

# Sample Preparation

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# **Protein Sample Preparation**

# Magnetic Beads and Racks for Immunoprecipitation

#### See Also

#### Antibodies: page 276. Western blotting: page 221. Chemiluminescence detection: page 238.

#### SureBeads<sup>™</sup> Magnetic Beads System

SureBeads magnetic beads are designed for bioseparation techniques like immunoprecipitation (IP), co-immunoprecipitation (co-IP), and protein pull-down assays. SureBeads beads are superparamagnetic beads with surface activated hydrophilic polymers and chemically conjugated protein A or protein G that specifically bind to the Fc region of immunoglobulin. This chemistry enables high IgG binding and low nonspecific binding from a variety of biological samples.

Product features include:

- Faster IP yes to magnetization, no to centrifugation
- Easier IP ergonomically designed SureBeads magnetic rack magnetizes beads in seconds
- Use less antibody patented\* surface chemistry enables proper antibody orientation for optimal antigen binding
- Reproducible consistent IgG binding capacity ensures accurate, reproducible results
- Low cost to go magnetic priced similarly to leading agarose beads, and the 16-tube magnetic rack is included in the IP starter kit with two 3 ml vials of beads



\* U.S. patent 20100105879A1.

For More Information Web: bio-rad.com/immunoprecipitation Request or download bulletin: 6560

Catalog #	Description
1614011	SureBeads Protein A Magnetic Beads, 1 x 1 ml vial conjugated magnetic immunoprecipitation beads (10 mg beads/ml suspension)
1614013	SureBeads Protein A Magnetic Beads, 1 x 3 ml vial conjugated magnetic immunoprecipitation beads (10 mg beads/ml suspension)
1614021	SureBeads Protein G Magnetic Beads, 1 x 1 ml vial conjugated magnetic immunoprecipitation beads (10 mg beads/ml suspension)
1614023	SureBeads Protein G Magnetic Beads, 1 x 3 ml vial conjugated magnetic immunoprecipitation beads (10 mg beads/ml suspension)
1614916	16-Tube SureBeads Magnetic Rack, with separable magnets
1614811	SureBeads Trial Kit Protein A, 1 x 1 ml vial conjugated magnetic immunoprecipitation beads (10 mg beads/ml suspension), 4-tube magnetic rack
1614821	SureBeads Trial Kit Protein G, 1 x 1 ml vial conjugated magnetic immunoprecipitation beads (10 mg beads/ml suspension), 4-tube magnetic rack
1614833	SureBeads Starter Kit Protein A and G, 1 x 3 ml vial protein A and 1 x 3 ml vial protein G conjugated magnetic immunoprecipitation beads (10 mg beads/ml suspension), 16-tube magnetic rack
1614813	SureBeads Starter Kit Protein A, 2 x 3 ml vials protein A conjugated magnetic immunoprecipitation beads (10 mg beads/ml suspension), 16-tube magnetic rack
1614823	SureBeads Starter Kit Protein G, 2 x 3 ml vials protein G conjugated magnetic immunoprecipitation beads (10 mg beads/ml suspension), 16-tube magnetic rack

# **Protein Extraction**

Protein extraction tools such as cell lysis and extraction kits, as well as mini grinders, are available for extracting proteins from cultured cells and tissues.

# **ReadyPrep<sup>™</sup> Protein Extraction Kit**

The ReadyPrep protein extraction kit (total protein) provides a simple, rapid, and reproducible method for preparation of total cellular protein extracts from a wide variety of biological samples. Use of this kit generates protein samples that can be applied directly to a variety of applications, including IEF and 2-D gel electrophoresis.

# For More Information

Web: bio-rad.com/proteinextraction

Description

#### **Ordering Information**

Catalog # 1632086

ReadyPrep Protein Extraction Kit (Total Protein), 20 preps

### **Kits for Cell Lysis**

#### **Cell Lysis Kits**

These kits offer a gentle nondetergent solution to cell disruption and generate total protein samples that are ready to be applied to SDS-PAGE, IEF, and 2-D gel electrophoresis. The kits are based on a chaotropic protein solubilization buffer (PSB), which contains nondetergent sulfobetaine 201 (NDSB 201) along with urea, thiourea, and CHAPS for particularly effective solubilization (Vuillard et al. 1995). Cell lysis and extraction protocols are tailored for mammalian, plant, yeast, or bacterial samples.

### **Bio-Plex® Cell Lysis Kit**

This cell lysis kit is used to prepare cell lysates for western blot analysis. Its protein extraction procedure yields western blotting results similar to those generated by routine cell lysis and protein extraction protocols.

#### For More Information

Web: bio-rad.com/proteinextraction and /bioplexcelllysis Request or download bulletins: 3033, 3034, and 5517







#### See Also

Protein assays: page 20.

# **Protein Sample Preparation**

**Protein Sample Cleanup** 

#### bio-rad.com/proteinsampleprep

#### See Also Protein assays: page 20.

# ReadyPrep<sup>™</sup> Mini Grinders

For grinding small biological samples for high recovery of proteins (and nucleic acids), each mini grinder includes a 1.5 ml grinding tube containing a grinding resin and a matching pestle. The grinding resin is a neutral abrasive material made of high tensile strength microparticles that do not bind to proteins or nucleic acids. ReadyPrep mini grinders are disposable and are nuclease- and protease-free. They are a component of the MicroRotofor<sup>™</sup> lysis kit (mammal) and are also sold separately as a pack of 20. The mini grinder tubes fit conveniently in most benchtop centrifuges.



