Fast HQ RNA Extraction Kit

For the efficient isolation RNA of cells

RUO *Research Use Only* REF *17213* Σ 50 15℃ 25℃

PRODUCT FEATURES

- Fast HQ RNA Extraction Kit is designed for rapid purification of RNA from animal cell, bacteria, fungi, yeast, protozoa and blood
- The fastest and the simplest procedure for purifying the highest quality RNA
- The high quality RNA can be used directly for the downstream application

INTRODUCTION

The method used in Fast HQ RNA Extraction Kit can obtain high quality RNA within 5 minutes and is different from traditional phenol / chloroform technology. The Fast HQ RNA Extraction Kit is an innovative product designed for the easy, reliable and rapid isolation of DNA-free RNA from a wide range of cell (up to 2.5 x 10^6). The procedure is simple and fast. The result is highly-concentrated, DNA-free RNA that is suitable for RT-PCR, qRT-PCR, hybridization, sequencing etc.

KIT CONTENTS

Components	50 col.
HQ RNA Buffer[1]	25 ml
HQ RNA Washing Buffer[2]	18 ml
HQ RNA Elution Buffer	7.5 ml
Column/Collection Tube	50 ea
Storage Conditions : Room Temperature	

[1] Add 35 ml of 100% isopropanol to HQ RNA Buffer. Mix well. Mark on the labels that isopropanol is added. Store it at room temperature.

[2] Add 42 ml of 100% ethanol to HQ RNA Washing Buffer. Mix well. Mark on the labels that ethanol is added. Store it at room temperature.

STORAGE CONDITION

The Fast HQ RNA Extraction Kit can be stored at room temperature (15-25℃) for up to 12 months without showing any reduction in performance and quality. For longer storage, this kit can be stored at 2-8℃.

INPORTANT NOTES

1. No damage to RNA quality during extraction procedure
2. Exclusive purification of RNA without any genomic DNA contamination
3. Every procedure at ambient temperature without any cold or freezing step
4. Final 5 ~ 20ug of RNA for more than 20 reactions of RT-PCR or qRT-PCR reaction
5. No Liquid nitrogen or an expensive homogenizing device

PROCEDURES FOR LYSATE PREPARATION

A. Cultured animal cells

- The maximum recommended input of cells is 2.5 x 10^6. As a reference, confluent 60mm and 100mm dish contain 3.2 x 10^6 and 8.8 x 10^6 cells respectively.
- The lysate can be stored at -20℃ for later RNA extraction.
- For RNA extraction from frozen cell, loosened the pellet by scraping the tube for 5 - 6 times over an uneven surface such as a microcentrifuge tube rack and add lysis solution directly to frozen sample.

< Cell grown in monolayer >

1. Aspirate media
2. Add 700ul of HQ RNA Buffer and mix with gentle swirling for 30 sec.
3. Transfer all lysate to a microcentrifuge tube and vortex for 15 sec.

< Cell grown in suspension / detached monolayer cells >

1. Transfer cell suspension to a microcentrifuge tube (not provied) and centrifuge for 30 sec.
2. Remove supernatant with pipetting or a gentle vacuum..
3. Repeat steps 1 and 2 as required.
4. Closed the cap and loosened the pellet by scraping the tube for 5 – 6 times over an uneven surface such as a microcentrifuge tube rack.
5. Add 700ul of HQ RNA Buffer and vortex for 30 sec.

B. Blood sample

- Blood sample should be fresh and used within a couple of hours after collection.
- Frozen blood sample is not appropriated for blood RNA extraction by this kit.

1. Invert mix for 5 – 6 times of whole blood in EDTA collection bottle.
2. Transfer 200ul of whole blood into a microcentrifuge tube.
3. Microcentrifuge for 30 sec.
4. Remove all liquid.
5. Closed the cap and loosened the pellet by scraping the tube for 5 – 6 times over an uneven surface such as a microcentrifuge tube rack.
6. Add 700ul of HQ RNA Buffer and vortex for 30 sec.

C. Bacteria, fungi, yeast, and protozoa

- It is recommended that no more than 1.5ml of saturated bacterial or yeast culture volume be used in this procedure in order to prevent clogging of the column.
- It is recommended that no more than 100mg of fungi or protozoa be used for this procedure in order to prevent clogging of the column.

1. Transfer cell in culture media or PBS to a microcentrifuge tube (not provided) and centrifuge for 30 sec.
2. Remove supernatant with pipetting or a gentle vacuum.
3. Closed the cap and loosened the pellet by scraping the tube for 5 – 6 times over an uneven surface such as a microcentrifuge tube rack.
4. Add 700ul of HQ RNA Buffer and vortex for 30 sec.

