflow

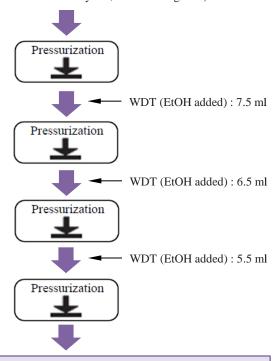


The Pressurization mark in the workflow indicates the following operations.

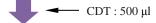
- 1. Set holder into system. *Please read the Operation Manual to know how to set the holder.
- 2. Rotate pressurizing switch toward the front side to start pressurizing.
- 3. Make sure that there is no residual liquid in the cartridge and return the pressurizing switch to original position.
- 4. Move the holder to pressurize the next row. Repeat 2. and 3.
- 5. Pull out holder from system.



Transfer whole lysate to the cartridge and set pressure seal plate. (If any aggregates are formed in lysate, transfer altogether)



Move the cartridge holder into the elution position. (Please read the Operation Manual to know how to set the holder.)



Incubation at room temperature for 90 sec.

