

# Purification Systems

## Isolate RNA...

...from a variety of sample materials, on a scale of your choosing.

Promega's Total RNA and mRNA Isolation Systems provide:

- **Your choice of purification method:** Promega provides a range of solution, membrane filtration-based and magnetic particle-based systems. You choose the separation method best suited to your needs.
- **Your choice of throughput:** Both membrane-based and magnetic-based RNA isolation systems are available in manual and automated formats.
- **Systems tailored to your purification scale:** Mini-isolation from small sample sizes to scaleable, solutions-based formats, there is a total RNA purification system for every need.
- **Direct isolation of total RNA or mRNA from biological materials:** Systems are available for direct isolation from a variety of starting materials.
- **Increased productivity:** Save time and labor with our easy to use protocols.

### Purify RNA From:

- Blood
- Tissue
- Cultured Cells
- Plant Material
- Bacteria

## How can we help you achieve your goals today?

### A Comparison of RNA Isolation Systems.

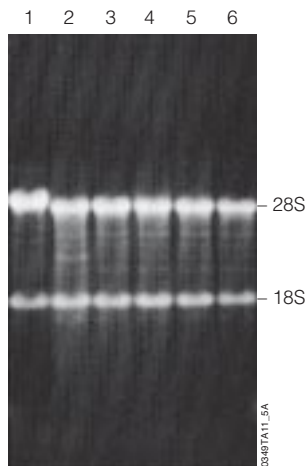
	Total RNA	Blood	Bacteria	Animal Tissue	Culture Cells	Plant Material	Manual Handling	Automated Handling	Magnetic Technology	Spin Protocol	Vacuum Protocol
<b>Total RNA Isolation Systems</b>											
RNAgents® Total RNA Isolation System (Cat.# Z5110)		●	●	●	●	●	●			●	
SV Total RNA Isolation System (Cat.# Z3100, Z3105)		●		●	●		●			●	●
SV 96 Total RNA Isolation System (Cat.# Z3500, Z3505)		●		●	●		●	●			●
MagneSil® Total RNA mini-Isolation System (Cat.# Z3351)		●		●	●			●	●		
<b>mRNA Isolation Systems</b>											
PolyAtract® Automated System (Cat.# Z5671)				●	●			●	●		
PolyAtract® System 1000 (Cat.# Z5400, Z5420)	●			●	●		●		●		
PolyAtract® mRNA Isolation Systems (Cat.# Z5200, Z5300)	●						●		●		

## RNAgents® Total RNA Isolation System

### Scalable solution-based total RNA purification for maximum flexibility

#### The RNAgents Total RNA Isolation System provides:

- **Speed:** The solution-based procedure can be completed in 90 minutes to 3 hours.
- **Scalable:** Isolate RNA from as little as 5mg, as much as 6g of tissue, or from 10<sup>5</sup>–10<sup>8</sup> cultured cells.
- **Simplicity:** A solution-based protocol for RNA isolation that can be performed with ease in any laboratory setting.
- **Performance:** Yield and purity for today's demanding applications (e.g. microarrays and qRT-PCR)
- **Complete system:** Purity total RNA from blood, tissue cells, plant leaf, etc.
- **Maximum flexibility:** Denaturing Solution available separately for maximum flexibility, allowing you the option to supply the other system components (phenol/chloroform, isoamyl alcohol and isopropanol).



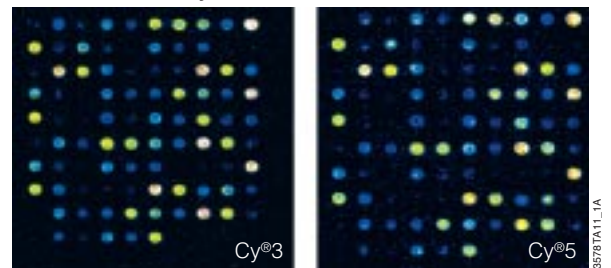
**Analysis of RNA isolated from various tissues with the RNAgents System.** RNA was isolated from the following sources: HeLa cells (lane 1), mouse intestine (lane 2), mouse spleen (lane 3), mouse lung (lane 4), mouse kidney (lane 5) and mouse liver (lane 6). Five micrograms of each sample was resolved on a 1% denaturing agarose gel (2.2M formaldehyde).

