

Marvel Life Junction

**Touch
Your Experiments**

MVPTMrep

**DNA^{out} Plus Total
RNA Kit (with DNase I)**

RP-30050PR



RP-30100PR

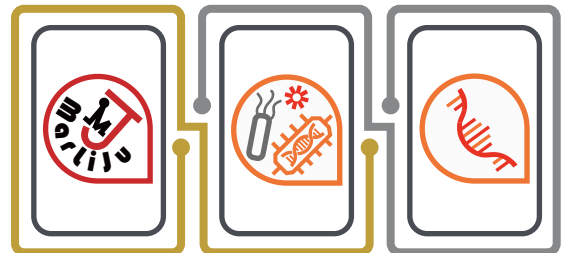


RP-30200PR



Version No. : 1.0R

User Quick Guide



www.marliju.com

REF

RP-30050PR / RP-30100PR / RP-30200PR

Cat. No.	RP-30050PR / RP-30100PR / RP-30200PR
Number of Preps	50 Preps / 100 Preps / 100 Preps x 2
Spin A Column (Collection Tube included, 50 ea / pk)	1 pk / 2 pk / 2 pk x 2
Spin B Column (Collection Tube included, 50 ea / pk)	1 pk / 2 pk / 2 pk x 2
Buffer OTR01	50 mL / 100 mL / 100 mL x 2
Buffer OTR02* (Add Ethanol)	22 mL / 44 mL / 44 mL x 2
Buffer OTR03* (Add Ethanol)	8 mL / 16 mL / 16 mL x 2
Buffer OTR04* (Add Ethanol)	8 mL / 16 mL / 16 mL x 2
Buffer OTR05	15 mL / 15 mL / 15 mL x 2
Buffer OTRA	45 mL / 90 mL / 90 mL x 2
DNase I ** , *** (Lyophilized)	1 ea / 1 ea / 1 ea x 2
DNase I resus. Solution	1 ea / 1 ea / 1 ea x 2
Protocol	1 ea / 1 ea / 1 ea x 2

* Before use, add Absolute Ethanol (ACS grade or higher) to the Buffer bottle according to the instructions on the label.

** Upon receiving the kit, store the DNase I, packaged in a small box, at -20°C until use.

*** When using the kit, completely thaw DNase I in DNase I resus. Solution and aliquot it into 5-10 tubes, storing them at -20°C.

(Caution) Do not vortex DNase I; gently thaw it by pipetting.

Introduction

The MVPPrep™ DNA^{OUT} Plus Total RNA Kit is devised with a two Spin Column and DNase I treatment system plus for the easy extraction of DNA-free total RNA from animal cells (up to 1 x 10⁷) and animal tissue (up to 25 mg), all without the use of harmful organic solvents (phenol, chloroform). It employs optimized buffers and a straightforward experimental procedure, ensuring the stable and high-purity extraction of total RNA.

The extracted total RNA can be applied to various downstream experiments, including cDNA synthesis and RT-PCR.

Storage Condition

The MVPPrep™ DNA^{OUT} Plus Total RNA Kit should be stored at room temperature (15-25°C) until its expiry date.

Preparation before the experiment and Precautions

Before the experiment, check the Buffers, especially Buffer **OTR02**, if there are precipitants, dissolve the precipitants completely at 37°C.

Do not perform vigorous vortexing when resuspending DNase I, which may lead to the inappropriate performance of DNase I.

1.5ml tube.

Absolute Ethanol (ACS or higher grade).

Heating block or water bath.

14.3 M β-mercaptoethanol (β-ME) or 2 M dithiothreitol (DTT) in water.

Prepare the DNase I Reaction Solution by mixing 5 ul of DNase I Solution and 75 ul of Buffer **OTRA** for each reaction.

All centrifuges should be performed at the speed of 13,000 rpm at room temperature (15-25°C) unless instructions are given.

Always wear gloves and eye protectors and follow standard safety precautions.



