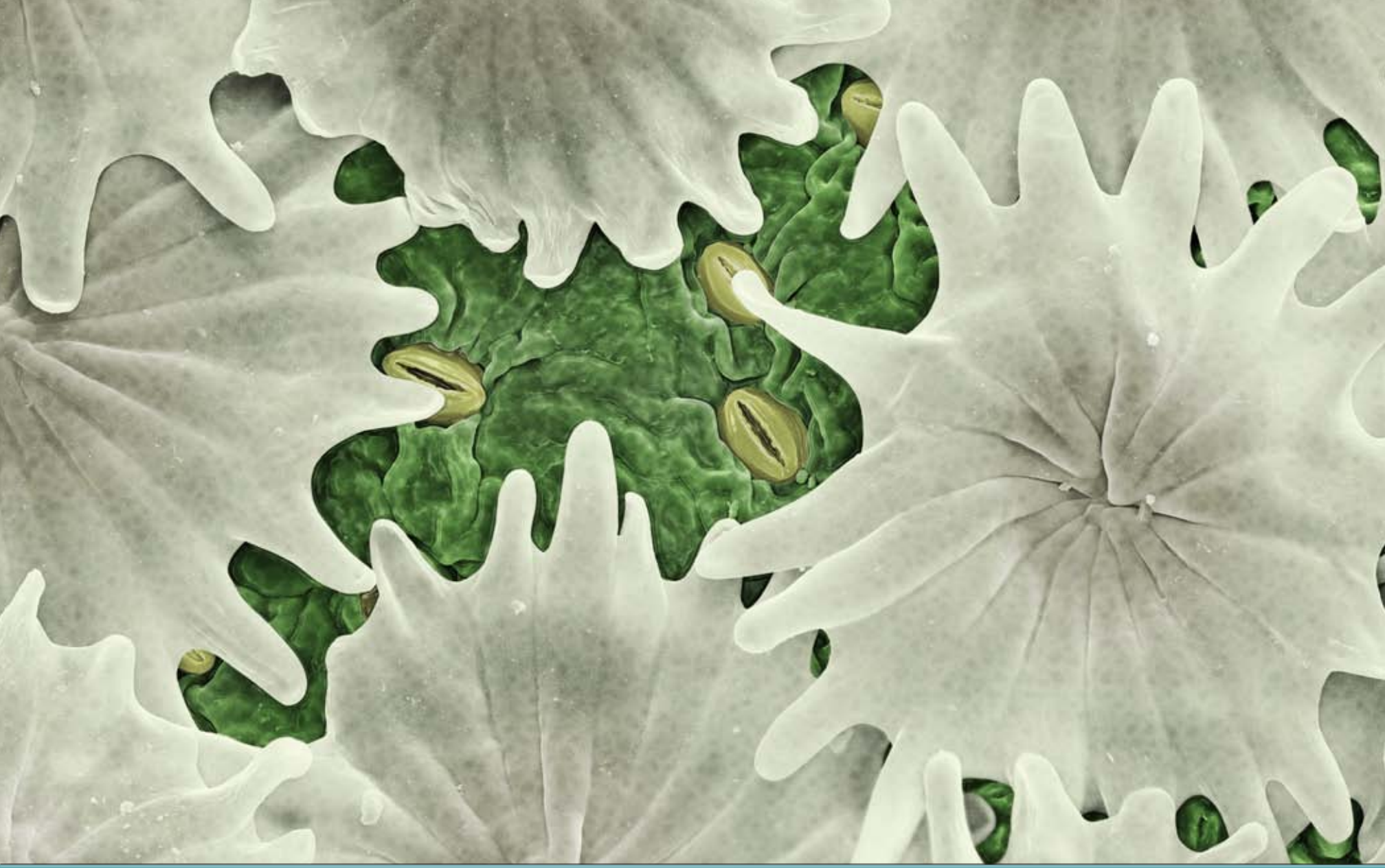

Life Science CATALOG 2011

Promega

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EXTRACTION

Promega

Maxwell® 16 System

Brings Personal Automation™ to your lab.

Compact, bench-top instrument for automated extraction of DNA, RNA or total nucleic acid for research, clinical and forensic applications. Convenient pre-filled reagent cartridges. Sample tracking system. Complete service and support. Affordable

See pages 84–88 and 198–202 for more information or visit:

www.promega.com/maxwell16

Promega

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GoTaq® Hot Start Polymerase

Product	Size	Cat.#
GoTaq® Hot Start Polymerase	100 u	M5001
	500 u	M5005
	2,500 u	M5006
	10,000 u	M5008
GoTaq® Hot Start Green Master Mix	100 reactions	M5122
	1,000 reactions	M5123
GoTaq® Hot Start Colorless Master Mix	100 reactions	M5132
	1,000 reactions	M5133

For Research Use Only. Not for use in diagnostic procedures.

Description: GoTaq® Hot Start Polymerase contains the high-performance GoTaq® DNA Polymerase bound to a proprietary antibody that blocks polymerase activity. The polymerase activity is restored during the initial denaturation step when the amplification reactions are heated at 94–95°C for two minutes. This allows for hot-start PCR, where polymerase activity is eliminated or minimized at temperatures below 70°C. GoTaq® Hot Start Polymerase exhibits 5′→3′ exonuclease activity. The system is supplied with a tube of 25mM $MgCl_2$, allowing optimization of the magnesium concentration in your reactions. It is also supplied with 5X Green GoTaq® Flexi Buffer and 5X Colorless GoTaq® Flexi Buffer. The buffers contain a compound that increases sample density, so that samples sink easily into the wells of an agarose gel. The green buffer also contains two dyes (yellow and blue) that separate to allow easy monitoring during electrophoresis. Use the green reaction buffer for direct-to-gel analysis after amplification and the colorless reaction buffer for amplifications where the dyes may interfere with post-amplification analysis such as fluorescence or absorbance testing.

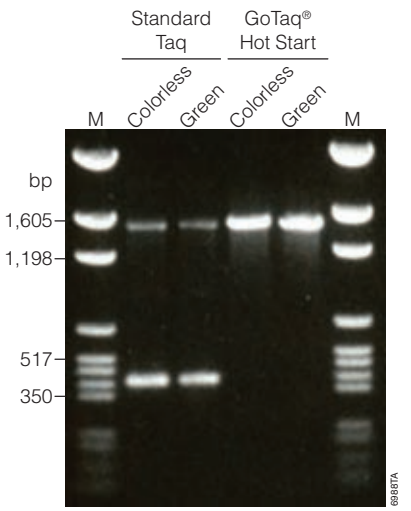
GoTaq® Hot Start Master Mixes are premixed, ready-to-use solutions containing GoTaq® Hot Start Polymerase, magnesium, dNTPs and buffer. Reactions can be set up in less than a minute at room temperature; simply add your template, water and primers. Available with either green or colorless reaction buffers, which also serve as loading buffers, allowing you to go directly from thermal cycler to gel analysis. GoTaq® Hot Start Master Mixes offer the specificity and sensitivity of an antibody-based hot-start polymerase in a convenient, easy-to-use, time-saving format.

Features:

- **Enhance Yield, Sensitivity and Specificity:** The proven, robust amplification and sensitivity of GoTaq® DNA Polymerase now with a built-in hot start to deliver even more superior results.
- **Ease of Use:** Set up your reaction at room temperature—no need to set up on ice.
- **Higher Yield:** Two-minute activation saves time and ensures maximum enzyme activity.
- **Higher Specificity:** Minimize nonspecific amplification and primer-dimers.
- **Improve Productivity:** Go directly from PCR to gel analysis. Green GoTaq® Reaction Buffer serves as both reaction buffer and gel-loading solution.
- **Convenient:** One tube, one pipetting step. Only add template and primers when using the master mixes.
- **Optimization:** Control the magnesium in your reaction for specialized templates when using the standalone polymerase.

Protocol	Part#
GoTaq® Hot Start Polymerase Product Information	9PIM500
GoTaq® Hot Start Green Master Mix Product Information	9PIM512
GoTaq® Hot Start Colorless Master Mix Product Information	9PIM513

Storage Conditions: Store at –20°C.



Improve amplification of targets that require hot start using GoTaq® Hot Start Polymerase. A 1.5kb fragment of a *Corynephage* omega gene that requires hot start PCR was amplified from 500pg of plasmid DNA using either standard *Taq* or GoTaq® Hot Start Polymerase in Green and Colorless Flexi Reaction Buffers. Use of GoTaq® Hot Start Polymerase resulted in amplification of only the target 1.5kb fragment. Using standard *Taq* DNA Polymerase, a nonspecific 410bp product was also amplified.

*Taq*Bead™ Hot Start Polymerase

Product	Size	Cat.#
*Taq*Bead™ Hot Start Polymerase, 1.25u/bead, Nonbarrier	100 reactions	M5661

Description: *Taq*Bead™ Hot Start Polymerase consists of spherical beads of wax containing *Taq* DNA polymerase. *Taq*Bead™ Hot Start Polymerase facilitates hot-start PCR by keeping the enzyme sequestered in paraffin wax until the reaction temperature reaches approximately 60°C. This increases PCR specificity by keeping the polymerase separate from the rest of the reaction components until a critical temperature is reached, decreasing the probability of amplifying products that are the result of nonspecific binding of primers. The nonbarrier format of the bead is intended for use with heated-lid thermal cyclers or with the addition of a mineral oil overlay.

Thermophilic DNA Polymerase 10X Reaction Buffer: 500mM KCl, 100mM Tris-HCl (pH 9.0) at 25°C, 1.0% Triton® X-100.

Magnesium Chloride: 25mM $MgCl_2$ Solution included.

Features:

- **Higher Yield PCR:** Hot-start PCR protocols increase amplimer yield by minimizing nonspecific priming, primer-dimer formation or other reactions that can occur at low temperatures once all the PCR amplification components are mixed.
- **Increased Specificity:** Hot-start format reduces the incidence of nonspecific amplification products.
- **Reliable:** Minimized nonspecific priming results in increased reproducibility of amplification reactions.
- **Flexible:** Sufficient 25mM $MgCl_2$ is provided separately to allow optimization of enzyme performance under different conditions.

Protocol	Part#
Technical Bulletin	TB247

Storage Conditions: Store at –20°C.