Product	Size	Cat.#
RNAgents® Total RNA Isolation System	Scalable	Z5110
For Laboratory Use.		

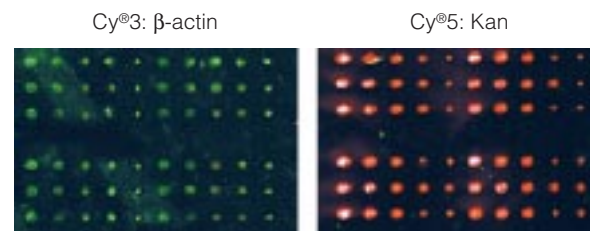
#### Average Yields of Total RNA from a Variety of Tissues.

Tissue	RNAgents Total RNA Yield
HeLa cells	1.6mg RNA/10 <sup>8</sup> cells
Human white blood cells	1.3mg RNA/10 <sup>8</sup> cells
Mouse intestine	2.3mg RNA/g tissue
Mouse spleen	8.3mg RNA/g tissue
Mouse lung	1.9mg RNA/g tissue
Mouse kidney	3.1mg RNA/g tissue
Mouse liver	6.6mg RNA/g tissue

#### E. coli Control Array



#### B. Jurkat Control Array



**Hybridization of fluorescent cDNA targets to E.Coli and Jurkat Cell Arrays. Panel A:** Hybridization results for the E. coli control array. One array was hybridized with a Cy<sup>3</sup> target and another with a Cy<sup>5</sup> target. **Panel B:** Hybridization results for the Jurkat Control Array. Fluorescent Cy<sup>3</sup> and Cy<sup>5</sup> targets were co-hybridized to the same array, demonstrating specificity for their complementary sequences.

# Purification Systems

## SV Total RNA Isolation System

**Simple and reliable total RNA purification from tissues, cultured cells or white blood cells**

The SV Total RNA Isolation System provides:

- **Speed:** The procedure can be completed in 60 minutes.
- **Safety and convenience:** Silica membrane based, no phenol extraction required.
- **Confidence in your results:** Includes a DNase I treatment step to ensure removal of genomic DNA. No detectable DNase I carryover to the final RNA preparation.
- **Peace of mind:** Only system that includes DNase treatment step in the standard protocol.
- **Applications tested RNA:** RNA suitable for all routine molecular biology applications, including RT-PCR and Northern blotting.

### Average Yields of Total RNA Using SV Total RNA Isolation System.

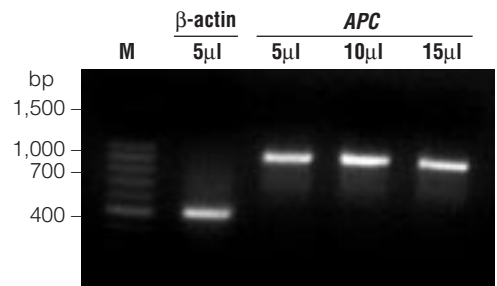
Samples	Maximum Amount to Process	Avg. Yield per Prep (µg)	Avg. Yield per mg Tissue (µg)	A <sub>260</sub> /A <sub>280</sub>
<b>Mouse Tissues</b>				
Liver	30mg	131	4.4	1.9
Kidney	20mg	44	2.2	1.9
Spleen	15mg	79	5.3	1.9
Brain	60mg	39	0.65	2.1
Muscle	30mg	22	0.73	2.1
<b>Rat Tissues</b>				
Pancreas	30mg	100	3.5	1.9
Heart	60mg	16	0.27	2.1
Lung	60mg	36	0.6	2.1
<b>Bacteria</b>				
<i>E. coli</i>	1 x 10 <sup>9</sup> cells	36	N/A	2.0
<b>Yeast</b>				
<i>S. cerevisiae</i>	4 x 10 <sup>7</sup> cells	19	N/A	2.1
<b>Plant</b>				
Tomato Leaf	30mg	4.6	0.15	2.0
<b>Cell Line</b>				
RAW264.7	5 x 10 <sup>6</sup> cells	51	N/A	2.1

N/A = Not Applicable.

Average yield is that of RNA only. Other commercial systems may appear to give higher yields, but this is frequently due to DNA carryover.