0 Add β -mercaptoethanol (β -ME) or 2M DTT to the appropriate volume of Buffer **OTR01** for a single experiment to make a 1% solution. (e.g., add 10 μ l per 1 ml of Buffer **OTR01**)

- **Cultured Cell** : Make cell pellet a maximum of 1×10^7 cells through centrifugation and discard supernatant.
- **Animal Tissue** : Transfer a maximum of 25 mg of tissue into a 1.5 ml tube and then add 400 μ l of **OTR01** and homogenize as finely as possible.

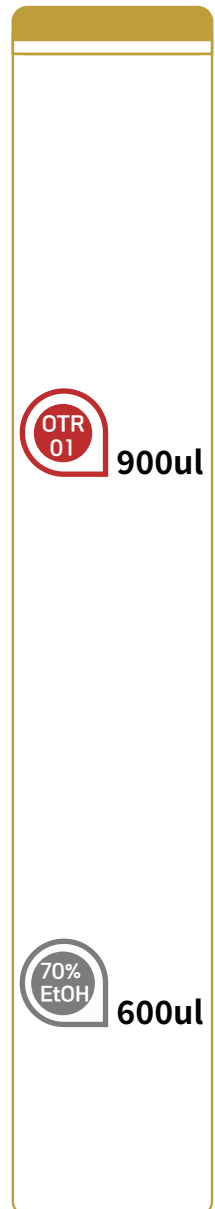
1 Add 900 μ l of Buffer **OTR01** to the Ttube for the cell pellet or 500 μ l for the homogenized tissue mixture with 400 μ l of **OTR01** and completely dissolve it through pipetting or vortexing.
(Caution) Prepare only the amount needed for a single experiment by mixing 10 μ L of β -ME or 2 M DTT with Buffer **BR02** per 1 mL.

2 Transfer 700 μ l of the Sample mixture into Spin A Column and centrifuge for 1 min. Transfer the filtered Sample mixture from the Collection Tube into a new 1.5 ml tube (not provided). After transfer, reattach the Collection Tube with the Spin A Column.
(Caution) Do not discard the filtered Sample Mixture in the Collection Tube.

3 Repeat Step 2 with the remaining Sample mixture.

4 Add 600 μ l of 70% Ethanol into the 1.5 ml tube and thoroughly mix through pipetting (more than 10 times) or pulse-vortexing.

5 Transfer 700 μ l of the filtered Sample mixture into Spin B Column and centrifuge for 1 min. After centrifugation, discard the filtrate in the Collection Tube and reattach it with the Spin B Column.



- 6 Repeat Step 5 with the remaining Sample mixture.
- 7 Add 400 ul of Buffer **OTR02** into the Spin B Column and centrifuge for 1 min. After centrifugation, discard the filtrate in the Collection Tube and reattach it with the Spin B Column.
- 8 Add 80 ul of DNase I Reaction Solution into the Spin B Column and wait for 20 min at room temperature (15-25°C).
(Caution) Prepare the DNase I Reaction Solution by mixing 5 ul of DNase I Solution and 75 ul of Buffer **OTRA** for each reaction.
- 9 Add 400 ul of Buffer **OTR02** into the Spin B Column and centrifuge for 1 min. After centrifugation, discard the filtrate in the Collection Tube and reattach it with the Spin B Column.
- 10 Add 700 ul of Buffer **OTR03** into the Spin B Column and centrifuge for 1 min. After centrifugation, discard the filtrate in the Collection Tube and reattach it with the Spin B Column.
- 11 Add 700 ul of Buffer **OTR04** into the Spin B Column and centrifuge for 1 min. After centrifugation, discard the filtrate in the Collection Tube and reattach it with the Spin B Column.
- 12 Centrifuge for an additional 1 min.
(Caution) If the liquid is still in the Spin Column after centrifugation, perform an additional 1-minute centrifugation.
- 13 Transfer the Spin Column to a new 1.5 ml tube (not provided) and reattach it.
- 14 Add 30-100 µl of Buffer **OTR05** or Nuclease-free water into the center of the Spin B Column and wait for 1 min at room temperature (15-25°C).
- 15 Centrifuge for 1 min.

OTR02 400ul

OTRA

OTR02 400ul

OTR03 700ul

OTR04 700ul

OTR05 30ul~100ul

Note.



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