For More Information Web: bio-rad.com/proteinextraction

Ordering Inf	formation
Catalog #	Description
1632141	Mammalian Cell Lysis Kit, 15 preps, includes 50 ml protein solubilization buffer (PSB), ReadyPrep mini grinders (2 packs of 10 each)
1632142	Plant Cell Lysis Kit, 10 preps, includes 50 ml protein solubilization buffer (PSB), ReadyPrep 2-D cleanup kit (50 reaction size)
1632143	Yeast Cell Lysis Kit, 15 preps, includes 50 ml protein solubilization buffer (PSB), 15 ml yeast suspension buffer, 2 x 0.5 ml lyticase (1.5 U/µl)
1632144	Bacterial Cell Lysis Kit, 15 preps, includes 50 ml protein solubilization buffer (PSB), 25 ml bacteria suspension buffer, 1 ml lysozyme (1,500 U/µl)
Cell Lysis Kit	t Components
1632145	Protein Solubilization Buffer (PSB), makes 50 ml of solution
1632146	ReadyPrep Mini Grinders, includes 20 mini grinders, sufficient for twenty 100 mg extractions
Bio-Plex Cel	I Lysis Kits
171304011	Bio-Plex Cell Lysis Kit, 1 x 96-well, includes cell lysis and wash buffers, factor 1 and factor 2
171304012	Bio-Plex Cell Lysis Kit, 10 x 96-well, includes cell lysis and wash buffers, factor 1 and factor 2

# Protein Sample Cleanup

General-purpose cleanup kits and columns are available for the removal of salts and other contaminants.

#### **Protein Sample Cleanup Kit Selection Guide**

Kit Type and Catalog Number	Applications	Procedure	Preparation Time	Number of Preps
Salt Removal				
ReadyPrep 2-D cleanup kits (#1632130, #1632140)	Cleanup of protein samples for 1-D and 2-D applications	TCA-like precipitation to remove salts, detergents, lipids, phenolic compounds	<1 hr	50
	Reduction of streaking on 2-D gels Concentration of dilute samples			
Micro Bio-Spin <sup>™</sup> 6 columns (see pages 16–17)	Removal of salts and other contaminants	Size exclusion chromatography	5 min	25, 100
Removal of Other Contamina	ants			
ReadyPrep 2-D cleanup kits (#1632130, #1632140)	Reduction of streaking on 2-D gels Concentration of dilute samples	TCA-like precipitation to remove salts, detergents, lipids, phenolic compounds	<1 hr	50
Reduction and Alkylation				
ReadyPrep reduction- alkylation kit (#1632090)	Reduction of streaking on 2-D gels Improved resolution of basic proteins	Reduction, then alkylation of sample to remove disulfide bonds and prevent their re-formation	<2 hr	100

#### For More Information

Web: bio-rad.com/proteincleanup Request or download bulletins: 2934 and 2961

#### bio-rad.com/proteinsampleprep

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**Protein Fractionation** 

Ordering In	formation
Catalog #	Description
ReadyPrep	Kits
1632130	ReadyPrep 2-D Cleanup Kit, 50 preps
1632140	ReadyPrep 2-D Cleanup Kit, 5 preps
1632105	ReadyPrep 2-D Starter Kit, includes protein sample and reagents sufficient to rehydrate, focus, and transfer
	to second-dimension gels; six 17 cm, ten 11 cm, or sixteen 7 cm ReadyStrip IPG strips (ReadyStrip IPG
	strips*, precast SDS-PAGE gels, and gel stains not included in kit)
1632090	ReadyPrep Reduction-Alkylation Kit, 100 preps
ReadyPrep	Kit Components and Related Products
1632091	ReadyPrep Proteomics Grade Water, 500 ml
1632083	ReadyPrep 2-D Rehydration/Sample Buffer 1, 10 ml, 7 M urea, 2 M thiourea, 1% ASB-14, 40 mM Tris,
	0.001% bromophenol blue
1632106	ReadyPrep 2-D Starter Kit Rehydration/Sample Buffer, 10 ml, 8 M urea, 2% CHAPS, 50 mM DTT, 0.2%
	Bio-Lyte 3/10 ampholyte, 0.001% bromophenol blue
Reducing an	nd Alkylating Agents
1610611	Dithiothreitol (DTT), 5 g
1632101	Tributylphosphine (TBP), 200 mM, 0.6 ml
1632109	lodoacetamide, 30 g
* See the pre	packed Bio-Spin and Micro Bio-Spin gel filtration columns on pages 16–17.

# **Protein Fractionation**

Fractionation kits reduce sample complexity, helping to identify low-abundance proteins. Fractionation kits can be subdivided into three groups that fractionate based on differential solubility, cellular location of the proteins of interest, and protein charge.

# **Fractionation by Solubility**

#### **Fractionation Using Differential Protein Solubility**

The ReadyPrep sequential extraction kit and the ReadyPrep protein extraction kit (soluble/insoluble) both reduce sample complexity using differential solubilization. The two kits can be used independently, or the rehydration/sample buffer from the soluble/insoluble kit can be used with the sequential extraction kit to create a fourth fraction for even better resolution.

 ReadyPrep sequential extraction kit — enables the isolation of 3 different fractions of increasing solubility. These fractions are isolated sequentially, allowing the visualization of proteins that might not otherwise be seen. Increasing solubilization strength is provided through the use of stronger detergents for each subsequent fraction

 ReadyPrep protein extraction kit (soluble/insoluble) uses a single fractionation step

# ReadyPrep Kit Components and Related Products

Individual ReadyPrep kit components and related products such as reducing agents are also available. See ordering information on pages 4–5.

For More Information Web: bio-rad.com/proteinfractionation Request or download bulletins: 2934 and 2961

# Ordering Information

Catalog #	Description
ReadyPrep K	lits
1632100	ReadyPrep Sequential Extraction Kit, 15 preps
1632085	ReadyPrep Protein Extraction Kit (Soluble/Insoluble), 20 preps
ReadyPrep K	Cit Components and Related Products
1632102	ReadyPrep Sequential Extraction Kit Reagent 1, 50 ml, 40 mM Tris base
1632103	ReadyPrep Sequential Extraction Kit Reagent 2, 10 ml, 8 M urea, 4% CHAPS, 40 mM Tris base, 0.2% Bio-Lyte 3/10 ampholyte
1632104	ReadyPrep Sequential Extraction Kit Reagent 3, 10 ml, 5 M urea, 2 M thiourea, 2% CHAPS, 2% SB 3–10, 40 mM Tris base, 0.2% Bio-Lyte 3/10 ampholyte
1632083	ReadyPrep 2-D Rehydration/Sample Buffer 1

See Also Bio-Spin gel filtration columns: page 16. Micro Bio-Spin gel filtration columns: page 16.