GoTaq® Amplification Family

Product	Size	Conc.	Cat.#
GoTaq® Flexi DNA Polymerase	100 u	5 u/µl	M8291
	500 u	5 u/µl	M8295
	2,500u (5 × 500 u)	5 u/µl	M8296
	5,000u (10 × 500 u)	5 u/µl	M8297
	10,000u (20 × 500 u)	5 u/µl	M8298
GoTaq® DNA Polymerase	100 u	5 u/µl	M3001
	500 u	5 u/µl	M3005
	2,500 u	5 u/µl	M3008
GoTaq® Green Master Mix	100 reactions		M7122
	1,000 reactions		M7123
GoTaq® Colorless Master Mix	100 reactions		M7132
	1,000 reactions		M7133

For Laboratory Use.

Description: Experience improved PCR performance with GoTaq® DNA Polymerase products. GoTaq® DNA Polymerase is a proprietary formulation of *Taq* DNA polymerase that gives robust amplification equal to and in some cases superior to that of standard *Taq* DNA polymerase. GoTaq® DNA Polymerase comes in a variety of formulations designed to give you maximum flexibility, control and convenience.

GoTaq® Flexi DNA Polymerase allows you to optimize enzyme and magnesium concentration in your PCR. The supplied 5X Green and Colorless Flexi Reaction Buffers do not contain magnesium. A separate tube of 25mM $MgCl_2$ is supplied, giving you maximum control over your reaction conditions. $MgCl_2$ is also available separately.

GoTaq® DNA Polymerase provides improved amplification with the convenience of reaction buffers containing magnesium. The 5X GoTaq® Green and Colorless Reaction Buffers supplied with GoTaq® DNA Polymerase contain $MgCl_2$ at a concentration of 7.5mM, giving a final concentration of 1.5mM in the 1X reaction.

The **5X Green and 5X Colorless Reaction Buffers** supplied with GoTaq® enzymes allow you to go directly from thermal cycler to gel analysis. These buffers contain a compound that increases sample density, so that samples sink easily into the wells of an agarose gel. The green buffer also contains two dyes (yellow and blue) that separate to allow easy monitoring during electrophoresis. The blue dye comigrates at the same rate as 3–5kb DNA fragments in a 1% agarose gel. The yellow dye migrates ahead of primers.

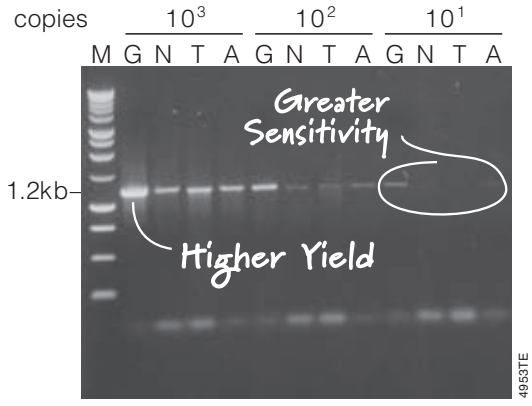
For ultimate convenience, choose **GoTaq® Colorless Master Mix** or **GoTaq® Green Master Mix**. Both are premixed, ready-to-use 2X solutions containing GoTaq® DNA Polymerase, dNTPs, $MgCl_2$ and reaction buffers at optimal concentrations for efficient amplification of DNA templates by PCR. GoTaq® Green Master Mix also includes two dyes (blue and yellow) that allow monitoring of progress during electrophoresis. GoTaq® Colorless Master Mix has the same formulation as the GoTaq® Green Master Mix but does not include the dyes. Both also include Nuclease-Free Water. Reactions assembled with the GoTaq® Master Mixes have sufficient density for direct loading onto agarose gels.

Features:

- **Improve Performance:** Experience better PCR performance with this new buffer and enzyme formulation. With GoTaq® Flexi, you also have the option to titrate Mg^{2+} concentration in your reactions.
- **Improve Productivity:** Go directly from PCR to gel analysis. Green GoTaq® Reaction Buffer serves as both reaction buffer and gel-loading solution.
- **Keep Your Cycling Conditions:** Directly substitute GoTaq® products, with either Colorless or Green Reaction Buffer, in your current PCR application—no need to change cycling parameters.
- **Use With PCR Enhancers:** GoTaq® DNA Polymerase is compatible with PCR enhancers such as betaine and DMSO. Neither compound affects the color or characteristics of the GoTaq® Green Reaction Buffer.
- **Fast and Convenient:** GoTaq® Green Master Mix and PCR Master Mix offer the ultimate in convenience. Reactions can be set up in less than a minute; simply add your template, water and primers and go!

Protocol	Part#
GoTaq® Flexi DNA Polymerase Product Information	9PIM829
GoTaq® DNA Polymerase Product Information	9PIM300
GoTaq® Green Master Mix Product Information	9PIM712
GoTaq® Colorless Master Mix Product Information	9PIM713

Storage Conditions: Store enzymes at –20°C. PCR Master Mix can be stored at 4°C for 3 months; GoTaq® Green Master Mix can be stored at 4°C for 6 weeks.



GoTaq® Green Master Mix outperforms standard *Taq* DNA Polymerase. Amplification of a 1.2kb fragment of the α-1-antitrypsin gene from indicated copies of Human Genomic DNA (Cat.# G3041).Lanes G, GoTaq® Green Master Mix; Lanes N, competitor N; Lanes T, competitor T; Lanes A, competitor A; and Lane M, BenchTop 1kb DNA Ladder (Cat.# G7541).

GoTaq® Reaction Buffers and Magnesium Chloride

Product	Size	Conc.	Cat.#
5X Green GoTaq® Reaction Buffer	20 ml		M7911
5X Colorless GoTaq® Reaction Buffer	20 ml		M7921
5X Colorless GoTaq® Flexi Reaction Buffer	20 ml		M8901
5X Green GoTaq® Flexi Reaction Buffer	20 ml		M8911
Magnesium Chloride Solution	1.5 ml	25 mM	A3511
	25 ml	25 mM	A3513

For Laboratory Use.

Description: The 5X Green GoTaq® Reaction Buffer contains two dyes (blue and yellow) that separate during electrophoresis to show migration progress. The buffer also contains a compound that increases sample density. This means that samples can be loaded directly onto gels without the need for loading dye. The blue dye migrates at the same rate as a 3–5kb DNA fragment in a 1% agarose gel. The yellow dye migrates at a rate faster than primers (The 5X Colorless GoTaq® Reaction Buffer has the same formulation as the 5X Green GoTaq® Reaction Buffer but does not contain dyes and is recommended for any applications where absorbance or fluorescence measurements are necessary prior to PCR cleanup. Both buffers are supplied at pH 8.5.

Cat.# M7911 and M7921 contain $MgCl_2$ at a concentration of 7.5mM, giving a final concentration of 1.5mM in the 1X reaction. Cat.# M8901 and M8911 do not contain magnesium.

Storage Conditions: Store at –20°C.

Section Contents
Table of Contents

GoTaq® PCR Core Systems

Product	Size	Cat.#
GoTaq® PCR Core System I	200 reactions	M7660
GoTaq® PCR Core System II	200 reactions	M7665

For Laboratory Use.

Description: The GoTaq® PCR Core Systems I and II are designed for the exponential amplification of specific regions of DNA using the polymerase chain reaction. Both systems include GoTaq® DNA polymerase and PCR Nucleotide Mix, along with high-performance buffers and magnesium chloride. The GoTaq® PCR Core System II also includes a Positive Control Plasmid DNA and Positive Control Primers, providing increased confidence in PCR control reactions. Each system's components are performance-tested in PCR and are sufficient for 200 reactions.

Features:

- **Convenient:** PCR-tested components are provided in optimized volumes for 200 reactions.
- **Flexible:** Optimization tools are provided for reaction flexibility.
- **Positive Controls:** The GoTaq® PCR Core System II provides Positive Control Plasmid DNA and Control Primers to help troubleshoot PCR parameters.
- **Performance Guarantee:** Promega PCR systems, enzymes and reagents are proven in PCR to ensure reliable, high-performance results. If you are not completely satisfied with any Promega PCR product, we will send a replacement or refund your account.

Protocol	Part#
Technical Bulletin	TB254

Storage Conditions: Store all components at –20°C.

PCR Master Mix

Product	Size	Conc.	Cat.#
PCR Master Mix	10 reactions	2 X	M7501
	100 reactions	2 X	M7502
	1,000 reactions	2 X	M7505

For Laboratory Use. Please contact Promega for information on bulk purchases.

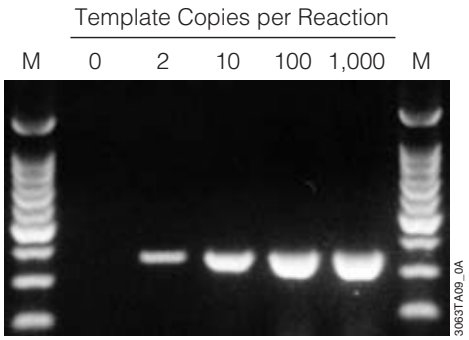
Description: PCR Master Mix is a premixed, ready-to-use solution containing *Taq* DNA polymerase, dNTPs, $MgCl_2$ and reaction buffers at optimal concentrations for efficient amplification of DNA templates by PCR. The PCR Master Mix has been optimized for use in routine PCR for amplifying DNA templates in the range of 0.2–2kb.

Features:

- **Fast:** Set up reactions in less than a minute.
- **Sensitive:** Amplify as little as 2 copies of target template.
- **Convenient:** One tube, one pipetting step.
- **Complete:** Reagents, including *Taq* DNA polymerase, $MgCl_2$, dNTPs and buffers, in one tube.
- **Scalable:** Set up 10µl, 25µl or 50µl reactions.
- **Stable:** Store at 4°C for up to 3 months.
- **Performance Guarantee:** Promega PCR systems, enzymes and reagents are proven in PCR to ensure reliable, high-performance results. If you are not completely satisfied with any Promega PCR product, we will send a replacement or refund your account.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Promega Product Information	9PIM750

Storage Conditions: Store at –20°C. PCR Master Mix can be stored at 4°C for up to 3 months.



