



Volume 26, Number 3, 2010

INSIDE

Phenol-Free Total RNA Purification Kit p. 2

High quality total RNA purified without phenol/chloroform

Formaldehyde-Free RNA Gel Kit p. 6

Denaturing RNA gel electrophoresis without formaldehyde



Singing changes expression of over 800 genes in Zebra Finches and activates micro RNA's that are implicated as regulators of song learning.

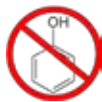
Wesley C. Warren et.al. [Nature](#) 2010, 464:757-762



Go Green with
AMRESCO RNA products

RNA Purification

Phenol-Free Total RNA Purification Kit



Eliminate phenol and chloroform

Save Time

- Rapid spin-column purification and concentration
- Total RNA purification from multiple samples in 20 minutes

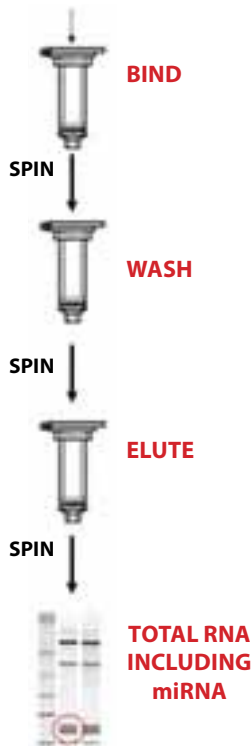
Reduce hazardous chemical use

- Eliminate phenol and chloroform
- Reduce interference by residual phenol in downstream applications

Enhance recovery of total RNA

- Efficient recovery from small sample sizes
- Applicable to a wide range of sources

Lyse cultured cells, tissues, bacteria, etc.



AMRESCO's Phenol-Free Total RNA Purification Kit provides a rapid method for the isolation and purification of total RNA. In as little as 20 minutes, all sizes of RNA, from large mRNA and ribosomal RNA down to microRNA (miRNA) and small interfering RNA (siRNA), can be extracted without the use of phenol or chloroform. The kit is an excellent tool for studies comparing expression of miRNA to other RNA species since both miRNA and mRNA can be analyzed from the same sample without further purification. The kit can be used with samples from a wide range of organisms.

KIT INCLUDES

- Spin Columns
- Elution Buffer
- Lysis Solution
- Collection Tubes
- Wash Solution
- Product Insert

PRODUCT DESCRIPTION	CODE	SIZE	PRICE
Phenol-Free Total RNA Purification Kit	N788-KIT	1 Kit	\$178.00
Sufficient for 50 RXNs			