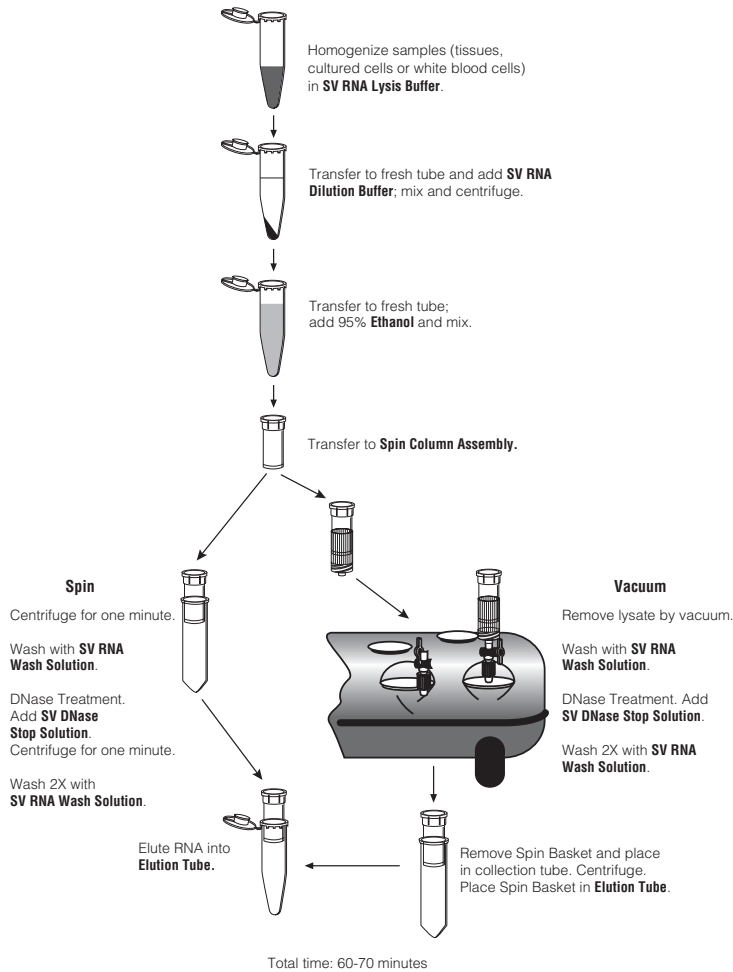
Product	Size	Cat.#
SV Total RNA Isolation System	10 preps	Z3101
	50 preps	Z3100
	250 preps	Z3105

For Laboratory Use.



RNA isolated from 1ml of human blood using the SV Total RNA Isolation System. RT-PCR was performed using the indicated volumes of eluted RNA and primers complementary to human  $\beta$ -actin or human *Adenomatous Polyposis Coli* (APC) gene with the Access RT-PCR System<sup>(a,b)</sup> (Cat.# A1280, A1250).

*The vacuum protocol for the SV Total RNA Isolation System requires use of the Vacuum Adapters and a vacuum manifold such as Promega's Vac-Man<sup>®</sup> Laboratory Vacuum Manifold (20-sample capacity).*

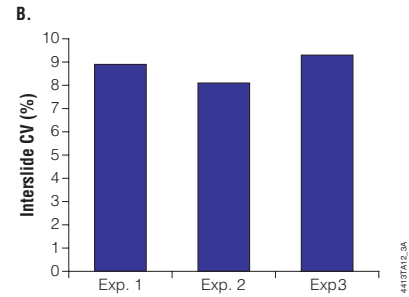
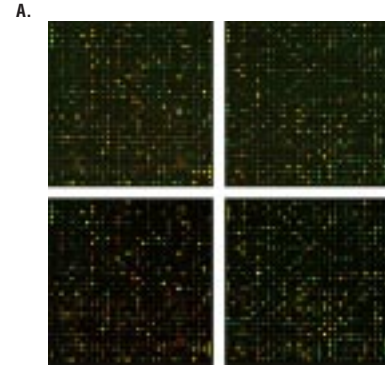


Schematic of the SV Total RNA Isolation System protocol.