Detection of low-copy-number templates using PCR Master Mix. Use of PCR Master Mix to detect low number of copies of the α-1-antitrypsin gene. PCR was performed on Human Genomic DNA (Cat.# G3041) using primers targeting a 360bp fragment of the α-1-antitrypsin gene (single copy per genome). Lane M, 100bp DNA Ladder (Cat.# G2101).

Tfl DNA Polymerase

Product	Size	Conc.	Cat.#
Tfl DNA Polymerase	100 u	5 u/µl	M1941
	1,000 u	5 u/µl	M1945

For Laboratory Use.

Description: *Tfl* DNA Polymerase is a thermostable enzyme of approximately 94kDa isolated from *Thermus flavus*. The enzyme replicates DNA at 74°C and exhibits a half-life of 40 minutes at 95°C. *Tfl* DNA Polymerase catalyzes the polymerization of nucleotides into duplex DNA in the 5′→3′ direction in the presence of magnesium and the polymerization of nucleotides into DNA using an RNA template in the 5′→3′ direction in the presence of manganese. The enzyme also possesses a 5′→3′ exonuclease activity. *Tfl* DNA Polymerase is recommended for use in PCR and primer extension reactions at elevated temperatures.

***Tfl* DNA Polymerase 10X Reaction Buffer:** 200mM Tris-acetate (pH 8.9 at 25°C), 100mM ammonium sulfate.

Magnesium Sulfate: 25mM $MgSO_4$ Solution included.

Features:

- **Flexible:** Provided with a 10X Reaction Buffer that does not contain magnesium. Sufficient 25mM $MgSO_4$ is provided separately to allow optimization of enzyme performance under different conditions.
- **Performance Guarantee:** Promega PCR systems, enzymes and reagents are proven in PCR to ensure reliable, high-performance results. If you are not completely satisfied with any Promega PCR product, we will send a replacement or refund your account.

Storage Conditions: Store at –20°C.

Pfu DNA Polymerase

Product	Size	Conc.	Cat.#
Pfu DNA Polymerase	100 u	2–3 u/µl	M7741
	500 u	2–3 u/µl	M7745

Product may not be available in all countries. Please contact your local representative for more information.

Description: *Pfu* DNA Polymerase is a thermostable enzyme of approximately 90kDa isolated from *Pyrococcus furiosus*. The enzyme replicates DNA at 75°C, catalyzing the polymerization of nucleotides into duplex DNA in the 5′→3′ direction in the presence of magnesium. *Pfu* DNA Polymerase also possesses 3′→5′ exonuclease (proofreading) activity. Base misinsertions that may occur during polymerization are rapidly excised by the proofreading activity of the polymerase. Consequently, *Pfu* DNA Polymerase is recommended for use in PCR and primer extension reactions that require high-fidelity synthesis. *Pfu* DNA Polymerase-generated PCR fragments are blunt-ended.

***Pfu* DNA Polymerase 10X Reaction Buffer with $MgSO_4$:** 200mM Tris-HCl (pH 8.8 at 25°C), 100mM KCl, 100mM $(NH_4)_2SO_4$, 20mM $MgSO_4$, 1.0% Triton® X-100 and 1mg/ml nuclease-free BSA.

Features:

- **High Fidelity:** *Pfu* DNA Polymerase exhibits the lowest error rate of any thermostable DNA polymerase.
- **Complete:** Provided with 10X Buffer containing 20mM $MgSO_4$.
- **Performance Guarantee:** Promega PCR systems, enzymes and reagents are proven in PCR to ensure reliable, high-performance results. If you are not completely satisfied with any Promega PCR product, we will send a replacement or refund your account.

Protocol	Part#
Promega Product Information	9PIM774

Storage Conditions: Store at –20°C.

GoTaq® 2-Step RT-qPCR System

Product	Size	Cat.#	
GoTaq® 2-Step RT-qPCR System	50 RT reactions + 200 qPCR reactions	A6010	435

For in vitro research use only. Not for use in diagnostic procedures.

Description: GoTaq® 2-Step RT-qPCR System is a reagent system for quantitative analysis of RNA using a two-step reverse transcription-quantitative PCR (RT-qPCR) protocol. The components and protocol allow cDNA synthesis using the GoScript™ Reverse Transcription System and quantitation using the GoTaq® qPCR Master Mix. The GoTaq® 2-Step RT-qPCR System contains a new fluorescent DNA-binding dye that often exhibits greater fluorescence enhancement upon binding to double-stranded DNA (dsDNA) than SYBR® Green I. GoTaq® qPCR Master Mix can be used with any real-time instrument capable of detecting SYBR® Green I or FAM™ dye. GoTaq® qPCR Master Mix contains a low level of CXR reference dye. A separate tube of CXR Reference Dye is included for use with instruments that require a higher level of reference dye.

Features:

GoTaq® 2-Step RT-qPCR System combines the ultra-active GoScript™ Reverse Transcriptase with the ultra-bright fluorescence of GoTaq® qPCR Master Mix to provide optimized, sensitive quantification of a full range of RNA targets.

- High-efficiency, full-length cDNA synthesis.
- Sensitive detection of low- and high-copy targets.
- Linear quantitation over a wide sample range.
- Robust activity in the presence of inhibitors.

Protocol	Part#
Technical Manual	TM337
Quick Protocol Card	FB115

Storage Conditions: Upon arrival, store all components at –20°C, protected from light. For immediate use, components maybe stored at 2–8°C, protected from light, for up to 3 months.

GoTaq® qPCR Master Mix

Product	Size	Cat.#
GoTaq® qPCR Master Mix	200 reactions	A6001
	1,000 reactions	A6002

For in vitro Research Use Only. Not for use in diagnostic procedures. Reaction size is based on a 50µl reaction volume of the mix (2X).

Description: GoTaq® qPCR Master Mix is a new reagent system for use in real-time quantitative PCR (qPCR). The system contains a new proprietary fluorescent DNA binding dye that exhibits greater fluorescence enhancement upon binding to double-stranded DNA (dsDNA) than SYBR® Green I. Combined with the GoTaq® Hot Start Polymerase, optimized buffer and proprietary dye, GoTaq® qPCR Master Mix provides robust real-time PCR with increased reliability, reproducibility and sensitivity. GoTaq® qPCR Master Mix is provided as a pre-mixed, ready-to-use stabilized 2X formulation that includes all components for qPCR except sample DNA, primers and water to dilute the DNA standards. This formulation, which includes a proprietary dsDNA-binding dye, a low level of carboxy-X-rhodamine (CXR) reference dye (identical to ROX™ dye), GoTaq® Hot Start Polymerase, $MgCl_2$, dNTPs and a proprietary reaction buffer, produces optimal results in qPCR experiments. A separate tube of CXR Reference Dye is included for use with instruments that require a higher level of reference dye than that in the GoTaq® qPCR Master Mix.

Advantages of the GoTaq® qPCR Master Mix

The proprietary dye provides brighter dsDNA-dependent fluorescence than SYBR® Green I. With less PCR inhibition than SYBR® Green I, the dye enables efficient amplification, often resulting in earlier cycle threshold (C_t) values and an expanded linear range using the same filters and settings as SYBR® Green I. The CXR reference dye can be detected using the same filters and settings as those used for ROX™. GoTaq® Hot Start Polymerase contains full-length *Taq* DNA polymerase bound to a proprietary antibody that prevents polymerase activity at room temperature. Thermal activation is achieved by incubating the assembled reaction at 95°C for 2 minutes. The proprietary polymerase/buffer formulation accommodates extended cycle numbers (45–50 cycles) and is compatible with thermal cycling programs that require extended activation (95°C for 10 minutes). GoTaq® qPCR Master Mix can be used with any real-time instrument capable of detecting SYBR® Green I or FAM™ dye. GoTaq® qPCR Master Mix contains a low level of CXR reference dye. If your real-time thermal cycler requires a high level of reference dye, add CXR Reference Dye to a final concentration of 1X in the reactions.

Features:

- Detection of low copy targets.
- Enhanced stability for automated setup.
- Direct substitute for SYBR® Green I products.
- The robust, reliable performance of GoTaq® Hot Start.
- The Promega PCR Performance Guarantee.

Protocol	Part#
Technical Manual	TM318
Quick Protocol Card	FB103

Storage Conditions: Store all components at –20°C, protected from light. For immediate use, components may be stored at 2–8°C, protected from light, for up to 3 months.

Plexor® qPCR and qRT-PCR Systems

Product	Size	Cat.#
Plexor® qPCR System	200 reactions	A4011
Plexor® One-Step qRT-PCR System	200 reactions	A4021
Plexor® Two-Step qRT-PCR System	200 reactions	A4051

For Research Use Only. Not for use in diagnostic procedures.

Description: The Plexor® qPCR and qRT-PCR Systems are multiplex-capable real-time amplification systems using novel base pair chemistry. Each target is measured directly during the amplification process and not through a secondary reaction. Plexor® reactions require only two primers for each target. Multiplex assay design is further simplified by the use of the web-based Plexor® Primer Design program.

The Plexor® Systems work by measuring a reduction in fluorescent signal during amplification. Amplification uses only two primers, one of which contains both a fluorescent tag and a modified base. As amplification proceeds, fluorescence is reduced by site-specific incorporation of a fluorescent quencher inserted opposite the complementary modified base. The quencher is in close proximity to a fluorescent dye located on the end of the primer, resulting in a reduction in the fluorescent signal. After PCR, a melt analysis can be performed to provide an internal control for the final assay design or to expedite troubleshooting during development. The system also includes a proprietary reagent to minimize primer-dimer formation.