Small RNAs are Efficiently Recovered

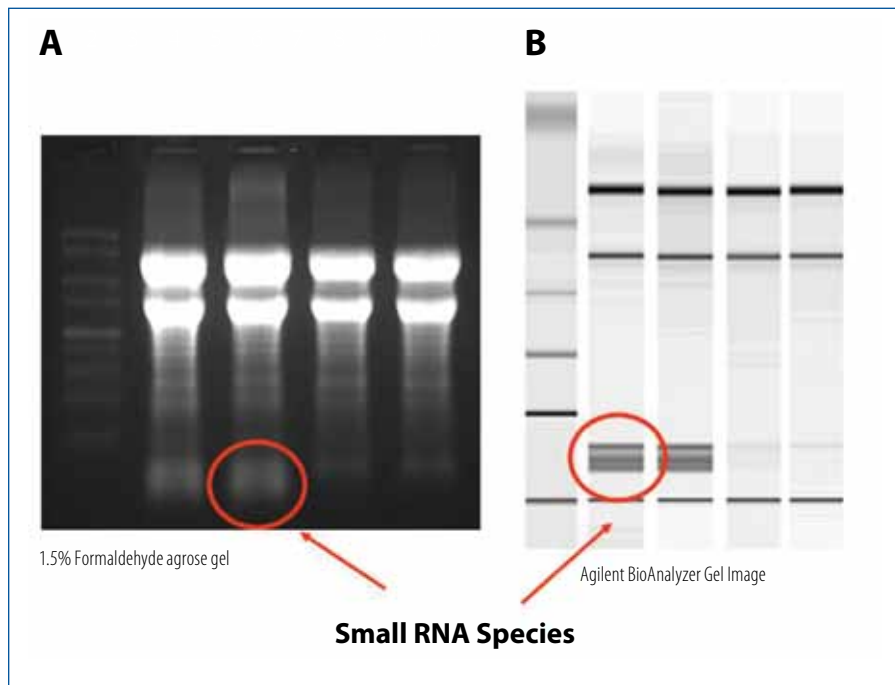


Figure 1. Size Range of RNA purified with Phenol-Free Total RNA Purification Kit versus a competitor's kit. Total RNA was isolated from 1×10^9 *E. coli* cells with Phenol-Free Total RNA Purification Kit and a competitor's kit. Five μL and 1 μL of isolated RNA are analyzed on an agarose gel (Panel A) and the Agilent[®] 2100 BioAnalyzer RNA Nano 6000 chip (Panel B), respectively. **Note the presence of small RNA species (red circle) in the samples isolated via Phenol-Free Total RNA Purification Kit and the absence of these RNA species in the competitor RNA preparation.**

RNA Recovered from as Little as One Cell

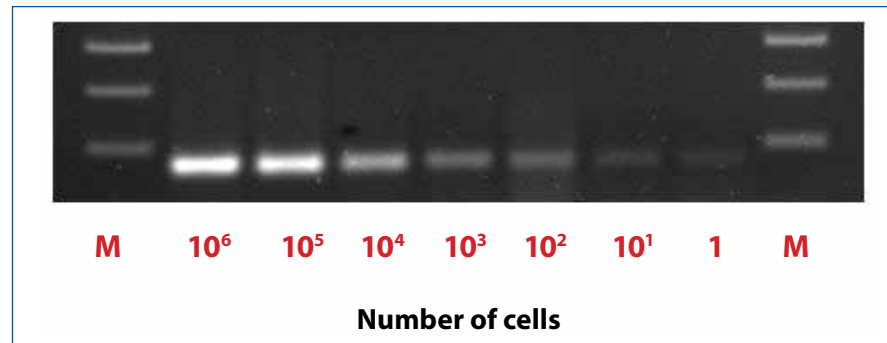


Figure 2. RNA Recovery by Phenol-Free Total RNA Purification Kit Approaches Single Cell Levels. Total RNA was extracted from serial dilutions of 293 HEK cells over a range of 1×10^6 cells to a single cell. The RNA was polyadenylated and reverse transcribed with an oligo dT primer. Three μL of the reverse transcription reaction was amplified by PCR with primers to the human beta-actin gene. M is the marker lane. **PCR products of beta-actin were detected from as little as a single cell.**

RELATED PRODUCTS

- **Formaldehyde-Free RNA Gel Kit**
 - Prepare and run RNA gels on your bench top
 - Instant band visualization
- **RNA Markers**
 - RiboReady[™] 100bp RNA Ladder (N603)
 - RiboReady[™] 1kb RNA Ladder (N604)

To Place an Order in the USA Call: 800-829-2805 or Order On-line at www.amresco-inc.com

Both Small and Large RNAs are Easily Amplified from the Same Sample

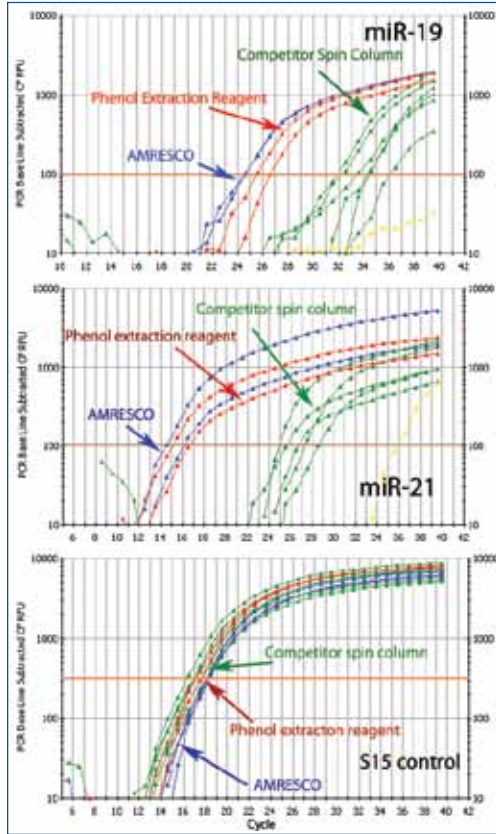


Figure 3. Amplification of total RNA isolated from Phenol-Free Total RNA Purification Kit versus competitor kits.