# **Fractionation by Cellular Location**

### **Fractionation by Cellular Location**

ReadyPrep<sup>™</sup> protein extraction kits facilitate fractionation of proteins from different cellular locations such as the membrane, nucleus, or cytoplasm.

- ReadyPrep protein extraction kit (cytoplasmic/ nuclear) — prepares fractions enriched in cytoplasmic or nuclear proteins from eukaryotic samples
- ReadyPrep protein extraction kit (membrane I) offers a quick and effective protocol for isolating most membrane proteins. It does not require ultracentrifugation or preparation of density gradients
- ReadyPrep protein extraction kit (membrane II) offers a protocol for isolating more complex membrane proteins
- ReadyPrep protein extraction kit (signal) for isolating proteins involved in intracellular membrane trafficking and signaling pathways. These include GPIanchored proteins, caveolin and associated proteins, acylated tyrosine kinases, and G proteins

# For More Information

Web: bio-rad.com/proteinfractionation Request or download bulletins: 2934 and 2961

Ordering Information			
Catalog #	Description		
ReadyPrep I	Kits		
1632089	ReadyPrep Protein Extraction Kit (Cytoplasmic/Nuclear), 50 preps		
1632088	ReadyPrep Protein Extraction Kit (Membrane I), 50 preps		
1632084	ReadyPrep Protein Extraction Kit (Membrane II), 10 preps		
1632087	ReadyPrep Protein Extraction Kit (Signal), 50 preps		
Related Pro	ducts		
1632083	ReadyPrep 2-D Rehydration/Sample Buffer 1		
1632085	ReadyPrep Protein Extraction Kit (Soluble/Insoluble), 20 preps		

### **Fractionation by Charge**

Aurum<sup>™</sup> ion exchange (AEX or CEX) mini columns allow selective purification of acidic or basic proteins, respectively. Also available in easy-to-use kits, these mini columns selectively enrich either acidic or basic proteins and can be used with a variety of starting samples.

#### For More Information

Web: bio-rad.com/proteinfractionation Request or download bulletin: 2928



#### **Ordering Information**

Cata	llog #	Description
7326	6703	Aurum CEX Mini Columns, 25 pack
7326	6706	Aurum AEX Mini Columns, 25 pack

#### bio-rad.com/proteinsampleprep

# **Protein Depletion**

Complex samples often require depletion of high-abundance proteins to allow the detection of the low-abundance ones. Bio-Rad offers two different methodologies for protein depletion — the ProteoMiner<sup>™</sup> protein enrichment system (which utilizes hexapeptide libraries) and Aurum<sup>™</sup> serum and Affi-Gel<sup>®</sup> Blue products (which utilize resins).

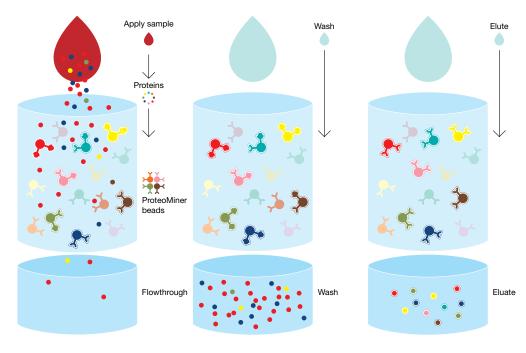
### **ProteoMiner<sup>™</sup> Protein Enrichment System**

The ProteoMiner protein enrichment system is a novel sample preparation tool for reducing the dynamic range of protein concentrations in complex biological samples. The ProteoMiner system:

- Enriches and concentrates low-abundance proteins that cannot be detected through traditional methods
- Works with a variety of sample types (serum, plasma, urine, bile, cell lysates, tissues, and platelets) and is not limited by the species' origin
- Utilizes a combinatorial library of hexapeptides rather than immunodepletion, minimizing dependence on available antibodies and preventing the codepletion of low-abundance proteins

**ProteoMiner Protein Enrichment Kits** — these kits can be used with a variety of biological samples and are compatible with all major downstream proteomics applications. Small- and large-capacity kits are available for processing two or ten samples.

**ProteoMiner Sequential Elution Kits** — these kits utilize multiple elution reagents to sequentially elute proteins based on different properties.



ProteoMiner technology is based on the interaction of complex protein samples with a large, highly diverse library of hexapeptides bound to chromatographic supports. In theory, each unique hexapeptide binds to a unique protein sequence. Because the bead capacity limits binding capacity, high-abundance proteins quickly saturate their ligands (red and yellow beads) and excess protein is washed out during the procedure. In contrast, low-abundance proteins are concentrated on their specific ligands (pink and teal beads), thereby decreasing the dynamic range of proteins in the sample. When analyzed in downstream applications, the number of proteins detected is dramatically increased.

# **Protein Depletion**

#### bio-rad.com/proteinsampleprep

#### ProteoMiner Small- and Large-Capacity Kits

ProteoMiner kits for protein enrichment are now offered in formats optimized for varying starting amounts of sample protein.