Features:

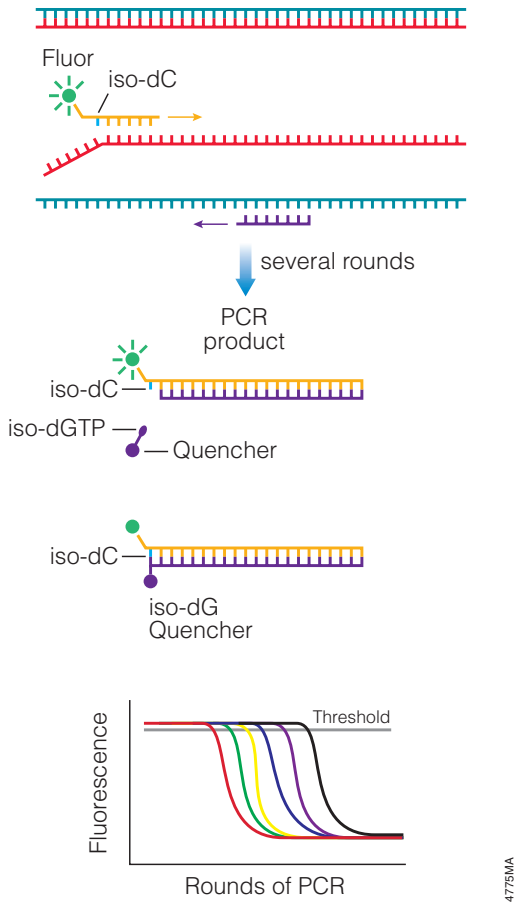
- **Multiplex Performance:** Only two primers are required for each target, making the design of multiplex assays much easier.
- **Improve Productivity:** Less labor, less time and less cost per assay. Measure controls and targets at the same time in the same well.
- **Enjoy Convenience:** The master mix format provides everything you need in one tube. Combine with your template and primers.
- **Obtain Strong Data:** Plexor® Systems measure a reduction in fluorescent signal during amplification. Quenching is directly proportional to amplicon accumulation. After amplification, a melt analysis can be performed to provide an internal control for specificity.
- **Use Your Existing Real-Time Instruments:** Plexor® technology works on most currently available real-time instruments capable of measuring more than one fluor. The free Plexor® Analysis Software allows users to import and analyze data from their preferred instrument platform.
- **Use Free Design and Analysis Tools:** Our free web-based design program will assist in your multiplex assay design. Once you have performed the assay, export the raw data and analyze it with the Plexor® Analysis Software.

Three Simple Steps to Use Plexor® Systems:

- **Step 1. Design Your Assay:** Simple online tools for design of your multiplex assay. Choose your primer sets, then order from your preferred oligo provider.
- **Step 2. Run the Assay:** Instruction manuals are available for a wide variety of real-time instruments including Applied Biosystems and Roche. All Plexor® assays are designed for the same cycling conditions.
- **Step 3. Analyze Your Data:** Export the raw data from your real-time instrument, then import into our free Plexor® Analysis Software. The Plexor® software converts the quenching data into cycle threshold (C_t) values and generates standard curves.

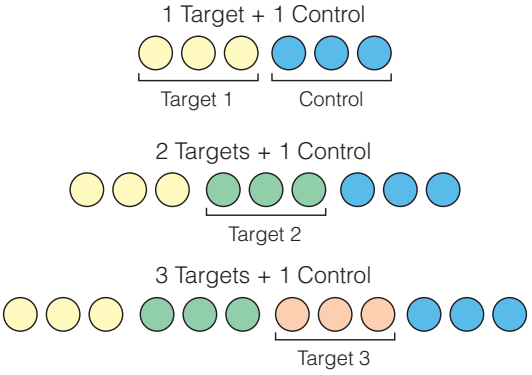
Protocol	Part#
Plexor® qPCR System Technical Manual	TM262
Plexor® One-Step qRT-PCR System Technical Manual	TM263
Plexor® Two-Step qRT-PCR System Technical Manual	TM264

Storage Conditions: Store at –20°C.

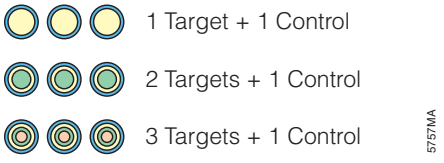


Overview of the core technology in the Plexor® Real-Time PCR and RT-PCR Systems.

Monoplex Method of Assaying Multiple Targets



Plexor® Method of Assaying Multiple Targets



Comparison of Plexor® method to monoplex analysis. The multiplexing power of the Plexor® System allows you to perform the equivalent of 12 wells of monoplex analysis in only 3 wells. Productivity is increased and savings are realized because you use less master mix and less plasticware, and you have to perform fewer steps.

MOPS/EDTA Buffer

Product	Size	Cat.#
MOPS/EDTA Buffer	3 × 10 ml	Y5101

Description: MOPS/EDTA Buffer is provided at pH 7.4 for the resuspension and dilution of primers and templates used in the Plexor® qPCR and qRT-PCR Systems. It is critical that this MOPS/EDTA Buffer be used with the Iso-dC-containing primers used in the Plexor® Systems, as these primers are sensitive to pH below 7.0.

Storage Conditions: Store at any temperature.

StemElite™ Gene Expression System

Product	Size	Cat.#
StemElite™ Gene Expression System	100 qPCR reactions	B1001
StemElite™ Gene Expression System Plus	100 qPCR reactions + 50 RT reactions	B1002

For in vitro Research Use Only. Not for use in diagnostic procedures.

Description: The StemElite™ Gene Expression System is a novel real-time quantitative PCR (qPCR) system for the detection and relative quantification of RNA expression levels associated with the differentiation state or "potency" of cells. The StemElite™ Gene Expression System is optimized to quantitatively amplify a two-color duplex, enabling the user to amplify a transcript of interest as well as a reference gene in a single reaction.

Features:

- Achieve sensitive real-time detection and relative quantification of RNA expression levels for monitoring of stem cell differentiation in a multiplexed amplification
- Quantitatively amplify a two-color duplex.
- Amplify in a single tube the transcript of interest and reference transcript (GAPDH and Actb).
- Reduce the number of reactions required for the measurement of multiple transcripts.
- Improve experimental data quality by measuring all the transcripts in the same well.

Protocol	Part#
Technical Manual	TM323

Storage Conditions: Store at –20°C.

StemElite™ Human Pluripotent Transcripts

Product	Size	Cat.#
StemElite™ NANOG/GAPDH Primer Pair (20X)	100 µl	B1011
StemElite™ SOX2/GAPDH Primer Pair (20X)	100 µl	B1021
StemElite™ POU5F1/GAPDH Primer Pair (20X)	100 µl	B1031
StemElite™ LIN28/GAPDH Primer Pair (20X)	100 µl	B1041
StemElite™ KLF4/GAPDH Primer Pair (20X)	100 µl	B1051
StemElite™ MYC/GAPDH Primer Pair (20X)	100 µl	B1061
Available Separately	**Size**	**Cat.#**
StemElite™ Gene Expression System	100 qPCR reactions	B1001
StemElite™ Gene Expression System Plus	100 qPCR reactions + 50 RT reactions	B1002

For Research Use Only. Not for use in diagnostic procedures.

Description: NANOG, SOX2, POU5F1, LIN28, KLF4 and MYC are functionally associated with maintenance of the undifferentiated human embryonic stem cell.

Features:

- Achieve sensitive real-time detection and relative quantification of RNA expression levels for monitoring of stem cell differentiation in a multiplexed amplification
- Quantitatively amplify a two-color duplex.
- Amplify in a single tube the transcript of interest and reference transcript (GAPDH).
- Reduce the number of reactions required for the measurement of multiple transcripts.
- Improve experimental data quality by measuring all the transcripts in the same well.

Protocol	Part#
StemElite™ Gene Expression System Technical Manual	TM323

Storage Conditions: Store at –20°C.

StemElite™ Human Heart-Associated Transcripts

Product	Size	Cat.#
StemElite™ NPPA/GAPDH Primer Pair (20X)	100 µl	B1071
StemElite™ MYL7/GAPDH Primer Pair (20X)	100 µl	B1081
StemElite™ MYL2/GAPDH Primer Pair (20X)	100 µl	B1091
StemElite™ MYH6/GAPDH Primer Pair (20X)	100 µl	B1101
StemElite™ MYH7/GAPDH Primer Pair (20X)	100 µl	B1111
StemElite™ NKX2-5/GAPDH Primer Pair (20X)	100 µl	B1121
StemElite™ TNNT2/GAPDH Primer Pair (20X)	100 µl	B1131
StemElite™ TNNI3/GAPDH Primer Pair (20X)	100 µl	B1141
StemElite™ MEF2C/GAPDH Primer Pair (20X)	100 µl	B1151
StemElite™ PLN/GAPDH Primer Pair (20X)	100 µl	B1161
StemElite™ GATA4/GAPDH Primer Pair (20X)	100 µl	B1171
Available Separately	**Size**	**Cat.#**
StemElite™ Gene Expression System	100 qPCR reactions	B1001
StemElite™ Gene Expression System Plus	100 qPCR reactions + 50 RT reactions	B1002

For Research Use Only. Not for use in diagnostic procedures.

Description: Pluripotential stem cells can give rise to differentiated cells and tissues for all three embryonic germ layers. NPPA, MYL7, MYL2, MYH6, MYH7, NKX2-5, TNNT2, TNNI3, MEF2C, PLN and GATA4 are mesodermal markers associated with the differentiation of cardiac muscle.

Features:

- Achieve sensitive real-time detection and relative quantification of RNA expression levels for monitoring of stem cell differentiation in a multiplexed amplification
- Quantitatively amplify a two-color duplex.
- Amplify in a single tube the transcript of interest and reference transcript (GAPDH).
- Reduce the number of reactions required for the measurement of multiple transcripts.
- Improve experimental data quality by measuring all the transcripts in the same well.

Protocol	Part#
StemElite™ Gene Expression System Technical Manual	TM323

Storage Conditions: Store at –20°C.

StemElite™ Human Pancreatic-Associated Transcripts

Product	Size	Cat.#
StemElite™ HNF4A/GAPDH Primer Pair (20X)	100 µl	B1301
StemElite™ HNF1B/GAPDH Primer Pair (20X)	100 µl	B1311
StemElite™ PDX1/GAPDH Primer Pair (20X)	100 µl	B1321
StemElite™ INS/GAPDH Primer Pair (20X)	100 µl	B1331
Available Separately	**Size**	**Cat.#**
StemElite™ Gene Expression System	100 qPCR reactions	B1001
StemElite™ Gene Expression System Plus	100 qPCR reactions + 50 RT reactions	B1002

For Research Use Only. Not for use in diagnostic procedures.

Description: Pluripotential stem cells can give rise to differentiated cells and tissues for all three embryonic germ layers. HNF4A, HNF1B, PDX1 and INS are mesodermal markers associated with the differentiation of pancreatic cells.