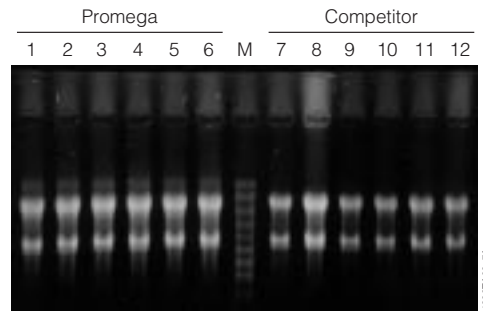
*High-quality RNA from the SV Total RNA isolation system ensures success in subsequent experiments such as microarrays.*

(a) The PCR process is covered by patents issued and applicable in certain countries\*. Promega does not encourage or support the unauthorized or unlicensed use of the PCR process. Use of this product is recommended for persons that either have a license to perform PCR or are not required to obtain a license. \*In the U.S., effective March 29, 2005, U.S. Pat. Nos. 4,683,195, 4,965,188 and 4,683,202 will expire. In Europe, effective March 28, 2006, European Pat. Nos. 201,184 and 200,362 will expire.

(b) U.S. Pat. Nos. 4,966,964, 5,019,556 and 5,266,687, Australian Pat. Nos. 616,881 and 641,261 and other pending and issued patents, which claim vectors encoding a portion of human placental ribonuclease inhibitor, are exclusively licensed to Promega Corporation.



**High degree of experimental reproducibility using RNA purified by the SV Total RNA Isolation System.** **Panel A.** Subgrids from four representative 4K human cancer arrays (provided by Corning Incorporated) are shown. Labeled cDNA was generated with the ChipShot™ components of the Pronto!™ Plus System using 5µg of total RNA purified using the SV Total RNA System. **Panel B.** Reproducibility was assessed by determining the %CV for 4000 features from 3-4 arrays. The inter-slide %CV was <10% indicating a high degree of reproducibility within and between experiments as a result of high quality input total RNA.



**Formaldehyde gel electrophoresis of RNA.** RNA was isolated from 30mg of liver tissue following the SV Total RNA Isolation System protocol (lanes 1–6) and a competitor's protocol (lanes 7–12). Ten percent (10µl) of the final prep volume of 100µl for each sample was analyzed by electrophoresis in a 1% MOPS-EDTA-formaldehyde agarose gel and visualized by including 0.1µg/µl ethidium bromide in the sample loading buffer. Promega's system consistently yielded an average of 100µg. The competitor's system yielded an average of 70µg with wide variability.

# Purification Systems

## SV 96 Total RNA Isolation System

### High-throughput 96-well total RNA purification

#### The SV 96 Total RNA Isolation System provides:

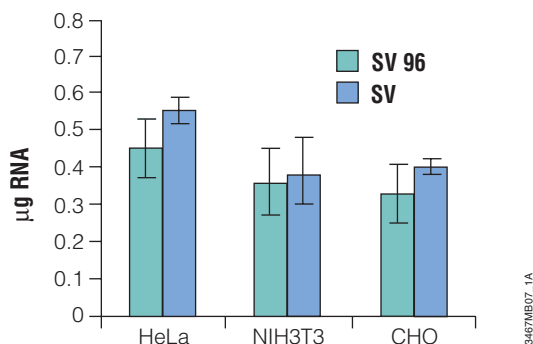
- **Efficiency:** Save labor with a walk-away high-throughput 96-well purification automated method. 96-samples processed is less than 1 hour.
- **A unique design:** The novel vacuum manifold eliminates waste handling, and the novel plate design prevents cross-contamination during sample processing.
- **Confidence in your results:** A DNase I treatment step is included in the protocol to ensure removal of genomic DNA contamination. There is no detectable DNase carryover into the final RNA preparation.
- **An applications-tested system:** Tested to ensure purified RNA performs optimally in RT-PCR.
- **Hassle-free automated methods:** Automated methods and support information available at: [www.promega.com/automethods/](http://www.promega.com/automethods/)

Product	Size	Cat. #
SV 96 Total RNA Isolation System	1 x 96 each	Z3500
	5 x 96 each	Z3505

For Laboratory Use.



Automated isolation of total RNA using the SV 96 Total RNA Isolation System on the Beckman Coulter Biomek® FX.



The SV 96 Total RNA Isolation System provides consistent and reliable recovery of high quality total RNA. Panel A: RNA yields from several cell lines using  $1 \times 10^6$  cells is consistently 0.3µg or more.