- Small-capacity kits optimized for use with limited sample material (minimum 10 mg of protein is recommended)
- Large-capacity kits optimized for use with samples in which at least 50 mg of protein is available

For More Information Web: bio-rad.com/proteominer Request or download bulletins: 5632, 5635, and 5841

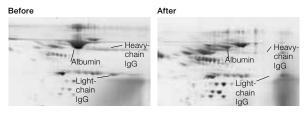
Catalog #	Description
1633006	ProteoMiner Protein Enrichment Small-Capacity Kit, 10 preps, for processing 10 mg of total protein, includes 10 spin columns, wash buffer, elution reagents, collection tubes
1633007	ProteoMiner Protein Enrichment Large-Capacity Kit, 10 preps, for processing 50 mg of total protein, includes 10 spin columns, wash buffer, elution reagents, collection tubes
1633008	ProteoMiner Protein Enrichment Introductory Small-Capacity Kit, 2 preps, for processing 10 mg of total protein, includes 2 spin columns, wash buffer, elution reagents, collection tubes
1633009	ProteoMiner Protein Enrichment Introductory Large-Capacity Kit, 2 preps, for processing 50 mg of total protein, includes 2 spin columns, wash buffer, elution reagents, collection tubes
ProteoMine	r Sequential Elution Kits
1633010	ProteoMiner Sequential Elution Small-Capacity Kit, 10 preps, for processing 10 mg of total protein, includes 10 spin columns, wash buffer, 4 sequential elution reagents, collection tubes
1633011	<b>ProteoMiner Sequential Elution Large-Capacity Kit</b> , 10 preps, for processing 50 mg of total protein, includes 10 spin columns, wash buffer, 4 sequential elution reagents, collection tubes
ProteoMine	r Kit Accessories
1633003	ProteoMiner Sequential Elution Reagents, 10 preps, includes reagents only (columns not included), to be used with #1633006 or #1633007

# Aurum<sup>™</sup> Kits and Columns

The Aurum<sup>™</sup> Affi-Gel<sup>®</sup> Blue and Aurum serum kits and columns use affinity chromatography to reduce albumin and IgG, which improves analysis of lower abundance proteins. These products utilize a quick and easy spin-column format and provide eluted proteins ready for analysis.

#### For More Information Web: bio-rad.com/proteindepletion

Request or download bulletin: 2823



Removal of albumin and IgG from serum using the Aurum serum protein mini kit. Total protein (1.32 mg) was purified on an Aurum serum protein mini column. Left, untreated serum; right, serum treated with the Aurum kit.

Ordering Inf	ormation
Catalog #	Description
7326701 7326708	Aurum Serum Protein Mini Kit, 10 pack, includes columns and buffers Aurum Affi-Gel Blue Mini Columns, 25 pack

# **Nucleic Acid Sample Preparation**

# **RNA** Isolation

Bio-Rad offers a broad range of sample preparation products for RNA isolation. Scalable and high-throughput products are available.

#### **RNA Isolation Product Selection Guide**

	Aurum <sup>™</sup> Total RNA Kits			PureZOL <sup>™</sup> RNA
	Mini Kit	Fatty and Fibrous Tissue Kit	96 Kit	Isolation Reagent
Format	Mini column filtration (vacuum or spin)	Mini column filtration (vacuum or spin)	96-well plate filtration (vacuum or spin)	Single solution organic extraction
Maximum starting material amounts				
Cultured cells	2 x 10 <sup>6</sup>	1 x 10 <sup>7</sup>	1 x 10 <sup>6</sup>	1 x 10 <sup>7</sup>
Bacterial cells	2.4 x 10 <sup>9</sup>	2.4 x 10 <sup>9</sup>	8 x 10 <sup>8</sup>	2.4 x 10 <sup>9</sup>
Yeast cells Hard animal tissue	3 x 10 <sup>7</sup> 20 mg	3 x 10 <sup>7</sup> 100 mg	2 x 10 <sup>7</sup>	3 x 10 <sup>7</sup>
Soft to moderately hard animal tissue	40 mg	100 mg	_	100 mg 100 mg
Plant tissue	40 mg	100 mg	_	100 mg
Isolation method	Silica membrane	Lysis with PureZOL reagent, purification on silica membrane	Silica membrane	Organic extraction
Number of preps	50	50	192 (2 x 96-well plate)	50 or 100 (1 ml/prep)
Number of washes	3	3	3	-
DNase I included*	Yes	Yes	Yes	No
DNase I digest time	15 min (animal tissue, 25 min)	15 min	10 min	-
Total preparation time**	<50–80 min (with DNase I digest)	<50–80 min (with DNase I digest)	<60 min (with DNase I digest)	<60 min
Binding capacity	>100 µg	>100 µg	>40 µg	-
Maximum elution volume	80 µl (2 x 40 µl elutions)	2 x 40 µl	80 µl	30–100 µl
Yield***	20–30 µg/2 x 10 <sup>6</sup> NIH 3T3 cells 20–40 µg/2 x 10 <sup>6</sup> HeLa cells 15–20 µg/40 mg brain 30–55 µg/20 mg thymus 15–30 µg/40 mg spleen 8–15 µg/40 mg liver 15–20 µg/40 mg kidney 8–15 µg/40 mg potato 20–30 µg/3 x 10 <sup>7</sup> <i>S. cerevisiae</i> cells 35–55 µg/2.4 x 10 <sup>9</sup> <i>E. coli</i> cells	140–150 μg/1 x 10 <sup>7</sup> 293H cells 90–100 μg/100 mg brain 50–60 μg/100 mg breast 80–90 μg/100 mg heart 60–70 μg/100 mg skin 50–60 μg/100 mg cartilage 20–30 μg/2.4 x 10 <sup>9</sup> <i>E. coli</i> cells 60–65 μg/3 x 10 <sup>7</sup> <i>S. cerevisiae</i> cells 90–100 μg/100 mg potato 5–10 μg/100 mg <i>Arabidopsis</i> 10–15 μg/50 mg <i>A. niger</i>	10 μg/1 x 10 <sup>6</sup> NIH 3T3 cells 17 μg/1 x 10 <sup>6</sup> HeLa cells 11 μg/2 x 10 <sup>7</sup> <i>S. cerevisiae</i> cells 5 μg/8 x 10 <sup>8</sup> <i>E. coli</i> cells 5 μg/8 x 10 <sup>8</sup> <i>B. cereus</i> cells	1.37 $\mu$ g/1 x 10 <sup>5</sup> 293H cells 1.15 $\mu$ g/mg brain 1.37 $\mu$ g/mg fat 1.2 $\mu$ g/mg heart 0.66 $\mu$ g/mg cartilage 1.2 $\mu$ g/mg skin 1.0 $\mu$ g/mg potato 0.1 $\mu$ g/mg <i>Arabidopsis</i> 0.38 $\mu$ g/mg <i>A. niger</i> 0.63 $\mu$ g/2.4 x 10 <sup>5</sup> <i>E. coli</i> cells 0.31 $\mu$ g/3 x 10 <sup>5</sup>
Purity	A <sub>260</sub> /A <sub>280</sub> of 1.9–2.1	A <sub>260</sub> /A <sub>280</sub> of 1.9–2.1	A <sub>260</sub> /A <sub>280</sub> of 1.9–2.1	A <sub>260</sub> /A <sub>280</sub> of 1.9–2.1