Features:

- Achieve sensitive real-time detection and relative quantification of RNA expression levels for monitoring of stem cell differentiation in a multiplexed amplification
- Quantitatively amplify a two-color duplex.
- Amplify in a single tube the transcript of interest and reference transcript (GAPDH).
- Reduce the number of reactions required for the measurement of multiple transcripts.
- Improve experimental data quality by measuring all the transcripts in the same well.

Protocol	Part#
StemElite™ Gene Expression System Technical Manual	TM323

Storage Conditions: Store at –20°C.

StemElite™ Differentiation-Associated Transcripts

Product	Size	Cat.#
StemElite™ FOXA2/GAPDH Primer Pair (20X)	100 µl	B1341
StemElite™ SOX17/GAPDH Primer Pair (20X)	100 µl	B1351
StemElite™ GATA6/GAPDH Primer Pair (20X)	100 µl	B1361
Available Separately	**Size**	**Cat.#**
StemElite™ Gene Expression System	100 qPCR reactions	B1001
StemElite™ Gene Expression System Plus	100 qPCR reactions + 50 RT reactions	B1002

For Research Use Only. Not for use in diagnostic procedures.

Description: Pluripotential stem cells can give rise to differentiated cells and tissues for all three embryonic germ layers. FOXA2, SOX17 and GATA6 are nonspecific differentiation markers.

Features:

- Achieve sensitive real-time detection and relative quantification of RNA expression levels for monitoring of stem cell differentiation in a multiplexed amplification
- Quantitatively amplify a two-color duplex.
- Amplify in a single tube the transcript of interest and reference transcript (GAPDH).
- Reduce the number of reactions required for the measurement of multiple transcripts.
- Improve experimental data quality by measuring all the transcripts in the same well.

Protocol	Part#
StemElite™ Gene Expression System Technical Manual	TM323

Storage Conditions: Store at –20°C.

StemElite™ Mouse Pluripotent Transcripts

Product	Size	Cat.#
StemElite™ Mus-Nanog/Actb Primer Pair (20X)	100 µl	B1371
StemElite™ Mus-Sox2/Actb Primer Pair (20X)	100 µl	B1381
StemElite™ Mus-Pou5f1/Actb Primer Pair (20X)	100 µl	B1391
StemElite™ Mus-Lin28/Actb Primer Pair (20X)	100 µl	B1401
StemElite™ Mus-Klf4/Actb Primer Pair (20X)	100 µl	B1411
StemElite™ Mus-Myc/Actb Primer Pair (20X)	100 µl	B1421
Available Separately	**Size**	**Cat.#**
StemElite™ Gene Expression System	100 qPCR reactions	B1001
StemElite™ Gene Expression System Plus	100 qPCR reactions + 50 RT reactions	B1002

For Research Use Only. Not for use in diagnostic procedures.

Description: Mus-Nanog, Mus-Sox2, Mus-Pou5f1, Mus-Lin28, Mus-Klf4 and Mus-Myc are functionally associated with maintenance of the undifferentiated mouse embryonic stem cell.

Features:

- Achieve sensitive real-time detection and relative quantification of RNA expression levels for monitoring of stem cell differentiation in a multiplexed amplification
- Quantitatively amplify a two-color duplex.
- Amplify in a single tube the transcript of interest and reference transcript (Actb).
- Reduce the number of reactions required for the measurement of multiple transcripts.
- Improve experimental data quality by measuring all the transcripts in the same well.

Protocol	Part#
StemElite™ Gene Expression System Technical Manual	TM323

Storage Conditions: Store at –20°C.

GoScript™ Reverse Transcription System

Product	Size	Cat.#
GoScript™ Reverse Transcription System	50 reactions	A5000
	100 reactions	A5001
Available Separately	**Size**	**Cat.#**
GoScript™ Reverse Transcriptase	100 reactions	A5003
	500 reactions	A5004

For Laboratory Use.

Description: The GoScript™ Reverse Transcription System includes a reverse transcriptase and a specialized set of reagents designed for efficient synthesis of first-strand cDNA optimized for quantitative PCR amplification. GoScript™ Reverse Transcriptase utilizes M-MLV and state-of-the-art buffer technology designed for qPCR to deliver robust, reliable cDNA synthesis of a full range of rare and abundant transcripts, even in the presence of inhibitors. GoScript™ Reverse Transcriptase is qualified for use in qPCR, including GoTaq® qPCR and Plexor® qPCR systems for performing RT-qPCR.

Features:

- **Sensitive:** Detect rare transcripts.
- **Processive:** Transcribe long messages.
- **Resilient:** Synthesize cDNA in the presence of strong inhibitors.

Protocol	Part#
Technical Manual	TM316

Storage Conditions: Store at –20°C.

ImProm-II™ Reverse Transcription System

Product	Size	Cat.#
ImProm-II™ Reverse Transcription System	100 reactions	A3800
Available Separately	**Size**	**Cat.#**
ImProm-II™ Reverse Transcriptase	10 reactions	A3801
	100 reactions	A3802
	500 reactions	A3803

For Laboratory Use.

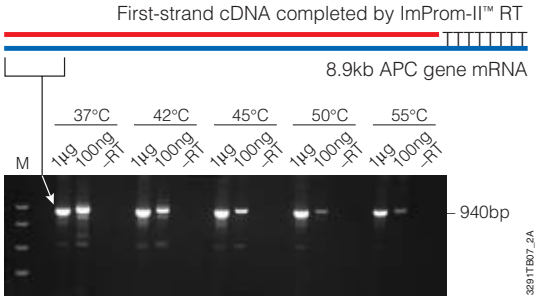
Description: The ImProm-II™ Reverse Transcription System produces efficient, robust synthesis of first-strand cDNA in preparation for PCR amplification. The components of the ImProm-II™ Reverse Transcription System can be used to reverse transcribe RNA templates starting with total RNA, poly(A)+ mRNA or synthetic transcript RNA. The optimized reaction buffer and powerful ImProm-II™ Reverse Transcriptase provided in the ImProm-II™ System together enable robust, full-length cDNA synthesis for the reproducible analysis of rare or long messages. The cDNA synthesis conditions have been formulated for standalone applications or for easy transition to gene-specific target amplification. From 1–20µl of the reverse transcription reaction can be directly amplified using *Taq* DNA polymerase in coupled or uncoupled PCR.

Features:

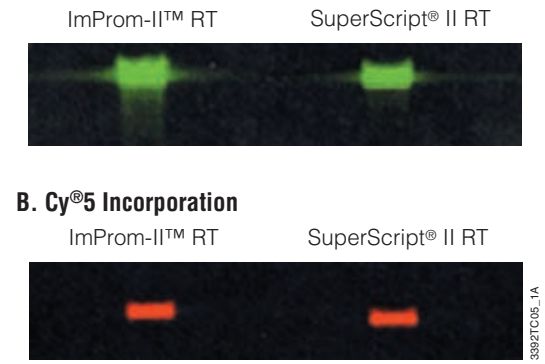
- **Full-Length RT-PCR:** Reverse transcribe long RNA template up to 8.9kb.
- **Microarray-Compatible:** May be used for incorporation of regular, Cy®3-modified, Cy®5-modified, and amino-allyl-modified nucleotides.
- **Easy to Use:** Kit format provides all reagents necessary for efficient reverse transcription.
- **Scalable and Flexible:** From 1–20µl from the initial RT reaction may be used in subsequent PCR, and the optimized buffer also allows for coupled RT-PCR.
- **Versatile:** Use with your thermostable DNA polymerase of choice.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Manual	TM236

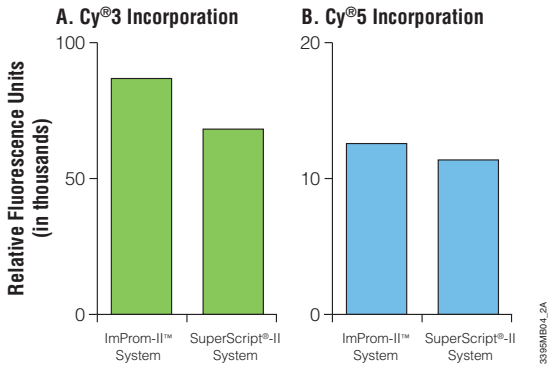
Storage Conditions: Store at –20°C. Store Positive Control RNA at –70°C.



Full-length cDNA synthesis of 8.9kb template over a range of temperatures using the ImProm-II™ Reverse Transcription System as demonstrated by selective amplification of terminal 3′ sequences in two-step RT-PCR. Entire 8.9kb message must be reverse transcribed by the ImProm-II™ RT from the oligo(dT) primer to amplify the terminal 940bp sequence. Message was amplified from either 1µg or 100ng of total RNA. Control reactions without the reverse transcriptase are shown (–RT) as well. Details of experiment available in the *ImProm-II™ Reverse Transcription System Technical Manual,* #TM236.



Incorporation studies of fluorescently labeled nucleotides. ImProm-II™ Reverse Transcription System allows for high-efficiency incorporation of Cy®3 and Cy®5 fluorescent nucleotides. This demonstrates fluorescent nucleotide incorporation by ImProm-II™ RT vs.SuperScript® II RT using a 1.2kb kanamycin transcript as template. A single fluorescent band is produced and visualized using an FMBIO® II Fluorescence Imaging System.



Relative Cy®3 and Cy®5 nucleotide incorporation by ImProm-II™ Reverse Transcription System in comparison to Superscript® II First Strand Synthesis System. Results with Cy®3 dUTP **(Panel A)** and Cy®5 dUTP **(Panel B)** incorporation are reported. Panels A and B correspond to Panels A and B in the figure above.

Reverse Transcription System

Product	Size	Cat.#
Reverse Transcription System	100 reactions	A3500
Available Separately	**Size**	**Conc.**	**Cat.#**
Magnesium Chloride Solution	1.5 ml	25 mM	A3511
Reverse Transcription 10X Buffer	1.4 ml		A3561

Cat.# A3511, A3561 For Laboratory Use.

Description: The Reverse Transcription System provides reagents to efficiently reverse transcribe RNA into cDNA in fifteen minutes. The cDNA prepared from each reaction using this system may be used directly in multiple PCR amplifications using *Taq* DNA polymerase. The AMV Reverse Transcriptase synthesizes single-stranded cDNA from total or poly(A)+ RNA. Both Oligo(dT)$_{15}$ and Random Primers are included, allowing cDNA synthesis from virtually any RNA source. The system contains sufficient reagents for 100 cDNA synthesis reactions, processing 1µg of RNA per reaction. Each cDNA synthesis reaction may be divided and used in up to 20 separate PCR amplifications. A polyadenylated 1.2kb RNA transcript is provided as a control template for the cDNA synthesis reaction.

