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# INSTRUCTION INSTRUCTION

# **RNA Pure Kit**

IB47640 (4 rxns) IB47641 (50 rxns) IB47642 (100 rxns)

## Introduction

The RNA Pure Kit uses a simple and efficient spin column procedure to purify Total RNA stored in RNase-free water, elution buffer or TE Buffer following extraction using acid-guanidinium-phenol-chlorofom based methods such as TRIzol<sup>®</sup> Reagent and IBI Isolate. Contaminants such as RNases, DNA and residual phenol are effectively removed using a simple 4 step procedure. The high-quality, total RNA is eluted in RNase-free Water or TE (RNase-free) and is ready for use in a variety of sensitive downstream applications.

# **Quality Control**

The RNA Pure Kit is tested on a lot-to-lot basis. Following RNA purification using the RNA Pure Kit, 10 µl from a 50 µl eluate of purified RNA is analyzed by electrophoresis on a 0.8% agarose gel.

## Advantages

- Purify up to 50  $\mu g$  of total RNA within 10 minutes
- Recovery: up to 80% of high quality RNA (A260/A280 = 1.9-2.0)
- Elution volume: 20-50 µl

• Compatibility: purify RNA stored in RNase-free water, elution buffer or TE Buffer following extraction using IBI Isolate,

TRI-Reagent<sup>®</sup>, TRIzol<sup>®</sup>, RNAzol<sup>®</sup> and QIAzol<sup>®</sup> etc.

# Applications

RT-PCR, Northern Blotting, Primer Extension, mRNA Selection, cDNA Synthesis, RNase Protection Assay

# Caution

During operation, always wear a lab coat, disposable gloves, protective goggles and (anti-fog) procedure mask. Disposable/non-disposable glassware, plasticware and automatic pipettes should be sterile (RNase-free) and used only for RNA procedures.

# Applications

Absolute ethanol, 1.5 ml microcentrifuge tubes (RNase-free).

# **Kit Contents**

ltem	Volume	Product	Shipping	Storage
RNA Pure Buffer	3 ml	PR004	room temperature	dry at room temperature
	30 ml	PR050		
	60 ml	PR100		
Wash Buffer <sup>1</sup> (Add Ethanol)	1 ml (4 ml)	PR004		
	12.5 ml (50 ml)	PR050	room temperature dry at room	dry at room temperature
	25 ml (100 ml)	PR100		

ltem	Volume	Product	Shipping	Storage
RNase-free Water	1 ml	PR004	room temperature	dry at room temperature
	6 ml	PR050		
	6 ml	PR100		
PR Columns	4 pcs	PR004		dry at room temperature
	50 pcs	PR050	room temperature	
	100 pcs	PR100		
2 ml Collection Tubes	4 pcs	PR004		
	50 pcs	PR050	room temperature	dry at room temperature
	100 pcs	PR100		

<sup>1</sup> Add absolute ethanol (see the bottle label for volume) to Wash Buffer prior to initial use.

### **RNA Purification Protocol Procedure**

Please read the entire instruction manual prior to starting the Protocol Procedure.

#### **1. Sample Preparation**

1. Transfer up to 100 µl of RNA product (stored in RNase-free water, elution buffer, TE buffer) to a 1.5 microcentrifuge tube (RNase-free). 2. Add 5 volumes of RNA Pure Buffer to 1 volume of the sample then shake vigorously.

#### 2. RNA Binding

- 1. Add an equal volume of 70% ethanol (if the sample mixture is 600 µl, add 600 µl of 70% ethanol) to the sample mixture from step 1.
- 2. Shake the mixture vigorously and break up any precipitate with a pipette.
- 3. Place a PR Column in a 2 ml Collection Tube then transfer 500 µl of the ethanol-added mixture to the PR Column.
- 4. Centrifuge at 14-16,000 x g for 1 minute then discard the flow-through and transfer the remaining mixture to the same PR Column.
- 5. Centrifuge at 14-16,000 x g for 1 minute then discard the flow-through and place the PR Column back in the 2 ml Collection Tube.