*Total RNA can be purified from 96 samples at once in less than an hour.*

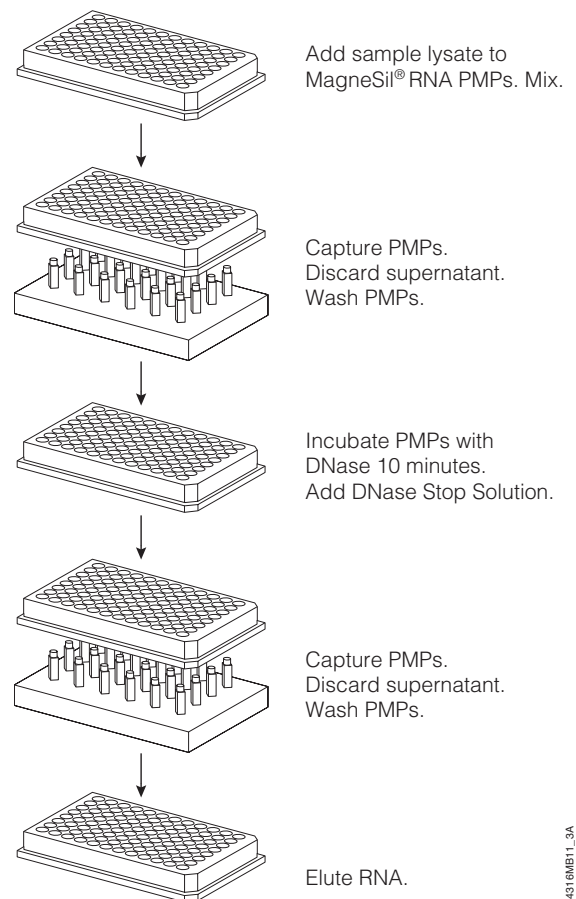
## MagneSil® Total RNA mini-Isolation System

### High-throughput 96-well or 384-well purification of highly concentrated total RNA

#### The MagneSil Total RNA mini-Isolation System provides:

- **A simple, fast method:** Elution volumes as low as 15µl give concentrated RNA. Only 30 minutes to process one 96-well plate and 50 minutes to process a single 384-well plate on a Beckman Coulter Biomek® FX laboratory workstation.
- **Confidence in your results:** A DNase I treatment step is included in the protocol to ensure removal of genomic DNA contamination.
- **A performance-tested system:** RNA rigorously tested in end-point RT-PCR and quantitative, real-time RT-PCR.
- **Efficiency:** Save labor with a walk-away high-throughput 96-well purification automated method. 96-samples processed in only 30 minutes.
- **Designed for screening protocols:** Total RNA isolation from small sample sizes (up to 10<sup>5</sup> cultured cells, 20mg tissue or 20µl whole blood/well in a 96-well format, and up to 10<sup>3</sup> cultured cells or 5µl whole blood/well in a 384-well format).
- **Hassle-free automated methods:** Automated methods and support information available at: [www.promega.com/automethods/](http://www.promega.com/automethods/)

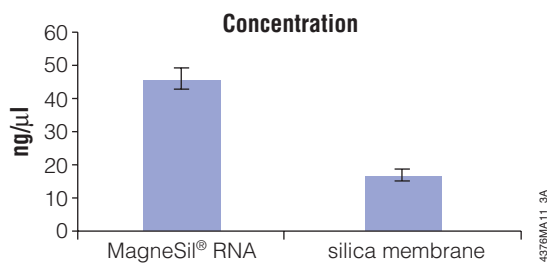
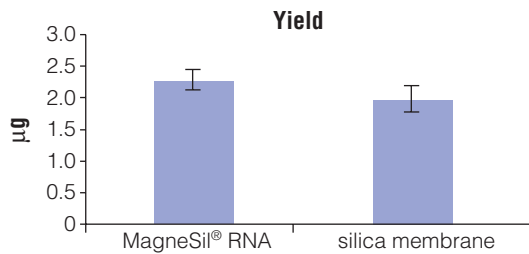
Product	Size	Cat.#
MagneSil® Total RNA mini-Isolation System	4 plates	Z3351
For Laboratory Use.		



