



IG® RNA Cleanup Kit

Manual

Catalog #	4003	4005
Package Size	25 preps	100 preps



15-27°C Storage

Room Temperature



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Intact Genomics, Inc.

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Description:

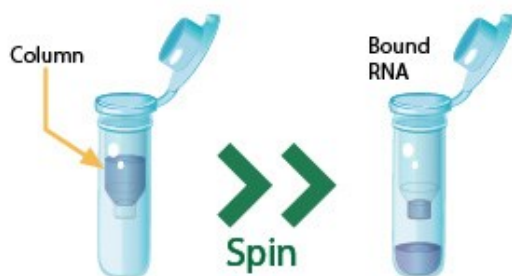
Intact Genomics is your resource for simple and fast cleanup of RNAs including sgRNAs. This kit purifies and concentrates up to 50 µg of RNA from enzymatic reactions. We recommend using this kit following sgRNA synthesis via the IG[®] sgRNA Synthesis Kits to remove proteins, salts and most unincorporated nucleotides. The Intact Genomics RNA Cleanup Kit has a simplified workflow, as shown below, and is ideal for cleanup and concentration of RNA after enzymatic treatments such as DNase I, Proteinase K, labeling, capping or *in vitro* transcription (IVT).

Workflow

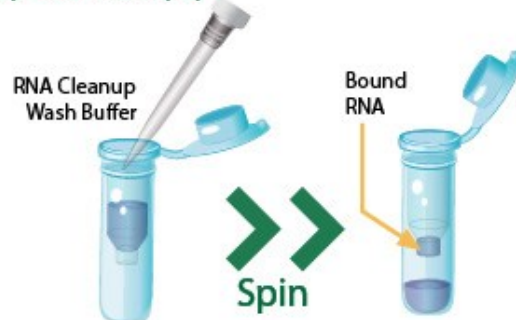
1.) Dilute Sample and Mix with Ethanol/Binding Buffer



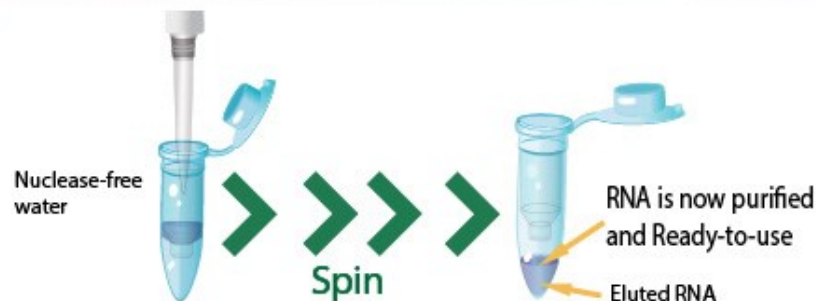
2.) Bind RNA to Matrix



3.) Wash Matrix (x2)



4.) Elute RNA



Benefits:

The Intact Genomics IG® RNA Cleanup Kit is a perfect choice for simple and fast cleanup of up to 50 µg of RNA. Below is a sampling of the key benefits.

- Rapid and simplified workflow with a single wash buffer
- Efficiently purify RNA ≥ 25 nt (a simple modification enables purification of RNA ≥ 15 nt)
- Option to Elute in 20-50 µL for more concentrated RNA than the standard 100 µL
- Standard 70-100% RNA recovery
- Customizable buffer options for difficult reactions
- Purified RNA is ready for use in a wide variety of downstream applications including: RT-PCR, RNA library prep for NGS, formation of RNP complexes for genome editing, microinjection, RNA labeling, and transfection.

Components and Storage:

The IG® RNA Cleanup Kit contains the items below. Store all components at Room Temperature.

- RNA Cleanup Columns (50 µg capacity)
- Collection Tubes
- RNA Cleanup Binding Buffer
- RNA Cleanup Wash Buffer (concentrated)
- Nuclease-free Water

IG® RNA Cleanup Kit Protocol:

- The RNA Cleanup Binding Buffer may form a precipitate. Should this occur, warm to room temperature to re-dissolve before use.
- Add 4 volumes of molecular biology grade ethanol ($\geq 95\%$) to one volume of RNA Cleanup Washing Buffer.
- Centrifugation steps should be at room temperature.
- Intact Genomics IG® RNA Cleanup Kit will purify RNA ≥ 25 nt. A modification in step 2 allows for the purification of RNA as small as 15 nt (the addition of 2 volumes of ethanol shifts the cutoff size of RNA binding from 25 nt down to 15 nt).

1. Add 100 µl RNA Cleanup Binding Buffer to the 50 µl sample.

The starting sample volume of 50 µl is recommended, although for smaller samples, nuclease-free water can be used to adjust the volume. If your sample is larger than 50µl, please scale buffer columns accordingly.

2. Add 150 µl of ethanol (≥ 95%) to your sample and mix by pipetting or flicking the tube. Do not vortex.

This is for binding RNA ≥ 25 nt in length. Should you need to bind RNA ≥15 nt, add 300 µl of ethanol to your sample instead of 150 µl.

3. Insert the included column into a collection tube and load your sample onto the column. Close the cap and then spin for 1 minute. Discard the flow-through.

If your sample is diluted > 900 µl, load a portion of the sample. Then spin and repeat as necessary.

4. Insert the column back into the collection tube. Add 500 µl RNA Cleanup Wash Buffer, spin for 1 minute and discard the flow-through. Repeat this step 1X.

5. Transfer the column to an RNase-free 1.5 ml microfuge tube (not included).

It is important to note that the tip of the column should not come into contact with the flow through. If this occurs, re-spin for 1 minute to ensure traces of salt/ethanol are not carried over to the final sample.

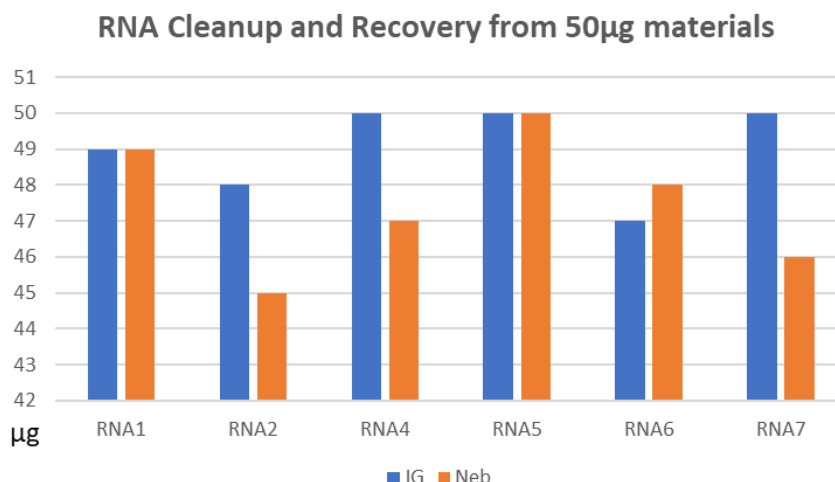
6. Elute RNA with nuclease-free water according to the following table:

Kit	Elution Volume	Spin Time
Intact Genomics RNA Cleanup Kit	20-100µl	1 Minute

The eluted RNA should be stored at –80°C. Yield will slightly increase if a larger volume is used, however the RNA will be less concentrated.

Comparison Data:

We compared Intact Genomics’ IG[®] RNA Cleanup Kit with New England Biolabs’ Monarch[®] RNA Cleanup Kit (50 µg) with 7 RNA samples (50 µg each) and 50 µl of elution buffer. The differences in the eluted amounts are not statistically significant.



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