\* Removal not required.

\*\* Total preparation time will vary depending on the tissue or cell type and on which format is used (vacuum or spin).

\*\*\* Yields will vary depending on the developmental stage, tissue type or cell line, and growth conditions used.

#### electrophoresis system: pages 267–272.

See Also PCR reagents: pages 372–385. Real-time PCR systems: pages 361–366. Experion automated

DNA amplification/PCR pages 349–402.

# **Nucleic Acid Sample Preparation**

**RNA** Isolation

### See Also

PureZOL reagent: page 11. Lipid transfection reagents: pages 337–338. DNA amplification/PCR: pages 349–402.

### Aurum<sup>™</sup> Total RNA Kits

Aurum total RNA kits are designed and formulated to assist in the isolation of highly pure and intact RNA from a variety of starting materials. Isolated RNA is compatible with downstream applications including real-time qPCR, northern blotting, microarray analysis, and cDNA library construction.

- Isolate high yields of RNA from a wide range of sample types in less than 60 min
- DNase treatment ensures genomic DNA removal
- Produce ready-to-use RNA suitable for the most demanding downstream applications

Aurum total RNA kits offer streamlined processing using a choice of vacuum- or spin-mediated protocols. If vacuum-mediated purification is preferred, the protocol may be performed with a vacuum manifold.\*

#### For More Information

Web: bio-rad.com/rna-isolation Request or download bulletins: 2919, 2920, and 5282

#### Aurum Total RNA Mini Kit

The Aurum total RNA mini kit produces DNA-free total RNA from a wide range of starting materials, including cultured cells, bacteria, and yeast, as well as animal and plant tissues. The kit utilizes a stringent reagent composed of guanidine isothiocyanate and  $\beta$ -mercaptoethanol for efficient sample lysis and quick RNase inactivation, followed by purification on silica membrane in a spin-column format using a spin- or vacuum-mediated protocol. The kit can also be used for RNA cleanup and desalting.

#### Aurum Total RNA Fatty and Fibrous Tissue Kit

The Aurum total RNA fatty and fibrous tissue kit isolates total RNA from samples that are difficult to disrupt. The kit is effective for purifications involving fatty and fibrous tissues or samples, such as fungi, that are rich in RNases. The kit utilizes PureZOL<sup>™</sup> RNA reagent, a potent phenol-based reagent, which effectively lyses tissues and cells. The kit includes spin columns that can be processed using a choice of vacuum- or spin-mediated protocols.

\* The Aurum vacuum manifold is no longer available. Please contact Bio-Rad Technical Support for information on alternative products.



Aurum Total RNA Fatty and Fibrous Tissue Kit

### Aurum Total RNA 96 Kit

The Aurum total RNA 96 kit reproducibly isolates DNA-free total RNA from cultured cells, bacteria, and yeast in under 60 minutes in a 96-well format for high-throughput total RNA isolation. The total RNA binding plate is designed for use on the Aurum vacuum manifold.\*

#### **Consumables and Accessories**

The reagents and plasticware used in the Aurum RNA kits are available as refill orders. These products can be ordered separately from the Aurum total RNA kits.

#### bio-rad.com/nasampleprep

**RNA** Isolation

Catalog #	Description
7326820	Aurum Total RNA Mini Kit, 50 preps, includes 50 RNA binding columns, 50 capless collection tubes (2.0 ml), 100 capped sample tubes (2.0 ml), 50 capped sample tubes (1.5 ml), 1 vial lyophilized DNase I, RNase-free reagents
7326830	Aurum Total RNA Fatty and Fibrous Tissue Kit, 50 preps, includes #7326870, 50 ml PureZOL RNA isolation reagent
7326870*	Aurum Total RNA Fatty and Fibrous Tissue Module, 50 preps, includes 50 RNA binding columns, 50 capless collection tubes (2.0 ml), 100 capped sample tubes (2.0 ml), 50 capped sample tubes (1.5 ml), 1 vial lyophilized DNase I, RNase-free reagents, and plasticware
7326800	Aurum Total RNA 96 Kit, 2 x 96-well preps, includes 2 grow blocks, sealing tape, 2 RNA binding plates, 2 collection microplates, 2 vials lyophilized DNase I, RNase-free reagents
Aurum Cons	umables and Accessories
7326828	DNase I, RNase-free, lyophilized, 1 vial
7326801	Aurum Total RNA Elution Solution, RNase-free, 20 ml
7326802	Aurum Total RNA Lysis Solution, RNase-free, 85 ml
7326803	Aurum Total RNA Wash High-Stringency Solution, RNase-free, 150 ml
7326804	Aurum Total RNA Wash Low-Stringency Solution, 60 ml
7326805	Aurum DNase Dilution Solution, 20 ml
7326826	Aurum RNA Binding Mini Columns, 50

\* Does not include PureZOL RNA isolation reagent (see catalog #7326880 or #7326890 to order separately).