Features:

- **Fast:** Efficiently reverse transcribe poly(A)+ mRNA or total RNA in fifteen minutes.
- **Convenient:** PCR-compatible components are provided in optimized volumes for 100 reactions.
- **Positive Controls:** A polyadenylated RNA transcript is provided to help troubleshoot RT-PCR parameters.

Protocol	Part#
Technical Bulletin	TB099

Storage Conditions: Store at –20°C. Store Positive Control RNA at –70°C.

AccessQuick™ RT-PCR System

Product	Size	Cat.#
AccessQuick™ RT-PCR System	20 reactions	A1701
	100 reactions	A1702
	500 reactions	A1703

Cat.# A1702, A1703 For Laboratory Use.

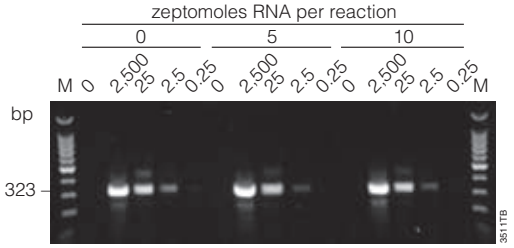
Description: The AccessQuick™ RT-PCR System is an easy and convenient master mix system for setting up one-tube RT-PCR. It is designed to increase convenience by combining the following components in a single tube: *Tfl* DNA Polymerase, dNTPs, magnesium sulfate and reaction buffer. The AMV RT enzyme is provided in a separate tube to allow important, no-RT control reactions. The AccessQuick™ Master Mix is simply added to RNA templates in reaction vials, followed by the AMV RT, primers and water. The AccessQuick™ RT-PCR Master Mix is intended for routine RT-PCR applications that have been previously optimized and do not require extreme conditions.

Features:

- **Achieve Maximum Convenience:** Save yourself four pipetting steps. Simply combine the AccessQuick™ Master Mix, AMV RT, your gene-specific primers, your RNA template and water. Separate AMV RT allows important, no-RT control reactions.
- **Use Less Template:** Amplify from zeptomole (10^{-21}mol) levels of RNA.
- **No Buffer Additions Required:** Set up reactions in a single tube, drop into a thermal cycler, come back later for results—no additions between the reverse transcription and DNA amplification steps.
- **Stable:** System components are stable over many freeze-thaw cycles.

Protocol	Part#
Promega Product Information	9PIA170

Storage Conditions: Store all system components at –20°C.



Stability of AccessQuick™ Master Mix through multiple freeze-thaw events. Rapid freeze-thaw events were performed 0, 5 and 10 times by removing a sample of the AccessQuick™ Master Mix from –70°C storage and placing it in a 50°C heat block. After 5 cycles, and again after 10 cycles, we added AMV RT, primers and RNA. All samples were then used in RT-PCR reactions to amplify a 323bp fragment using the indicated amounts of the 1.2kb Kanamycin Positive Control RNA (Cat.# C1381) template.Lane M = 100bp DNA Ladder (Cat.# G2101).

Access RT-PCR System

Product	Size	Cat.#
Access RT-PCR Introductory System	20 reactions	A1260
Access RT-PCR System	100 reactions	A1250
	500 reactions	A1280

Cat.# A1250, A1280 For Laboratory Use.

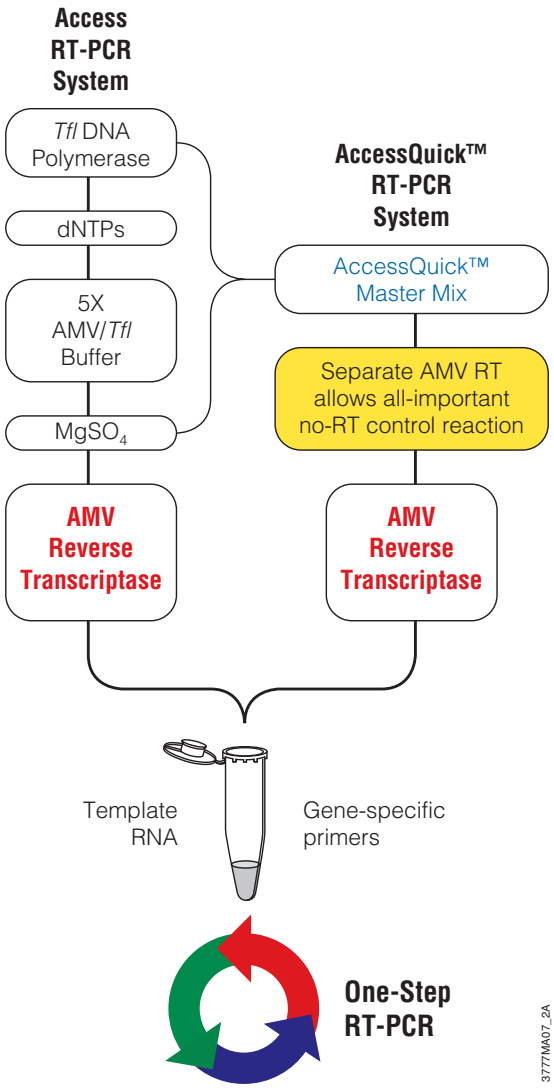
Description: The Access RT-PCR System is designed for the reverse transcription (RT) and polymerase chain reaction (PCR) amplification of a specific target RNA from either total RNA or mRNA. This one-tube, two-enzyme system provides sensitive, quick, and reproducible analysis of even rare RNAs. The system uses AMV Reverse Transcriptase (AMV RT) from Avian Myeloblastosis Virus for first-strand DNA synthesis and the thermostable *Tfl* DNA Polymerase from *Thermus flavus* for second-strand cDNA synthesis and DNA amplification. The Access RT-PCR System includes an optimized single-buffer system that permits extremely sensitive detection of RNA transcripts, without a requirement for buffer additions between the reverse transcription and PCR amplification steps. This simplifies the procedure and reduces the potential for contaminating the samples. In addition, the improved performance of AMV Reverse Transcriptase at elevated temperatures in the AMV/*Tfl* 5X Reaction Buffer minimizes problems encountered with secondary structures in RNA.

Features:

- **Achieve Maximum Control:** Separate tubes of each component allow you to control every step of the reaction. You can optimize Mg^{2+}, perform no-reverse transcriptase control reactions, etc.
- **Use Less Template:** Detect message from as little as 1pg of total RNA or mRNA.
- **No Buffer Additions Required:** The AMV/*Tfl* 5X Reaction Buffer provides results in optimal enzyme activity without buffer additions between the reverse transcription and DNA amplification steps.
- **Rely on a Performance-Tested System:** Promega PCR Systems, enzymes and reagents are proven in PCR to ensure reliable, high-performance results. If you are not completely satisfied with any Promega PCR product, we will send a replacement or refund your account.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB220

Storage Conditions: Store all system components at –20°C. For long-term storage, the Positive Control RNA with Carrier must be stored at –70°C.



Features of Access and AccessQuick™ RT-PCR Systems.

	Access RT-PCR System *Maximum Control*	AccessQuick™ RT-PCR System *Maximum Convenience*
Components	Individual tubes of *Tfl* DNA Polymerase, AMV RT, dNTPs and reaction buffer	*Tfl* DNA Polymerase, dNTPs and reaction buffer combined in master mix. AMV RT in separate tube
Mg^{2+} Concentration	Adjustable	1.5mM
Controls Included	Yes	No

9478LA

Tth DNA Polymerase

Product	Size	Conc.	Cat.#
Tth DNA Polymerase	100 u	5 u/µl	M2101
	500 u	5 u/µl	M2105

Description: *Tth* DNA Polymerase is a thermostable enzyme of approximately 94kDa isolated from *Thermus thermophilus* HB-8. The enzyme replicates DNA at 74°C and exhibits a half-life of 20 minutes at 95°C. *Tth* DNA Polymerase catalyzes the polymerization of nucleotides into duplex DNA in the 5′→3′ direction in the presence of magnesium and the polymerization of nucleotides into DNA using an RNA template in the 5′→3′ direction in the presence of manganese. The enzyme also possesses a 5′→3′ exonuclease activity. *Tth* DNA Polymerase is recommended for use in PCR, RT-PCR, reverse transcription and primer extension reactions at elevated temperatures.

10X Reverse Transcription Buffer: 100mM Tris-HCl (pH 8.3 at 25°C), 900mM KCl.

10X Chelate Buffer: 100mM Tris-HCl (pH 8.3 at 25°C), 1M KCl, 7.5mM EGTA, 0.5% Tween® 20, 50% glycerol.

Thermophilic DNA Polymerase 10X Reaction Buffer: 500mM KCl, 100mM Tris-HCl (pH 9.0 at 25°C) and 1% Triton® X-100. Buffer is optimized for use with 0.2mM of each of the dNTPs.

Manganese and Magnesium Chloride: 10mM $MnCl_2$ and 25mM $MgCl_2$ Solutions provided.

Features:

- **Increased Specificity for RT-PCR:** The ability to reverse transcribe at higher temperatures results in increased specificity of primer hybridization and extension.
- **Minimized Secondary Structures:** Higher temperature RT-PCR minimizes problems associated with strong secondary structures in RNA.
- **Performance Guarantee:** Promega PCR systems, enzymes and reagents are proven in PCR to ensure reliable, high-performance results. If you are not completely satisfied with any Promega PCR product, we will send a replacement or refund your account.

Protocol	Part#
Promega Product Information	9PIM210

Storage Conditions: Store at –20°C.

Primer Pairs for RT-PCR

Product	Size	Cat.#
β-Actin Primer Pair	20 reactions	G5740
CNTF Primer Pair	20 reactions	G5770
NT-3 Primer Pair	20 reactions	G6801
p75 Primer Pair	20 reactions	G6861

Description: Promega Primer Pairs for RT-PCR provide convenient primers for analyzing the expression of specific mRNAs by RT-PCR. Each Primer Pair includes a 5′ and 3′ oligonucleotide primer provided in individual vials at 100µM ready for addition to the amplification reaction. The Primer Pairs have been confirmed in two-step conventional RT-PCR (Reverse Transcription System, Cat.# A3500) using rat brain total RNA. They also have been tested in one-step RT-PCR (Access RT-PCR System, Cat.# A1260) with human and mouse brain total RNA.