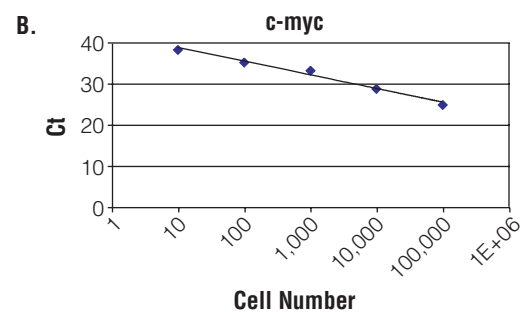
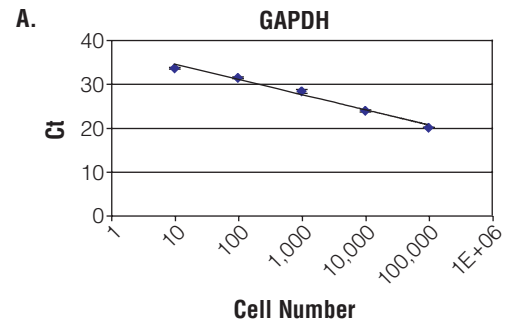
Overview of the MagneSil Total RNA mini-Isolation System protocol.

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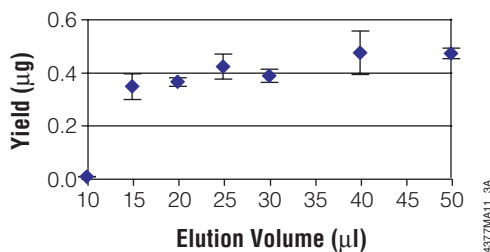
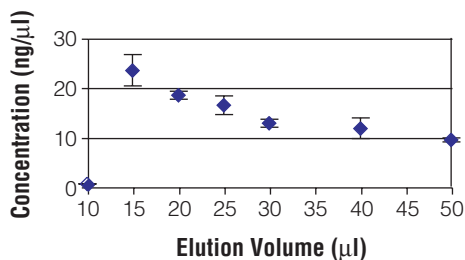
# Purification Systems



**Yield of total RNA from HeLa cells using the MagneSil Total RNA mini-Isolation System.** Total RNA was isolated from  $1 \times 10^5$  HeLa cells/well using either the MagneSil Total RNA mini-Isolation System or a silica membrane based purification method. Total RNA concentration and yield were calculated using RiboGreen<sup>®</sup> RNA Quantitation Reagent (Molecular Probes).



**Real-time RT-PCR analysis of RNA purified using the MagneSil Total RNA mini-Isolation System.** To measure linearity and sensitivity of RNA purification, GAPDH (abundant mRNA) and c-myc (rare mRNA) were amplified in real-time RT-PCR from RNA purified from various titrations of HeLa cells. Twenty microliter aliquots of total RNA isolated from a dilution series of HeLa cells in a 96-well plate were used as template in 100µl reverse transcription reactions. **Panel A:** Five microliter aliquots ( $n=3$ ) of the RT reaction were used for PCR of a GAPDH target. **Panel B:** Five microliter aliquots ( $n=1$ ) of the RT reaction were used for PCR of a c-myc target. GAPDH signal and c-myc signal were detected from as little as 10 cells.



**MagneSil Total RNA mini-Isolation System: titration of elution volume.** Total RNA was isolated from  $2 \times 10^4$  HeLa cells/well using the MagneSil Total RNA mini-Isolation System, and eluted in various volumes from 50µl down to 10µl. Yield and concentration obtained for each elution volume are shown. Total RNA concentration was calculated using RiboGreen<sup>®</sup> RNA Quantitation Reagent (Molecular Probes).

