DNA free RNA SYSTEM
PROTOCOL & TECHNICAL BULLETIN

BenevBio

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KIT CONTENTS

50 RXN Kit (DRS0001S):

ITEM	CATALOG #	50 RXN
DNA free RNA Buffer	DRS0002	1 vial/25ml
DNA free Wash Buffer Con.	DRS0004	1 vial/6ml
DNase I w/ 10x Buffer	DRS0006	100 Units
NucleoPur™ Columns	DRS0008	50 columns
Collection Tubes	CT0050	50 tubes

PRECAUTIONS

THIS PRODUCT IS NOT INTENDED OR APPROVED FOR USE IN HUMANS OR VETERINARY ANIMALS. THE USE OF THIS PRODUCT IN A THERAPEUTIC SETTING IS HAZARDOUS AND MAY RESULT IN ILLNESS, INJURY AND/OR DEATH.

Please read these instructions carefully before using this system.

The reagents in this system have been formulated and tested to work exclusively with the BenevBio DNA free RNA System. This system may not perform as described if any reagent or procedure is replaced and/or modified.

For research use only. Not for human or diagnostic use.

WARRANTY

BenevBio guarantees the performance of all products when used as directed for their intended purpose. Should any product fail to perform satisfactorily for any reason other than misuse, BenevBio's sole liability hereunder shall be limited to refund of the purchase price or, at the discretion of BenevBio, the replacement of all material that does not meet our specifications. BenevBio shall not be liable otherwise or for incidental or consequential damages including, but not limited to, the cost of handling. We reserve the right to change or modify any components to enhance the performance or design. The Buyer must give notice within thirty (30) days after receipt of material or shall constitute a waiver by Buyer of all claims hereunder with respect to said material.

BenevBio CUSTOMER CARE INFORMATION

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Internet: www.benevbio.com

STORAGE AND STABILITY

The system will perform as specified if stored dry at room temperature (20-25°C). Under these conditions the kit can be stored for up to 12 months without any decline in performance and/or quality.

ADDITIONAL ITEMS REQUIRED

1. Microcentrifuge capable of reaching 16,000 x g.
2. Adjustable pipettor.
3. Ethanol.
4. Microcentrifuge tubes.
5. A source of "UltraPure" water. Water used to elute samples must be deionized and free of trace organic contaminants.

Note: UltraPure water is available for purchase.

INTRODUCTION AND DESCRIPTION

The BenevBio DNA free RNA System is designed to eliminate contaminating DNA from crude RNA extractions for applications sensitive to small amounts of DNA. The NucleoPur™ Columns efficiently purify and concentrate RNA samples of enzymatic reactions, removing contaminants and organic solvents used in extraction processes. Samples are efficiently recovered and concentrated in as little as 6µl for sensitive downstream applications. (Scalable elution volume)

QUICK PROTOCOL (ABRIDGED)

1. Sample DNase digestion reaction is as follows (Adjust accordingly):

RNA in water or TE buffer	10.0 µls
10X reaction buffer	2.5 µls
1 unit DNase (1 unit/µl)	1.0 µl
Nuclease-free water	11.5 µls
Total reaction volume	**25.0 µls**

2. Mix well and incubate at 37°C for 10 minutes.

3. Add 4 volumes of RNA Buffer to every volume of RNA sample.

4. Spin through NucleoPur™ Column for 30 seconds. (10,000 x g)

5. Add 200µl wash buffer and spin for 30 seconds. (10,000 x g)
6. Repeat previous step and spin for 1 minute. (16,000 x g)
7. Place NucleoPur™ Column in a new microcentrifuge tube.

8. Add 6-35µl H_2O, wait for 30 seconds then spin for 1 minute at top speed.

Note: All centrifugation steps should be performed at room temperature.

PROTOCOL

NOTES: Be sure to add an appropriate volume of ethanol (95-100%) to the Wash Buffer Concentration before use. See bottle label for volume.

1. The sample DNase digestion reaction is as follows

(Adjust accordingly):

RNA in water or TE buffer	10.0 µls
10X reaction buffer	2.5 µls
1 unit DNase I (1unit/µl)	1.0 µl
Nuclease-free water	11.5 µls
Total reaction volume	**25.0 µls**

2. Mix well and incubate at 37°C for 10 minutes.
3. Add 4 volumes of RNA Buffer to every volume of RNA sample and mix.
   For RNA samples <25µl, add 100µl RNA Buffer.
4. Pipet mixture into a Benev NucleoPur™ Column with collection tube attached.
5. Centrifuge for 30 seconds. (10,000 x g)
6. Add 200µl of Wash Buffer to the column and centrifuge for 30 seconds. (10,000 x g)
7. Repeat step 4 and centrifuge for 1 minute. (16,000 x g)
8. Discard collection tube and place NucleoPur™ Column in a new microcentrifuge tube.
9. Add 6-35µl nuclease-free water directly to the column matrix. Wait for 30 seconds and centrifuge at top speed for 1 minute.

TROUBLESHOOTING

Low RNA yield:

Incorrect ratio of DNA RNA Buffer to RNA sample.	Make certain that 4 volumes of RNA Buffer is added to every 1 volume of RNA sample. For samples under 25µls, add 100µls of RNA Buffer.
Ethanol was not added to Wash Buffer.	Ethanol must be added to the Wash Buffer Concentrate. Add an appropriate volume of ethanol and repeat procedure.
Incorrect elution buffer.	DNA will be efficiently eluted with a low salt buffer such as TE pH 8.0 or water.
Centrifugation too slow.	Centrifuge at 16,000 x g.

RELATED PRODUCTS

ITEM	CATALOG #	SIZE
RNA Purification System	RPS0001S	50 RXN

ORDERING INFORMATION

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