# PureZOL<sup>™</sup> RNA Isolation Reagent

The PureZOL RNA isolation reagent protocol is an improvement over the rapid, widely used, and proven method of RNA isolation developed by Chomczynski and Sacchi (1987). PureZOL RNA isolation reagent is a potent monophasic combination of phenol and the chaotropic agent guanidine isothiocyanate, which effectively lyses cells and tissues, deproteinates RNA, and inactivates endogenous nucleases in a single step. DNA and protein are efficiently removed from the RNA following phase separation.

The ready-to-use PureZOL RNA isolation reagent is a versatile and efficient means of isolating high yields of RNA from a variety of sources including cultured cells, animal and plant tissue, yeast, virus, and bacteria samples. The single-solution format permits recovery of RNA from small quantities of tissues or cells, making it ideally suited for gene expression analysis or whenever sample quantities are limited. Total RNA isolated using PureZOL RNA isolation reagent is free of



DNA and protein and can be used for northern blot analysis, in vitro translation, poly(A)+ selection, RNase protection assays, RT-PCR, and molecular cloning. Since no spin columns are used in the protocol, the protocol is scalable to accommodate a wide range of sample sizes.

For More Information Web: bio-rad.com/purezol

#### **Ordering Information**

Catalog #	Description
7326880	PureZOL RNA Isolation Reagent, 50 ml
7326890	PureZOL RNA Isolation Reagent, 100 ml

#### See Also

Aurum total RNA kits: pages 10–11.

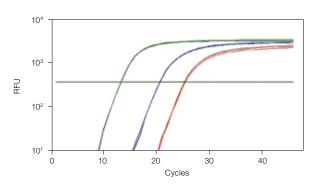
# iScript<sup>™</sup> RT-qPCR Sample Preparation Reagent

iScript RT-qPCR sample preparation reagent enables efficient cell lysis and RNA stabilization for sensitive quantitative gene expression analysis without RNA purification. This unique reagent accelerates and streamlines RT-qPCR analysis of cultured cells by eliminating the need to purify RNA. Reverse transcription and real-time PCR (qPCR) can be performed directly from cell lysates. This reagent is ideal for rapid, high-throughput gene expression analysis such as validation of siRNA-mediated gene knockdown.

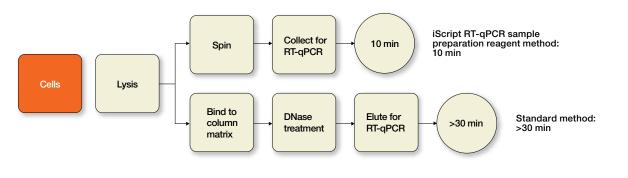
- Rapid protocol (5–10 min) efficiently stabilizes RNA
- Sensitive detection of high-, medium-, and low-copy number gene targets directly from cell lysates
- Reagent enables multiplex qPCR detection of up to 4 targets from as few as 10 cells

#### For More Information Web: bio-rad.com/iscript-sampleprep

Request or download bulletin: 5736



iScript RT-qPCR sample preparation reagent provides rapid yet sensitive gene expression results for high-, medium-, and low-copy number gene targets. HeLa cells were treated with iScript RT-qPCR sample preparation reagent at 125 cells/µl and the expression levels of three different genes were assessed. 18S rRNA (-), β-tubulin (-), and CMYC (-) expression levels were determined by performing reverse transcription (iScript cDNA synthesis kit) and qPCR (iQ<sup>™</sup> SYBR<sup>®</sup> Green supermix) directly from cell lysate preparations. RFU, relative fluorescence units.



Ordering Inf	formation
Catalog #	Description
1708898	iScript RT-qPCR Sample Preparation Reagent, 100 reactions, 10 ml, contains RNase inhibitors and RNA stabilizers
1708899	iScript RT-qPCR Sample Preparation Reagent, 500 reactions, 5 x 10 ml, contains RNase inhibitors and RNA stabilizers

# **DNA** Isolation

### **DNA Isolation Kit Selection Guide**

Kit		DNA Yield	Preparation Time	Bacterial Culture Volume	Purification Format	Binding Matrix	Growth Media
Quantum Prep <sup>®</sup> plasmid miniprep	I	≤40 µg	<15 min	1–10 ml	Spin	Diatomaceous earth	Enriched or standard
Quantum Prep plasmid midiprep	Ţ	≤400 µg	45 min	20–40 ml	Spin	Diatomaceous earth	Enriched or standard
Aurum <sup>™</sup> plasmid mini	8	≤20 µg	<10 min	1–5 ml	Vacuum* and spin	Silica membrane	Standard

\* Using the Aurum vacuum manifold. This product is no longer available. Please contact Bio-Rad Technical Support for information on alternative products.

# **Quantum Prep® Plasmid Kits**

#### **Quantum Prep Plasmid Miniprep Kit**

The Quantum Prep miniprep kit offers high quality and yield in an easy spin-based procedure that takes less than 15 minutes from cell culture to purified plasmid. Plasmid DNA is recovered in water or TE for immediate use in all downstream molecular biology applications.

### **Quantum Prep Plasmid Midiprep Kit**

The Quantum Prep plasmid midiprep kit uses a simple spin-column procedure that significantly reduces the time required to generate the large quantities of plasmid DNA needed to support applications such as transfection, subcloning, and other enzymatic manipulations.

#### For More Information Web: bio-rad.com/quantum-prep

Request or download bulletin: 2325



#### **Ordering Information**

Catalog #	Description
7326100	Quantum Prep Plasmid Miniprep Kit, 100 preps, includes 25 ml cell resuspension solution, 25 ml cell lysis
	solution, 25 ml neutralization solution, 20 ml Quantum Prep matrix, 63 ml wash solution, 100 spin columns
7326120	Quantum Prep Plasmid Midiprep Kit, 20 preps, includes 110 ml cell resuspension solution, 110 ml cell lysis
	solution, 110 ml neutralization solution, 20 ml Quantum Prep matrix, 125 ml wash solution, 20 midi spin columns

**DNA** Isolation

#### See Also

Lipid transfection reagents: pages 337–338. DNA amplification/PCR: pages 349–402.