PCR products from the use of the Primer Pairs have been sequence-confirmed for appropriate identity.

Storage Conditions: Store at –20°C.

Universal RiboClone® cDNA Synthesis System

Product	Size	Cat.#
Universal RiboClone® cDNA Synthesis System	1 system	C4360
Available Separately	**Size**	**Cat.#**
Oligo(dT)$_{15}$ Primer	20 µg	C1101
Random Primers	20 µg	C1181
Spin Columns	10 each	C1281
EcoRI Adaptors	150 pmol	C1291
1.2kb Kanamycin Positive Control RNA	5 µg	C1381
Sephacryl® S-400	10 ml	V3181

Cat.# C1181, C1381 and V3181 For Laboratory Use.

Description: The Universal RiboClone® cDNA Synthesis System contains the reagents required for the synthesis of double-stranded cDNA from mRNA and subsequent ligation into a suitable vector. The system is based on the method described by Okayama and Berg with modifications by Gubler and Hoffman. First-strand synthesis is driven by AMV (Avian Myeloblastosis Virus) Reverse Transcriptase and either Random Primers or an Oligo(dT)$_{15}$ Primer, followed directly by second-strand replacement synthesis using RNase H and DNA Polymerase I. After treatment with T4 DNA Polymerase to flush the ends, the double-stranded cDNA molecules are prepared for cloning by size fractionation and the addition of EcoRI Adaptors. The resulting cDNA preparation can then be cloned into a suitable vector.

Features:

- **Convenient:** Contains all the necessary reagents to synthesize double-stranded cDNA from RNA.
- **Flexible:** Both Oligo(dT)$_{15}$ Primer and Random Primers are included, providing the researcher a choice of priming methods.

Protocol	Part#
Technical Manual	TM038

Storage Conditions: Store control RNA at –70°C. Store Sephacryl® S-400 at 4°C and Spin Columns at room temperature. Store other components at –20°C.

Oligonucleotides and Primers: cDNA Synthesis and Cloning

Product	Size	Cat.#
Oligo(dT)$_{15}$ Primer	20 µg	C1101
Random Primers	20 µg	C1181
EcoRI Adaptors	150 pmol	C1291

Cat.# C1181 For Laboratory Use.

Description: Oligo(dT)$_{15}$ Primer is suitable for the use of a primer for first-strand cDNA synthesis with a reverse transcriptase. The primer hybridizes to the poly(A) tail of mRNA.

Random Primers can be used for first-strand cDNA synthesis and cloning; they are also available as components of the Universal Riboclone® cDNA Synthesis System (Cat.# C4360) and the Reverse Transcription System (Cat.# A3500). The primers are random hexadeoxynucleotides.

The **EcoRI Adaptors** consist of two complementary oligonucleotides: a 16mer and a 12mer phosphorylated at the 5′-end. The oligonucleotides are provided annealed in equimolar concentrations in water. The EcoRI Adaptors are designed to attach EcoRI "sticky" ends to blunt-ended DNA.

Storage Conditions: Store at –20°C.

PCR Nucleotide Mix

Product	Size	Conc.	Cat.#
PCR Nucleotide Mix	200 µl	10 mM	C1141
	1,000 µl	10 mM	C1145

For Laboratory Use.

Description: High-quality deoxynucleotide triphosphates (dNTPs) are critical for the success of the polymerase chain reaction (PCR). The PCR Nucleotide Mix is a premixed solution containing the sodium salts of dATP, dCTP, dGTP and dTTP, each at a concentration of 10mM in water; the total concentration of nucleotides, therefore, is 40mM (pH 7.5). This solution is ready to use and is optimized for standard polymerase chain reactions and specialty approaches including hot-start and reverse transcription PCR (RT-PCR). One microliter (1µl) is sufficient for PCR amplification in a typical 50µl reaction volume.

Features:

- **Optimized and Pretested in PCR:** Equimolar amounts of each dNTP ensure optimal PCR.
- **Convenient:** 1µl addition for 50µl PCR.
- **Easy to Use:** Reduced pipetting steps contribute to ease-of-use and reduce the risk of contamination.
- **Performance Guarantee:** Promega PCR systems, enzymes and reagents are proven in PCR to ensure reliable, high-performance results. If you are not completely satisfied with any Promega PCR product, we will send a replacement or refund your account.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Promega Product Information	9PIC114

Storage Conditions: Store at –20°C.

dNTP Mix

Product	Size	Conc.	Cat.#
dNTP Mix	200 µl	10 mM	U1511
	1,000 µl	10 mM	U1515

For Laboratory Use.

Description: dNTP Mix is a premixed solution containing sodium salts of dATP, dCTP, dGTP and dTTP, each at 10mM in water; the total concentration of nucleotides, therefore, is 40mM (pH 7.5). One microliter of the dNTP Mix into a 50µl reaction will give a final dNTP concentration of 200µM for each dNTP.

Features:

- **High Purity:** dNTPs are >98% pure.
- **Easy to Use:** Reduced pipetting steps contribute to ease-of-use and reduce the risk of contamination.

Storage Conditions: Store at –20°C.

Deoxynucleotide Triphosphates (dNTPs)

Product	Size	Conc.	Cat.#
dATP	25 µmol	100 mM	U1205
	40 µmol	100 mM	U1201
	200µmol (2 × 100 µmol)	100 mM	U1202
dGTP	25 µmol	100 mM	U1215
	40 µmol	100 mM	U1211
	200µmol (2 × 100 µmol)	100 mM	U1212
dCTP	25 µmol	100 mM	U1225
	40 µmol	100 mM	U1221
	200µmol (2 × 100 µmol)	100 mM	U1222
dTTP	25 µmol	100 mM	U1235
	40 µmol	100 mM	U1231
	200µmol (2 × 100 µmol)	100 mM	U1232
Set of dATP, dCTP, dGTP, dTTP	10 µmol each	100 mM	U1330
	25 µmol each	100 mM	U1420
	40 µmol each	100 mM	U1240
	200µmol (2 × 100 µmol each)	100 mM	U1410

For Laboratory Use.

Description: High-quality deoxynucleotide triphosphates (dNTPs) are critical for the success of many key procedures such as cDNA synthesis, sequencing and labeling. Promega dNTPs are greater than 98% triphosphate content and are provided at a concentration of 100mM in water at pH 7.5.

Features:

- **Dependable:** PCR-tested deoxynucleotides ensure optimal performance with all Promega amplification enzymes.
- **Consistent:** dNTPs are >98% pure, allowing highly consistent results.
- **Convenient:** Supplied at a convenient concentration (100mM in water) for ease-of-use in PCR and other applications.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Storage Conditions: Store at –20°C.

PCR Amplifications From Each Size of Individual dNTPs.
Each catalog number supplies each individual dNTP at 100mM. Reactions are based on 200µM each dNTP in a 50µl reaction.

Cat.#	Quantity	Volume	Reactions
U1330, U1335	10 µmol each	100 µl each	1,000
U1420	25 µmol each	250 µl each	2,500
U1240, U1245	40 µmol each	400 µl each	4,000
U1410	200 µmol each	2 × 1,000 µl each	20,000

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Deoxyuridine Triphosphate (dUTP)

Product	Size	Conc.	Cat.#
dUTP	40 µmol	100 mM	U1191
Set of dATP, dCTP, dGTP, dUTP	10 µmol each	100 mM	U1335
	40 µmol each	100 mM	U1245

For Laboratory Use.

Description: dUTP (2´-Deoxyuridine, 5´-Triphosphate) can be used in place of dTTP in PCR and RT-PCR protocols to prevent carryover from previous amplifications. The substitution of dUTP for dTTP in PCR results in uracil-containing PCR products that are suitable for most standard applications. The enzyme uracil-N-glycosylase, UNG (also referred to as UDG), can be added to a PCR premix to excise uracil from any contaminating PCR product, thereby preventing false positives. Each lot of dUTP is function-tested to ensure specific DNA amplification and the absence of nuclease activity.

Features:

- **Dependable:** PCR-tested deoxynucleotides ensure optimal performance with all Promega amplification enzymes.
- **Consistent:** dUTP is ≥98% pure, allowing highly consistent results.
- **Convenient:** Supplied at a convenient concentration (100mM in water) for ease-of-use in PCR and other applications.

Storage Conditions: Store at –20°C.

pGEM®-T Vector Systems

Product	Size	Cat.#
pGEM®-T Vector System I	20 reactions	A3600
pGEM®-T Vector System II	20 reactions	A3610

Description: The pGEM®-T Vector Systems are convenient systems for the cloning of PCR products. The pGEM®-T Vector is prepared by cutting the pGEM®-5Zf(+) Vector with EcoRV and adding a 3′ terminal thymidine to both ends. These single 3′-T overhangs at the insertion site greatly improve the efficiency of ligation of a PCR product into the plasmid by preventing recircularization of the vector and providing a compatible overhang for ligation of PCR products generated by certain thermostable polymerases. These polymerases often add a single deoxyadenosine, in a template-independent fashion, to the 3′-ends of amplified fragments.

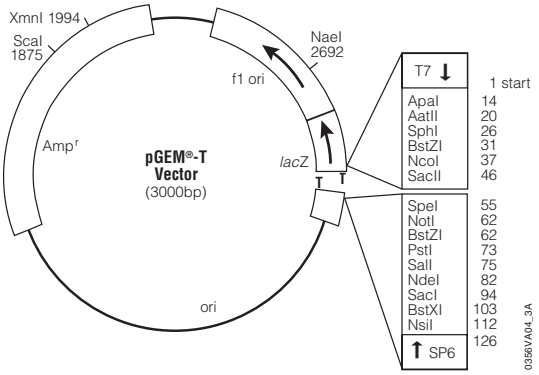
The multiple cloning site is flanked by recognition sites for the restriction enzyme BstZI, allowing release of the insert by a single-enzyme digestion. Alternatively, a double digestion may be used to release the insert from the vector. The pGEM®-T Vector System II contains JM109 Competent Cells in addition to all of the pGEM®-T Vector System I components.