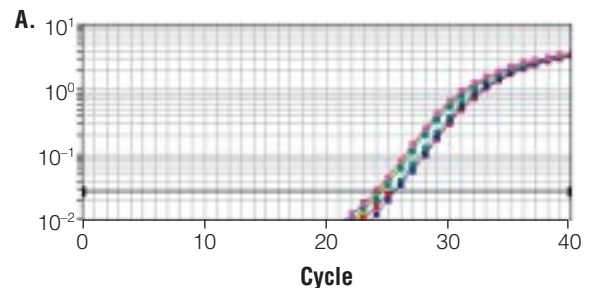
## PolyATtract® Automated System

### High-throughput, mRNA purification for sensitive applications

#### The PolyATtract Automated System provides:

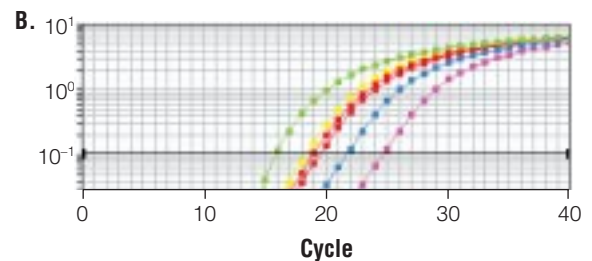
- **Your choice of starting material:** Linear recovery of poly(A)<sup>+</sup> RNA from 0.31–20 µg of eukaryotic total RNA. Alternatively, work with 0.02–5 mg of eukaryotic tissue in lysate or 10<sup>2</sup>–10<sup>6</sup> of cultured mammalian cells.
- **Strong, selective mRNA binding:** A magnetic-based purification method for mRNA capture that is based on hybridization of poly(A)<sup>+</sup> mRNA to a biotinylated oligo(dT) primer. The bound mRNA is then captured by streptavidin-coated paramagnetic particles. Removal of >99% of ribosomal RNA as judged by quantitative, real-time RT-PCR.
- **Confidence with expression profiles:** Expression profiles remain the same as samples from total RNA and poly(A)<sup>+</sup> RNA.
- **Freedom from well-to-well cross-contamination:** No detectable well-to-well contamination as judged by RT-PCR.
- **Efficiency:** Save labor with walk-away automation of mRNA purification directly from cultured cells, tissue lysates or purified total RNA.
- **Hassle-free automated methods:** Automated methods and support information available at [www.promega.com/automethods/](http://www.promega.com/automethods/)

Product	Size	Cat.#
PolyATtract® Automated System	4 x 96	Z5671
For Laboratory Use.		



#### GAPDH Target

- Total RNA (Avg. Ct = 23.0)
- mRNA From Total RNA (Avg. Ct = 24.9)
- mRNA From Cells (Avg. Ct = 23.8)



#### 18S rRNA Target

- 1:10 Total RNA
- 1:100 Total RNA
- mRNA From Total RNA
- mRNA From Cells
- 1:1,000 Total RNA
- 1:10,000 Total RNA

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**Detection of Ten copies of GAPDH and c-myc using real-time RT-PCR analysis of RNA purified using the MagneSil Total RNA Mini-Isolation System.** To measure linearity and sensitivity of RNA purification, GAPDH (abundant mRNA) and c-myc (rare mRNA) were amplified in real-time RT-PCR from RNA purified from various titrations of HeLa cells. Twenty microliter aliquots of total RNA isolated from a dilution series of HeLa cells in a 96-well plate were used as template in 100 µl reverse transcription reactions. **Panel A:** Five microliters aliquots (n=3) of the RT reaction were used for PCR of a GAPDH target. **Panel B:** Five microliter aliquots (n=1) of the RT reaction were used for PCR of a c-myc target.

# Purification Systems

## PolyATtract® mRNA Isolation Systems

### Scalable, flexible mRNA purification for all applications

#### The PolyATtract mRNA System provides:

- **Speed:** mRNA isolation is completed in 45 minutes or less.
- **Strong, selective mRNA binding:** A magnetic-based purification method for mRNA capture that is based on hybridization of poly(A)<sup>+</sup> mRNA to a biotinylated oligo(dT) primer. The bound mRNA is then captured by streptavidin-coated paramagnetic particles. Removal of >99% of ribosomal RNA as judged by quantitative, real-time RT-PCR.
- **Your choice of starting material:** Configurations available for large or small amounts of cells or tissues. mRNA isolation from total RNA in low throughput format (PolyATtract Systems I-IV) or directly from crude cell or tissue lysates in scaleable format (PolyATtract System 1000).
- **Convenience and ease-of-use:** mRNA isolation accomplished easily, with a simple, magnetic-based procedure.
- **Applications tested:** Highly pure mRNA suitable for molecular biology applications such as in vitro translation, cDNA synthesis, PCR, RNase protection, primer extension and Northern blots.