# Aurum<sup>™</sup> Plasmid Mini Kit

The easy-to-use Aurum plasmid mini kit improves the efficiency and throughput of plasmid purifications with a simple bind-wash-elute protocol using silica membranes, all in less than 10 minutes. Spin- and vacuum-mediated protocols are available. The purified plasmid DNA can be immediately used in any downstream molecular biology application.

The Aurum plasmid mini kit delivers high yields of reproducible plasmid DNA preparations for:

- Automated fluorescence-based sequencing
- Restriction digestion
- Ligation and transformation
- Transfection
- PCR

# For More Information

Web: bio-rad.com/aurum-plasmid Request or download bulletin: 2664



Ordering Information						
Catalog #	Description					
7326400	Aurum Plasmid Mini Kit, 100 preps, includes plasmid-binding mini columns, 100 capless collection tubes, reagents, protocol overview					

# See Also

Molecular biology and biotechnology grade resins: page 64.

# InstaGene<sup>™</sup> Matrix

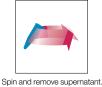
InstaGene matrix is designed for rapid isolation of small amounts of genomic DNA of sufficient purity for PCR in under an hour. The specially formulated 6% w/v Chelex<sup>®</sup> resin adsorbs cell lysis products that interfere with PCR, leaving genomic DNA template in the supernatant where it is immediately available for PCR reactions.

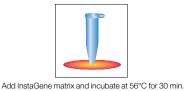
#### **For More Information**

Web: bio-rad.com/instagene-matrix Request or download bulletin: 2074

#### InstaGene Protocol







Vortex and incubate at 100°C for 8 min.



Spin; use supernatant for PCR.

#### 

Ordering Info	prmation
Catalog #	Description
7326030	InstaGene Matrix, 20 ml, sufficient for 100 extractions

# **Nucleic Acid Sample Preparation**

**DNA Cleanup** 

#### bio-rad.com/nasampleprep

### Chelex<sup>®</sup> 100 Molecular Biology Grade Resin

Nuclease and ligase inhibitor-free, this pipettable, smallscale resin is certified not to inhibit PCR and ensures complete removal of PCR inhibitors and metal ions.

For More Information Web: bio-rad.com/dna-isolation Request or download bulletin: 2074

#### See Also

Molecular biology and biotechnology grade resins: page 64.

#### **Ordering Information** Catalog # Description

1421253 Chelex 100 Resin, molecular biology grade, 50 g, sodium, 200-400 dry mesh, 75-150 µm wet bead

**DNA** Cleanup

Bio-Rad offers a variety of sample preparation and cleanup columns; many are also suitable for DNA fragment purification.

2 3 4 5

# Freeze 'N Squeeze<sup>™</sup> DNA Gel Extraction Spin Columns

Freeze 'N Squeeze spin columns offer a filtration-based purification method, which provides a quick and effective alternative to chemical extraction and electroelution methods. The Freeze 'N Squeeze method uses centrifugation to draw 50-23,000 bp DNA out of agarose gel slices that have been guickly frozen and thawed. Features include:

- No solutions to prepare save time and avoid using toxic chaotropic materials
- DNA immediately available for PCR, subcloning, ligations, and sequencing reactions
- Less than 1 min of hands-on time

# For More Information

Web: bio-rad.com/freeze-n-squeeze

#### Freeze 'N Squeeze Method



Cut out gel slice

**Ordering Information** 

Catalog # 7326165

7326166



Description



Chop into pieces.



Freeze 'N Squeeze DNA Gel Extraction Spin Columns, 25 columns

Freeze 'N Squeeze DNA Gel Extraction Spin Columns, 100 columns

Transfer to spin column and freeze for 5 min.

-20°C for 5 min



6

1,000 bp

200

100

Spin for 3 min.



Recovery of DNA

Freeze 'N Squeeze spin columns.

Extracted fragments

gel. Lane 1, precision

molecular mass ruler;

PCR molecular ruler.

lane 7, EZ Load<sup>™</sup> 100 bp

were rerun on a ReadyAgarose<sup>™</sup> 1% TAE

fragments using



# See Also

Agarose gel system: pages 262-263.

page 252. DNA ladders:

# **Prepacked Spin Columns**

Prepacked size exclusion spin columns allow easy cleanup and purification of DNA and proteins from lower MW contaminants. Bio-Spin<sup>®</sup>, Micro Bio-Spin<sup>™</sup>, and PCR Kleen<sup>™</sup> columns clean up DNA or protein samples quickly and easily using size exclusion chromatography. These columns are available in multiple sizes and offer multiple MW exclusion limits to accommodate a variety of needs. Use the chart below to choose the column that best meets your needs.