Amplification of RNA from Phenol-Free Total RNA Purification Kit (blue), a competitor's phenol-based extraction reagent (red), and a competitor's spin column kit (green). RNA isolated from 0.75 million HeLa cells was polyadenylated, and 4 microliters of the polyadenylated RNA was used in a 20 μ L reverse transcription reaction with a poly T adaptor primer. One microliter of the reverse transcription was used in a 20 μ L qPCR reaction with primers to the human microRNAs (*miR-19* and *miR-21*) and large mRNA (*S15*). **Amplification of RNA purified with Phenol-Free Total RNA Purification Kit matched amplification of both miRNAs and mRNA amplified with the phenol based extraction reagent. Amplification of microRNAs from the competitor's spin column technology was not detectable.**

Phenol-Free Total RNA Purification Kit is Sensitive and Linear for Both Large & Small RNAs

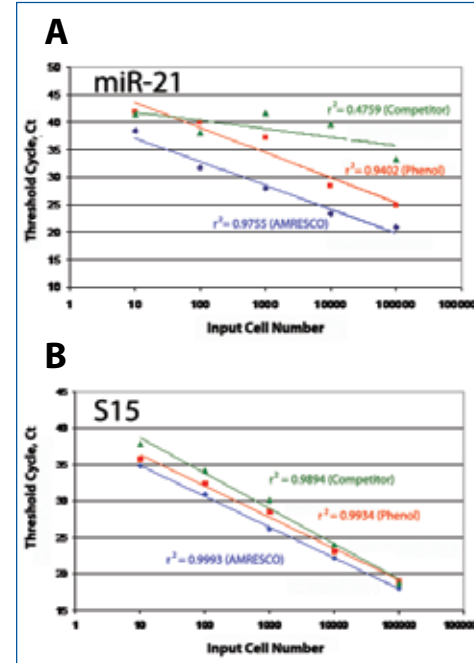


Figure 4. Linearity and sensitivity of large and small RNAs recovered from AMRESKO's kit versus competitor's spin column kit and phenol-based RNA extraction method.

Total RNA was isolated from 10 to 100,000 HeLa cells with Phenol-Free Total RNA Purification Kit (blue), a competitor's spin-column kit (green) and a phenol-based RNA extraction method (red). Relative expression of miR-21 (Panel A), and S15 (Panel B) was determined by RT-qPCR of total RNA samples. One microliter of isolated RNA was subjected to a 20 μ L reverse transcription using miR-21 stem-loop reverse primer or oligo dT primer. Two microliters of the reverse transcription was added to a 20 μ L real-time PCR reaction with primers to detect the human miR-21 (Panel A) and the S15 transcripts (Panel B). The resulting threshold cycle (Ct) values were plotted against input cell number. **RNA isolated with Phenol-Free Total RNA Purification Kit had the best linearity (higher R2) and sensitivity (lower Ct) for both large RNA (S15) and small RNA (miR-21).**

MicroRNA is Recovered From Many Sources, Including Plasma

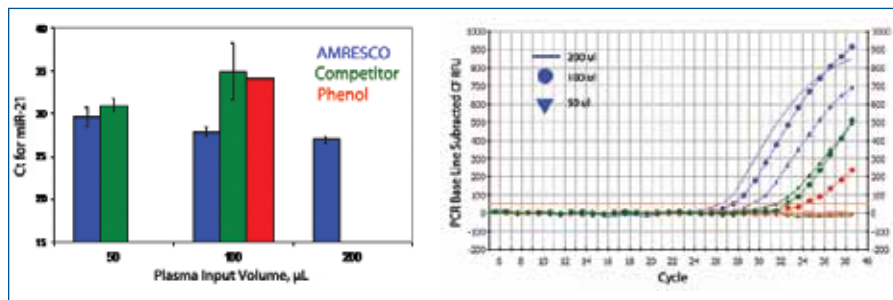


Figure 5. Isolation of Total RNA from Plasma by Phenol-Free Total RNA Purification Kit versus Competitors.