#### Average Yields of Poly(A)<sup>+</sup> RNA Using PolyATtract System 1000.

Sample	Size of Sample	Yield of mRNA	A <sub>260</sub> /A <sub>280</sub>	µg mRNA/g sample
Liver	1,000mg	220µg	2.08	220
Kidney	420mg	138µg	2.10	328
Spleen	390mg	282µg	2.15	72
Pancreas	400mg	148µg	2.11	370
Lung	160mg	16µg	2.09	100
NIH3T3	6 × 10 <sup>7</sup> cells	20µg	2.12	200

Product	Size	Cat.#
PolyATtract® System 1000 with Magnetic Stand	Scalable	Z5420
PolyATtract® System 1000 without Magnetic Stand	Scalable	Z5400
PolyATtract® mRNA Isolation System II with Magnetic Stand	3 isolations	Z5200
PolyATtract® mRNA Isolation System I (Refill for Z5200)	3 isolations	Z5210
PolyATtract® mRNA Isolation System III with Magnetic Stand	15 isolations	Z5300
PolyATtract® mRNA Isolation System IV (Refill for Z5300)	15 isolations	Z5310

For Laboratory Use.

## Ordering Information

Product	Size	Cat. #
RNAgents® Total RNA Isolation System*	Scalable	Z5110
RNAgents® Denaturing Solution*	120ml	Z5651

Product	Size	Cat. #
SV Total RNA Isolation System <sup>(a,b)*</sup>	10 preps	Z3101
	50 preps	Z3100
	250 preps	Z3105
SV RNA Red Blood Cell Lysis Solution*	200ml	Z3141
SV RNA Lysis Buffer*	50ml	Z3051

Product	Size	Cat. #
SV 96 Total RNA Isolation System <sup>(a)*</sup>	1 x 96 each	Z3500
	5 x 96 each	Z3505
SV RNA Lysis Buffer*	50ml	Z3051
SV RNA Wash Solution* (concentrated)	58.8ml	Z3091
Nuclease-Free Water*	150ml	P1195

Product	Size	Cat. #
MagneSil® Total RNA mini-Isolation System <sup>(a)*</sup>	4 plates	Z3351

Product	Size	Cat. #
PolyATtract® Automated System*	4 x 96	Z5671
Nuclease-Free Water*	150ml	P1195
Biotinylated Oligo(dT) Probe (50pmol/μl)*	35μl	Z5261
Streptavidin MagneSphere Paramagnetic Particles*	25ml	Z5482
PolyATtract® GTC Extraction Buffer*	120ml	Z5531

Product	Size	Cat. #
PolyATtract® System 1000 with Magnetic Stand <sup>(c)*</sup>	Scalable	Z5420
PolyATtract® System 1000 without Magnetic Stand <sup>(c)*</sup>	Scalable	Z5400

Product	Size	Cat. #
PolyATtract® mRNA Isolation System II with with Magnetic Stand*	3 isolations	Z5200
PolyATtract® mRNA Isolation System I* (Refill for Z5200)	3 isolations	Z5210
PolyATtract® mRNA Isolation System III with with Magnetic Stand*	15 isolations	Z5300
PolyATtract® mRNA Isolation System IV* (Refill for Z5300)	15 isolations	Z5310

### Hard Ware Devices/Accessories

Product	Size	Cat. #
Vac-Man® Laboratory Vacuum Manifold, 20-sample capacity	1 each	A7231
Vacuum Adapters	20 each	A1331
Vac-Man® 96 Vacuum Manifold	1 each	A2291
Wizard® SV 96 Binding Plates	10 pack	A2271
	100 each	A2278
MagneSil® Magnetic Separation Unit	1 each	A2231
MagnaBot® 96 Magnetic Separation Device	1 each	V8151
Heat Transfer Block	1 each	Z3271
MagnaBot® Spacer 1/8 inch	1 each	V8581
MagnaBot® Spacer	1 each	V8381
1/4 inch Foam Spacer	1 each	Z3301
PolyATtract® System 1000 Magnetic Separation Stand	1 each	Z5410

Cat.# P1195, Z5261, Z5482, Z5531 Cat.# A2271, A2278, Z3051, Z3091, P1195. For Laboratory Use.

<sup>(a)</sup>U.S. Pat. No. 6,218,531, Australian Pat. No. 745185 and other patents pending.

<sup>(b)</sup>Australian Pat. No. 730718 and other patents and patents pending.

<sup>(c)</sup>U.S. Pat. No. 5,693,784.

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 ChipShot and Pronto! are trademarks of Promega Corporation.  
 Cy is a registered trademark of Amersham Biosciences Ltd.  
 Biomek is a registered trademark of Beckman Coulter, Inc.  
 RiboGreen is a registered trademark of Molecular Probes, Inc.

