WelPrep™ Total RNA Prep Kit

Catalog Number **PR 103-01**
Storage Temperature **4°C or Room Temperature**

Product Description

WelPrep™ Total RNA Prep Kit is designed for easy, rapid isolation of total RNA. The kit is effectively isolate to small RNAs less than 100 nt from the samples. Because use the Spin Column, the kit is not required the general steps - phase separation, precipitation, drying - for total RNA isolation unlike the conventional method. The entire procedure ends within 15 minutes. (35 minutes include the DNase I treatment)

Contents

Lysis Solution	1 ea
Wash Buffer	1 ea
Elution Buffer	1 ea
Spin Column	50 ea
Collection Tube	50 ea

Storage/Stability

The Lysis Solution are stored at 4°C and two other reagents and columns are stored at room temperature.

Precautions

Prior to use the kit, 80 ml absolute ethanol must be added to Wash Buffer.
Because DNase I and 10X DNase I reaction buffer are not provided, the reagents must be purchased separately for genomic DNA digestion. The kit is recommended to Roche or Takara product.
All centrifugation should be proceed at more than 10,000 X g.

Protocols

I. Standard procedure

1. Prepare less than 4 X 10^6 cells or 50 mg tissue. Before add Lysis Solution, resuspend the cell pellets by vortex and homogenize the tissue using liquid nitrogen.

2. Add 0.35 ~ 1 ml Lysis Solution to the sample and vortex vigorously for 10 ~ 15 seconds.

3. Incubate for 5 ~ 10 minutes.

4. Add 100% ethanol of the same volume of Lysis Solution, mix by inversion but don't vortex.

5. Add the mixture into Spin Column and centrifuge for 1 minute. Discard the flow-through from Collection Tube.

6. Add 600 ul Wash Buffer to the column and centrifuge for 1 minute. Discard the flow-through from Collection Tube.

7. Repeat 6 step.

8. In order to remove for residual ethanol, centrifuge for 2 minutes at max speed.

9. Transfer the column into RNase-free tube.

10. Add 50 ul Elution Buffer or RNase-free D.W., following 1 minute incubation.

11. Centrifuge for 1 minute. The isolated total RNA is store to -70°C until use.

II. Procedure including genomic DNA elimination

1. Proceed to step 1 ~ 5 of the standard procedure.

2. Add 100 ul Wash Buffer to Spin Column and centrifuge for 1 minute. Discard the flow-through from Collection Tube.

3. Must be prepare DNase I Working Solution* immediately before use.
   *Composition of DNase I Working Solution

80 ul	Wash Buffer
10 ul	10X DNase I Reaction Buffer
X ul	RNase-free D.W.
5 ~ 10 units	DNase I (about 1 ~ 2 ul)
Adjust to 100 ul volumns	

   ※10X DNase I Reaction Buffer and DNase I are not included.

4. Add 100 ul DNase I Working Solution to Spin Column, following incubate at 37°C for 10 ~ 20 minutes, then centrifuge for 1 minute. Discard the flow-through.

5. Add 600 ul Wash Buffer to column and centrifuge for 1 minute. Discard the flow-through from Collection Tube.

6. Repeat 5 step.

7. Centrifuge for 2 minutes at max speed.

8. Transfer the column into RNase-free tube.

9. Add 50 ul Elution Buffer or RNase-free D.W., following 1 minute incubation.

10. Centrifuge for 1 minute. The isolated total RNA is store to -70°C until use.

www.welgene.com | Seoul Office | TEL. 82-2-2057-3288 | Headquarter & Factory | TEL 82-53-811-7081 | FAX 82-53-811-7096

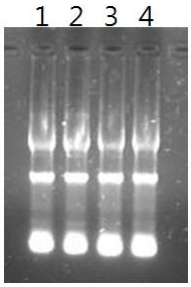

Fig 1. Comparison of total RNA isolation from different product
1, 2 : Coventional liquid product
3, 4 : WelPrep™ Total RNA Prep Kit

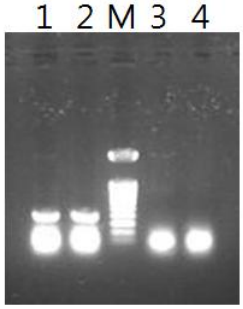

Fig 2. Verification of genomic DNA elimination through PCR of intron region of β-actin
1, 2 : Not treated total RNA
M : 100 bps size marker
3, 4 : DNase I treated total RNA

Troubleshooting Guide

Problem	Check point
Low RNA yield	Incomplete lysis of sample
	Increase the volume of Lysis Solution
RNA not detect	Degradate RNA
	Samples were not immediately lysis or treated with the RNase contaminated equipment
RNA smear	Total RNA was not kept in the right temperature. Check that the temperature of RNA storage.
	Washing step was not enough. Check that the addition of ethanol to Wash Buffer