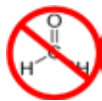
Total RNA was isolated from 50, 100 or 200 µL of rat plasma in triplicates using Phenol-Free Total RNA Purification Kit (blue), a competitor spin column kit (green) and a phenol-based RNA extraction method (red). Two microliters of the isolated RNA was subjected to a 20 µL reverse transcription with miR-21 stem-loop reverse primer or oligo dT primer. Three microliters of the reverse transcription reaction was transferred to a 20 µL real-time PCR reaction with primers to the human miR-21. **Phenol-Free Total RNA Purification Kit is the only product that showed (1) consistent detection of miR-21 transcripts across all input volumes and (2) Ct values correlated to input volume (decrease Ct with increase input).**

Kit Specifications

Binding Capacity per Column	Up to 50 µg RNA
Maximum Loading Volume Per Spin Column	650 µL
Size of Purified RNA	All sizes, including < 200 nt
Time to Complete 10 Purifications	20 minutes
RNA Yields	
Tissue	
Liver (10 mg)	30 µg
Kidney (10 mg)	10 µg
Brain (10 mg)	12 µg
Blood (hamster, 100 µL)	5 µg
Cells	
Hela (1.0 x 10 ⁶)	15 µg
CHO (1.0 x 10 ⁶)	11 µg
Yeast (1.0 x 10 ⁶)	30 µg
E. Coli (1.0 x 10 ⁶)	43 µg

RNA Electrophoresis

Formaldehyde-Free RNA Gel Kit



Eliminate formaldehyde and ethidium bromide

Save Time

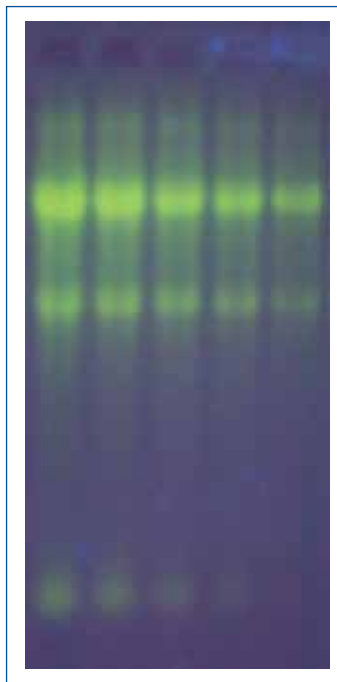
- Complete loading buffer denatures RNA, provides tracking dyes plus an RNA dye in a single solution

Reduce hazardous chemical use

- Run your RNA gels directly on your bench top
- Non-hazardous RNA dye stains the RNA bands a brilliant fluorescent green and eliminates the use of ethidium bromide (sensitivity is similar to Ethidium Bromide)

Versatile

- Compatible with downstream applications including Northern Blots



KIT INCLUDES

- Formaldehyde-Free RNA Gel Solution, 10X
- Formaldehyde-Free RNA Running Buffer, 10X
- Formaldehyde-Free RNA Loading Buffer, 2X
bromophenol blue and xylene cyanol included as tracking dyes.

PRODUCT DESCRIPTION	CODE	SIZE	PRICE
Formaldehyde-Free RNA Gel Kit	N726-KIT	1 Kit	\$179.90

RELATED PRODUCTS

- **Agarose I™ (0710)**
 - An all purpose agarose suitable for both analytical and preparative applications
- **NucleasEliminator® (E891)**
 - A ready-to-use liquid solution that removes and deactivates nuclease contamination even at high concentrations

Formaldehyde-Free Gels have Comparable Sensitivity to Ethidium Bromide Stained Gels