#### 3. RNA Wash

- 1. Add 600  $\mu l$  of Wash Buffer (make sure ethanol was added) to the CENTER of the PR Column.
- 2. Centrifuge at 14-16,000 x g for 30 seconds then discard the flow-through and place the PR Column back in the 2 ml Collection Tube.
- 3. Centrifuge at 14-16,000 x g for 3 minutes to dry the column matrix.

#### 4. RNA Elution

- 1. Place the dried PR Column in a clean 1.5 ml microcentrifuge tube (RNase-free).
- 2. Add 20-50  $\mu l$  of RNase-free Water or TE (RNase-free) to the CENTER of the column matrix.
- 3. Let stand for 2 minutes or until the RNase-free Water or TE (RNase-free) is absorbed completely by the matrix.
- 4. Centrifuge at 14-16,000 x g for 2 minutes to elute the purified RNA.

# Troubleshooting

Problem	Cause	Solution
Low Yield	A. Incorrect RNA Elution	<ul> <li>Make sure RNase-free Water is added to the center of the RB Column and is absorbed completely.</li> </ul>

Problem	Cause	Solution
Low Yield	A. Incorrect RNA Elution	<ul> <li>Make sure RNase-free Water is added to the center of the RB Column and is absorbed completely.</li> </ul>
Degraded RNA	A. Incorrect sample storage temperature	A. Extracted RNA should be stored at -70°C.
Low Yield	A. Incomplete wash step	<ul> <li>Wash the RB Column with ethanol added Wash Buffer 2 times.</li> </ul>
Eluted RNA does not perform well in downstream applications	A. Residual ethanol contamination	<ul> <li>Following the wash step, dry the RB Column with additional centrifugation at 14-16,000 x g for 5 minutes or incubate at 60°C for 5 minutes.</li> </ul>

### **Related RNA Extraction Products**

RNA Extraction and Purification				
Product	Package Size	Catalogue Number		
Total RNA Mini Kit (Blood/Cultured Cell)	50/100/300 preps	IB47321/322/323		
Total RNA Mini Kit (Tissue)	50/100/300 preps	IB47301/302/303		
Total RNA Mini Kit (Plant)	50/100/300 preps	IB47341/342/343		
rBAC Mini RNA Bacteria Kit	100/300 preps	IB47421/422		
rYeast Total RNA Mini Kit	100/300 preps	IB47411/412		
miRNA Isolation Kit	100 preps	IB47371		
IBI Isolate	100/200 rxns	IB47601/602		
IBI Tri-Isolate	100/200 rxns	IB47631/632		
RNA Pure Kit	50/100 rxns	IB47641/642		
Virus DNA/RNA Purification				
Product	Package Size	Catalogue Number		
Viral Nucleic Acid Extraction Kit II	50/100/300 preps	IB47401/402/403		

For additional product information, please visit www.ibisci.com. Thank you!

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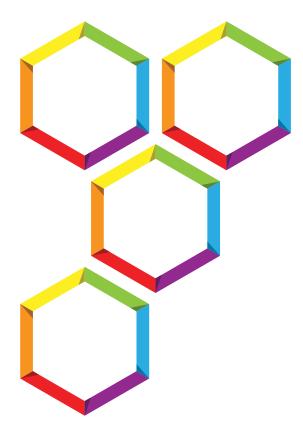


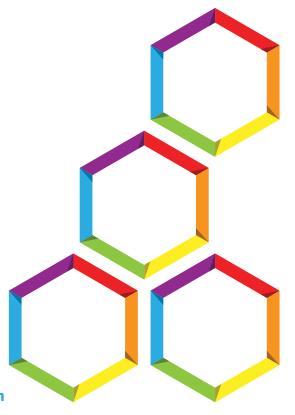


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