#### Prepacked Spin Column Selection Guide

	Bio-Spin 6	Micro Bio-Spin 6	Bio-Spin 30	Micro Bio-Spin 30	PCR Kleen
Packed support	Special grade Bio-Gel <sup>®</sup> P-6 gel	Special grade Bio-Gel P-6 gel	Special grade Bio-Gel P-30 gel	Special grade Bio-Gel P-30 gel	Special grade size exclusion gel
Equilibration buffer	10 mM Tris, pH 7.4, or SSC buffer*	10 mM Tris, pH 7.4, or SSC buffer*	10 mM Tris, pH 7.4, or SSC buffer*	10 mM Tris, pH 7.4, or SSC buffer*	10 mM Tris, 1 mM EDTA, pH 7.0
Applications Desalting of oligonucleotides >20 bases	•	•	-	-	-
Labeling reactions: removal of unincorporated nucleotides >20 bases or bp from DNA	_	_	•	•	_
Removal of primers and primer-dimers from PCR products >200 bp	_	-	_	-	•
Buffer exchange (restriction fragments, PCR products, enzyme reactions, sequencing templates)	·	•	_	•	_
DNA sequencing reaction mixture cleanup**	-	-	•	•	-
Riboprobe cleanup***	-	-	-	•	-
Desalting of antibody, enzyme, and protein solutions	•	•	_	•	-
Purification of proteins of molecular weight >6,000	•	•	_	_	_
Purification of proteins of molecular weight >40,000	_	-	•	•	-
Bed volume	1.1 ml	0.7 ml	1.1 ml	0.7 ml	0.6 ml
Retention and recovery	90% recovery of 20 bases or bp, 99% retention of salts	90% recovery of 20 bases or bp, 99% retention of salts	95% recovery of 22 bases or bp, 98% retention of ddNTPs	95% recovery of 22 bases or bp, 98% retention of ddNTPs	85% recovery of ≥700 bp, 95% retention of primers and primer-dimers
Molecular weight exclusion limit, globular proteins	6,000	6,000	40,000	40,000	8,000,000
Sample volume	50–100 µl	10–75 µl	50–100 µl	10–75 µl	25–100 µl
Centrifuge type	Swinging bucket	Microcentrifuge	Swinging bucket	Microcentrifuge	Microcentrifuge
Autoclavability	Yes	Yes	Yes	Yes	Yes

\* 150 mM NaCl, 17.5 mM sodium citrate, pH 7.0.

\*\* In Tris buffer.

\*\*\* In RNase-free Tris buffer.

#### bio-rad.com/nasampleprep

# **Nucleic Acid Sample Preparation**

**DNA Cleanup** 

See Also

Bio-Spin,

page 103.

Mini Bio-Spin

empty columns:

Empty columns:

pages 103-110.

Bio-Gel P resin: page 85.

Micro Bio-Spin, and

#### Prepacked Bio-Spin and Micro Bio-Spin Columns

Bio-Spin and Micro Bio-Spin columns clean up and remove salts, nucleotides, dye terminators, and small molecules from DNA, RNA, and protein samples in 10 minutes. Filled with specially sized Bio-Gel<sup>®</sup> P gels, these columns are shipped fully hydrated in Tris or SSC buffer. They yield 95% recovery of DNA >22 bp and allow sample loads of 10–100 µl. For safe riboprobe preparation use RNase-free Micro Bio-Spin P-30 Tris spin columns.

#### For More Information

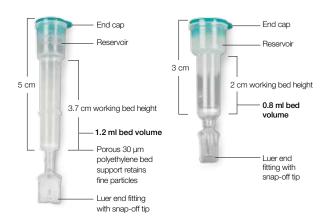
Web: bio-rad.com/dna-cleanup and /protein-cleanup

#### PCR Kleen Spin Columns

PCR Kleen columns are prepacked spin columns for purifying PCR products and other DNA molecules >200 bp directly from reaction mixtures. A simple 4-minute spin effectively removes salts, nucleotides, enzymes, primers, and primer-dimers. Purified DNA fragments are immediately available for secondary PCR, subcloning, restriction digests, ligations, sequencing, and other enzymatic manipulations.

#### For More Information

Web: bio-rad.com/dna-cleanup Request or download bulletin: 2311



Prepacked Bio-Spin and Micro Bio-Spin Columns



PCR Kleen Spin Columns

Catalog #	Description	
Micro Bio-S	pin Columns with Bio-Gel P-6 in Tris Buffer	
7326221	Micro Bio-Spin 6 Columns, includes 25 columns in Tris buffer, 50 collection tubes	
7326222	Micro Bio-Spin 6 Columns, includes 100 columns in Tris buffer, 200 collection tubes	
Micro Bio-S	pin Columns with Bio-Gel P-30 in Tris Buffer	
7326223	Micro Bio-Spin 30 Columns, includes 25 columns in Tris buffer, 50 collection tubes	
7326224	Micro Bio-Spin 30 Columns, includes 100 columns in Tris buffer, 200 collection tubes	
7326250	Micro Bio-Spin 30 Columns, includes 25 columns in Tris buffer, 50 collection tubes, RNase-free	
7326251	Micro Bio-Spin 30 Columns, includes 100 columns in Tris buffer, 200 collection tubes, RNase-free	
Micro Bio-S	pin Columns with Bio-Gel P-6 in SSC Buffer	
7326200	Micro Bio-Spin 6 Columns, includes 25 columns in SSC buffer, 50 collection tubes	
7326201	Micro Bio-Spin 6 Columns, includes 100 columns in SSC buffer, 200 collection tubes	
Micro Bio-S	pin Columns with Bio-Gel P-30 in SSC Buffer	
7326202	Micro Bio-Spin 30 Columns, includes 25 columns in SSC buffer, 50 collection tubes	
7326203	Micro Bio-Spin 30 Columns, includes 100 columns in SSC buffer, 200 collection tubes	
Bio-Spin Co	lumns with Bio-Gel P-6 in Tris Buffer	
7326227	Bio-Spin 6 Columns, includes 25 columns in Tris buffer, 50 collection tubes	
7326228	Bio-Spin 6 Columns, includes 100 columns in Tris buffer, 200 collection tubes	
Bio-Spin Co	lumns with Bio-Gel P-30 in Tris Buffer	
7326231	Bio-Spin 30 Columns, includes 25 columns in Tris buffer, 50 collection tubes	
7326232	Bio-Spin 30 Columns, includes 100 columns in Tris buffer, 200 collection tubes	
Bio-Spin Co	lumns with Bio-Gel P-6 in SSC Buffer	
7326002	Bio-Spin 6 Columns, includes 25 columns in SSC buffer, 50 collection tubes	
Bio-Spin Co	lumns with Bio-Gel P-30 in SSC Buffer	
7326006	Bio-Spin 30 Columns, includes 25 columns in SSC buffer, 50 collection tubes	
PCR Kleen S	Spin Columns	
7326300	PCR Kleen Spin Columns, 25	

# Sample Preparation Subject Index

# Subject Index

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