D. Nasal or Throat Swabs

1. Transfer 700ul of HQ RNA Buffer to a clean microcentrifuge tube (not provided).
2. Gently brush a sterile, single-use cotton swab (not provided) inside the nose or mouth of the subject.
3. Cut the cotton tip of swab with clean scissors and leave in the microcentrifuge tube.
4. Closed the cap and vortex for 30 sec.

PROTOCOL

- All centrifugation in 10,000 rpm at room temperature.
- If column is clogged, spin more until a complete flow through.
- Universal procedure for all lysate.
- All steps at ambient temperature.

1. Transfer all lysate to column. Do not centrifuge to collect the sample from the lid. Any centrifuge in this step would severely reduce the amount of purified RNA.
2. Centrifuge column for 15 sec.
3. Discard the flow through. Reassemble the spin column with its collection tube.
4. Repeat the steps of 1 and 3 if required.
5. Apply 700ul of HQ RNA Washing Buffer and centrifuge for 15 sec. Discard the flow through.
6. Apply 400ul of HQ RNA Washing Buffer and centrifuge for 30 sec. Discard the flow through.
7. Replace the collection tube a clean microcentrifuge tube (not supplied).
8. Add 100ul of HQ RNA Elution Buffer to column.
9. Close the cap and vortex for 15 sec.
10. Centrifuge for 15 sec.
11. Reload the flow through to the top of column and centrifuge for 30 sec.

NOTICE BEFORE USE

Fast HQ RNA Extraction Kit is intended for research use only. And Fast HQ RNA Extraction Kit is developed, designed, and sold for research purpose only. It is not intended to be used for human or animal diagnosis of diseases. Do not use internally or externally in humans or animals. When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. Please observe general laboratory precaution and utilize safety while using this kit.

PRODUCT WARRANTY AND SATISFACTION GUARANTEE

At iNtRON we take pride in the quality and availability of our technical support. iNtRON is staffed by experienced scientists with extensive practical and theoretical expertise in molecular biology and the use of iNtRON products. If you have any questions or experience any difficulties regarding the **Fast HQ RNA Extraction Kit**, please do not hesitate to contact us. iNtRON customers are a major source of information regarding advanced or specialized uses of our products. For technical assistance and more information please call iNtRON's local distributors.

TROUBLE SHOOTING GUIDE

This troubleshooting guide may be helpful in solving problems that may frequently arise. The scientists at iNtRON are always happy to answer any questions you may have about the information or protocol in this manual or other molecular biology applications.

Problem / Possible cause	Recommendation
Low yield or no RNA	
1)Too much starting material	• Do not overload the sample, overloading significantly reduces yield. Reduced the amount of starting material.
2)Sample integrity is poor	• Samples stored for extended duration at room temperature, frozen or refrigerated for extended periods will contribute to reduced RNA yield and integrity.
3)Step were not followed correctly or wrong reagent used	• Check the protocol; HQ RNA Washing Buffer(HQ RNA Buffer) did not contain 100% EtOH(100% isopropanol) so, 100% EtOH(100% isopropanol) must be added to the HQ RNA Washing Buffer(HQ RNA Buffer) before use.
RNA degradation	
1)RNA degraded during sample preparation	• It is essential to work quickly during sample preparation.
2)Inappropriately handled	• Use DEPC-treated glassware and wear gloves at all time.
RNA does not perform well in the downstream application	
1)Ethanol carryover	• Ensure that during the HQ RNA Washing Buffer, the column is spun at appropriate speed(10,000 rpm) 1min to dry column.

ORDERING INFORMATION

Product Name	Amount	Cat. No.
DNA-Eraser™ Genomic DNA Removal Kit	200 rxn.	21190
Maxime™ RT PreMix (Oligo (dT)15 Primer)	96 tubes	25081
Maxime™ RT PreMix (Random Primer)	96 tubes	25082
ONE-STEP RT-PCR PreMix Kit	50 rxn.	25101
Maxime™ RT-PCR PreMix	96 tubes	25131
RealMOD™ Green Fast qPCR mix	5 mL	25454
RealMOD™ Green Fast qPCR mix(LR)	5 mL	25453
RealMOD™ Green qRT-PCR mix	100 rxn.	25109
RealMOD™ Green qRT-PCR mix(LR)	100 rxn.	25107
AMV Reverse Transcriptase	200 Unit	27021
M-MLV Reverse Transcriptase	10000 Unit	27032

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