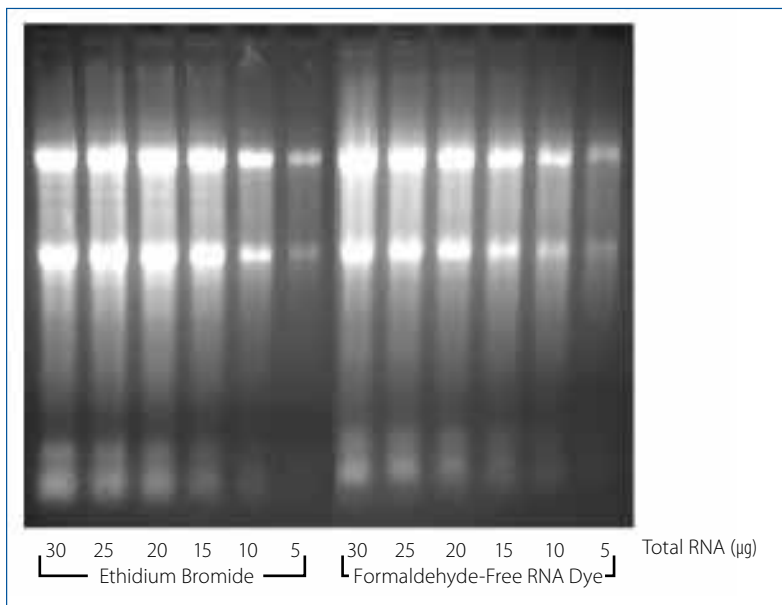


Figure 6. Comparison of Formaldehyde-Free RNA dye to ethidium bromide staining.

Total RNA was extracted from K562 cells with Ribozol® RNA Extraction Reagent (N580). Samples were denatured in Formaldehyde-Free RNA Loading Buffer containing either ethidium bromide or Formaldehyde-Free RNA dye and incubated for 10 minutes at 65°C. The samples were applied to a 2% Formaldehyde-Free RNA Agarose Gel (Agarose I™, Code:0710) and resolved for 1.5 hours at 5.1 V/cm. Image capture was performed with a Syngene GBox-HR Gel Doc System with a SYBR® Green filter. **Formaldehyde free RNA gel dye is as sensitive as ethidium bromide.**

Formaldehyde-Free Gels Compatible with Downstream Applications

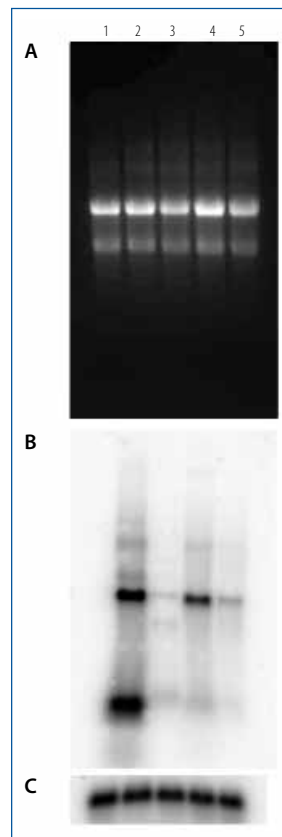


Figure 7. Northern blot analysis of PGK1 mRNA resolved on Formaldehyde-Free RNA Gel.

Data provided by Jeff Coller, Ph.D., Center for RNA Molecular Biology, Case Western Reserve University, Cleveland, Ohio. Gel A: 20 µg of total yeast RNA was applied per lane to an agarose Formaldehyde-Free RNA Gel and resolved at 8V/cm. Bands were visualized on a UV transilluminator Gel B: Bands were transferred to Hybond N membrane and probed overnight with a radiolabeled oligonucleotide to PGK1 mRNA. Gel C: Membranes were stripped and reprobed overnight with an oligonucleotide to SCR1 RNA loading control. **Formaldehyde free RNA gel kit is fully compatible with northern blotting.**

Lane 1: Untransformed control, Lane 2: High Copy PGK1 vector, Lane 3: Low Copy PGK1 vector, Lane 4: High Copy PGK1 vector, Lane 5: Low Copy PGK1 vector

*“ Even without formaldehyde . . .
[Formaldehyde-Free RNA Gel Kit]. . .
very good denaturing condition for RNA. ”*

*Dr. Bernard Lam
Norgen Biotek Corp.*

InvestiGator



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from our mailing list

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The logo for AMRESKO, featuring the word "AMRESKO" in white, bold, sans-serif capital letters inside a blue rounded rectangle with a registered trademark symbol.

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