Apostle MiniGenomics® Total RNA Isolation Kit, Automation Protocol

Isolation of total RNA from whole blood/cells suspension/dissociated tissue (100 µL) with MagTouch 2000, 96-well

Revision A.0

Product Description

The Apostle MiniGenomics® Total RNA Isolation Kit is designed for rapid, simple isolation of high-quality, ready-to-use total RNA from blood, cultured cells, or tissue samples. The kit uses proprietary Apostle MiniGenomics® technology, offering highly efficient, reproducible recovery of high-quality RNA with high yield. The protocol allows processing in either 96-well plates (manual or automated) or single tubes (manual). The isolated RNA are suitable for a broad range of subsequent applications, including sequencing, PCR, etc.

Required materials

Apostle MiniGenomics® Total RNA Isolation Kit (Cat# A220412-50 or A220412-200)

Apostle MagTouch 2000 (Cat# A201126-96)

Apostle 96-well deep well plates and tip comb

DNase/RNase-free tubes (1.5 mL and 15 mL)

Adjustable single and multi-channel micropipettes and tips (1000 µL, 200 µL, and 20 µL)

Vortex or shaker

β-mercaptoethanol (BME)

Ethanol, 200 proof, molecular biology grade

Isopropanol, 100%

Water, nuclease-free

PBS, nuclease-free (optional)

Sample requirements

Samples: Blood samples (Collected by anticoagulant blood collection tube. Do not freeze); Cultured cells samples (Trypsinized and suspended in nuclease-free PBS. Cell number of each 100 µL sample is $\leq 1\times10^6$); Tissue samples (≤30 mg of tissue sample homogenized with 200 µL nuclease-free PBS).

Sample storage: Process the sample immediately after collection, or temporarily stored at 4°C. Do not freeze.

Reagent Preparation

DNase and **Binding Enhancer** are shipped with dry ice. Immediately store them at -20°C after receiving and thaw them before use.

Before first use, add **8 mL isopropanol** into **Re-binding Solution** and mix well. Check the checkbox on label to record the completion of IPA addition.

Before first use, add **26.4 mL isopropanol** into **Wash Solution** and mix well. Check the checkbox on label to record the completion of IPA addition.

Before each use, prepare 80% ethanol with ethanol, 200 proof, molecular biology grade and nuclease-free water. 80% ethanol needed for each sample is 1.2 mL.

All solutions stored at room temperature (15 to 30°C) should be clear. If precipitate is observed in any of these reagents, warm the solution to 37°C until the precipitate dissolves.

Magnetic nanoparticle solution should be brown solution. Vortex magnetic nanoparticle solution to fully resuspend the nanoparticles before use.

Procedure for isolation of total RNA

A. Nucleic acid isolation and DNase treatment

1. Set up the 96-well plates according to the table below, outside the instrument
2. Select the [TotalRNAP1] program. Open the cabin door, and place the plates into the MagTouch 2000 as indicated on the instrument display. Press the position button to turn the rotary table and place all the plates in turn.
3. After placing all the plates, press the [Run] icon on the interface to start the program.

Plate Position	Plate Type	Name	Content	Reagent volume for each well
1	96 well plate with comb	-Load-	Comb	-
2	96 well plate	Bind	Lysis/Binding Solution	75 µL
			β-mercaptoethanol	1 µL
			Sample (blood/cells or tissue suspension)	100 µL
			Proteinase K	40 µL
			Isopropanol	170 µL
			Binding Enhancer	3 µL
			Magnetic Nanoparticles	10 µL
3	96 well plate	Wash1a	Wash Solution	400 µL
4	96 well plate	Wash1b	Wash Solution	400 µL
5	96 well plate	Wash2a	80% EtOH	400 µL
8	96 well plate	DNase	DNase buffer	100 µL
			DNase	1 µL

B. RNA rebinding and elution

1. Set up the 96-well plates according to the table below, outside the instrument

Plate Position	Plate Type	Name	Content	Reagent volume for each well
1	96 well plate with comb	-Load-	Comb (same as in previous step)	-
2	96 well plate	Bind	DNase treatment sample from step1	101 µL
			Re-binding Solution	215 µL
3	96 well plate	Wash2b	80% EtOH	400 µL
4	96 well plate	Wash2c	80% EtOH	400 µL
5	96 well plate	Elu	Elution Buffer	50 to 100 µL

2. Select the [TotalRNAP2] program. Open the cabin door, and place the plates into the MagTouch 2000 as indicated on the instrument display. Press the position button to turn the rotary table and place all the plates in turn.
3. After placing all the plates, press the [Run] icon on the interface to start the program.
4. When the program is complete, collect the supernatant that contains RNA from the elution plate.
5. Store the RNA sample at -80°C for long-term storage.