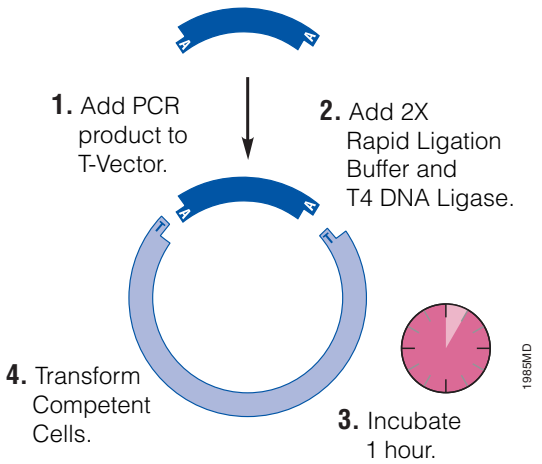
Features:

- **Rapid Ligation:** The 2X Rapid Ligation Buffer provided allows reactions to be completed in 1 hour at room temperature.
- **Blue/White Screening:** T7 and SP6 RNA polymerase promoters flank a multiple cloning region within the α-peptide coding region for β-galactosidase. Insertional inactivation of the α-peptide allows recombinant clones to be directly identified by color screening on indicator plates.
- **f1 Origin of Replication:** Allows the preparation of single-stranded DNA.

Protocol	Part#
Technical Manual	TM042

Storage Conditions: Store competent cells at –70°C; store all other components at –20°C.





The rapid ligation reaction reduces ligation time to just 60 minutes.

pGEM®-T Easy Vector Systems

Product	Size	Cat.#
pGEM®-T Easy Vector System I	20 reactions	A1360
pGEM®-T Easy Vector System II	20 reactions	A1380

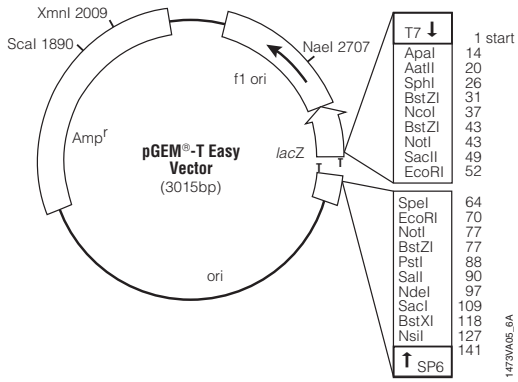
Description: The pGEM®-T Easy Vector Systems are convenient systems for the cloning of PCR products. They offer all of the advantages of the pGEM®-T Vector Systems with the added convenience of recognition sites for EcoRI and NotI flanking the insertion site. Thus several options for removal of the desired insert DNA with a single restriction digestion are provided. The pGEM®-T Easy Vector System II contains JM109 Competent Cells in addition to all of the pGEM®-T Easy Vector System I components.

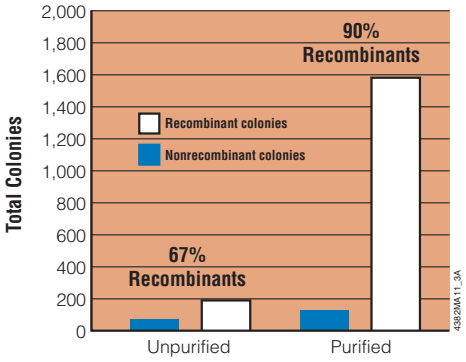
Features:

- **Flexible:** The multiple cloning site is flanked by restriction enzyme sites for BstZI, NotI and EcoRI, allowing three options for removal of the insert with a single digest.
- **Rapid Ligation:** The 2X Rapid Ligation Buffer provided allows reactions to be completed in 1 hour at room temperature.
- **Blue/White Screening:** T7 and SP6 RNA polymerase promoters flank a multiple cloning region within the α-peptide coding region for β-galactosidase. Insertional inactivation of the α-peptide allows recombinant clones to be directly identified by color screening on indicator plates.
- **f1 Origin of Replication:** Allows the preparation of single-stranded DNA.

Protocol	Part#
Technical Manual	TM042

Storage Conditions: Store competent cells at –70°C; store all other components at –20°C.





Purification of PCR products enhances cloning success. A 500bp PCR product was purified with the Wizard® SV Gel and PCR Clean-Up System and cloned into the pGEM®-T Easy Vector. Both the percent recombinants and total number of colonies increase with a pure PCR product. White bars represent recombinant colonies. Blue bars represent nonrecombinant colonies.

pTARGET™ Mammalian Expression Vector System

Product	Size	Cat.#
pTARGET™ Mammalian Expression Vector System	20 reactions	A1410

Description: The pTARGET™ Mammalian Expression Vector System is a convenient system for cloning PCR products and for expression of cloned PCR products in mammalian cells. The vector is prepared by digestion with EcoRV followed by addition of a 3′ terminal thymidine to each end. These single 3′-T overhangs at the insertion site greatly improve the efficiency of ligation of a PCR product into the plasmid in two ways. First, the overhangs prevent recircularization of the vector; second, they provide a compatible overhang for PCR products generated by certain thermostable polymerases. These polymerases often add a single deoxyadenosine, in a template-independent fashion, to the 3′-ends of amplified fragments. The pTARGET™ Vector also contains a modified version of the coding sequence of the α-peptide of β-galactosidase, which allows recombinants to be selected using blue/white screening.

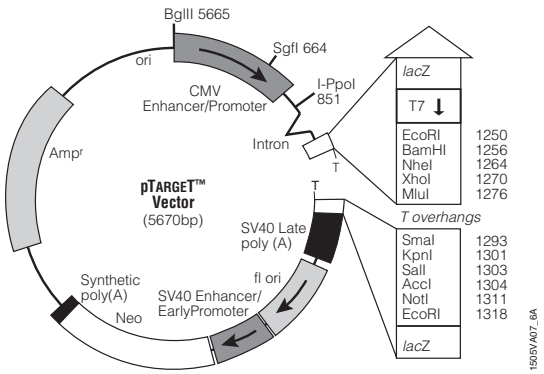
The pTARGET™ Vector carries the human cytomegalovirus (CMV) immediate-early enhancer/promoter region to promote constitutive expression of cloned DNA inserts in mammalian cells. This vector also contains the neomycin phosphotransferase gene, a selectable marker for mammalian cells. The pTARGET™ Vector can be used for transient expression or for stable expression by selecting transfected cells with the antibiotic G-418.

Features:

- **Simple PCR Cloning:** "T" overhangs permit direct ligation of PCR products generated by thermostable enzymes such as *Taq* DNA polymerase.
- **Strong, Constitutive Expression:** The CMV enhancer/promoter region allows strong, constitutive expression in many cell types. In transgenic mice, expression of the chloramphenicol acetyltransferase (CAT) gene under the regulation of the CMV enhancer/promoter was observed in 24 of the 28 tissues examined. The vector is maintained as an episome in cells expressing the SV40 large T antigen, leading to even higher levels of expression.
- **Blue/White Screening:** Allows the easy identification of recombinant clones. A single digest removes the insert DNA.
- **Stable Transfectants:** Select for stable transfectants using the neomycin phosphotransferase gene.

Protocol	Part#
Technical Manual	TM044

Storage Conditions: Store competent cells at –70°C; store all other components at –20°C or –70°C.



Promega Barrier Tips

Product	Size	Cat.#
Promega 10 Barrier Tips, 960/pk	0.5–10 µl	A1491
Promega 10E Barrier Tips, 960/pk	0.5–10 µl	A1501
Promega 10F Barrier Tips, 960/pk	0.5–10 µl	A1511
Promega 20 Barrier Tips, 960/pk	2–20 µl	A1521
Promega GEL Barrier Tips, 768/pk	0.5–30 µl	A1531
Promega 100 Barrier Tips, 960/pk	10–100 µl	A1541
Promega 200 Barrier Tips, 960/pk	50–200 µl	A1551
Promega 1000 Barrier Tips, 480/pk	100–1,000 µl	A1561

Description: Aerosol barrier tips eliminate false signals and contamination caused by aerosols. Scientifically designed and tested, Promega Barrier Tips offer performance and economy when working with amplified nucleic acids (PCR), radioactive isotopes, tissue culture fluids, infectious samples, forensic and serological specimens.

Promega Barrier Tips are made with an inert ultrahydrophobic HDPE plastic that offers the effectiveness of a self-sealing barrier with the convenience of sample retrieval. In retention tests, Promega Barrier Tips virtually eliminated tip retention and sample holdup.

Features:

- **Sterile:** Promega Barrier Tips are presterilized and certified RNase- and DNase-free. Tips are supplied packaged and sealed in covered trays.
- **Convenient:** Designed to fit perfectly on all major brands of pipettor.

Storage Conditions: Store at room temperature.

Tip/Pipette Compatibility Guide.

Tip	Size	Pipetman®	Eppendorf®	Oxford Benchmate®	Finn-pipette®
Promega 10	10µl	P-2 & P-10		0.5–10µl	0.5–10µl Digital
Promega 10E	10µl	P-2 & P-10	0.5–10µl	0.5–10µl	
Promega 10F	10µl				0.5–10µl
Promega 20	20µl	P-20	2–20µl		
Promega 100	100µl	P-100	10–100µl	10–50µl	5–40µl
Promega 200	200µl	P-200	EDP–250µl	40–200µl	40–200µl
Promega 1000 200–1,000µl	1,000µl	P-1,000		200–1,000µl	

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Biochemicals

Biochemicals 22

Acrylamide, Molecular Grade

Product	Size	Cat.#
Acrylamide, Molecular Grade	100 g	V3111
	500 g	V3115

Description: Acrylamide, Molecular Grade, is used for the electrophoretic separation of nucleic acids and proteins. Very small DNA fragments, such as those generated by sequencing reactions, can be resolved by polyacrylamide gel electrophoresis. Proteins can be separated by a variety of techniques, including denaturing gel electrophoresis using SDS or urea, isoelectric focusing and native gel electrophoresis in a wide variety of buffers.

Formula Weight: 71.08.

Form: White, free-flowing crystals.

Properties:
Purity: ≥99.9%.
Melting Point: 84–86°C.
Free Acrylic Acid: <0.001%
Iron: ≤1ppm.
Lead: ≤1ppm.
pH (10% in 0.1M NaCl at 25°C): 6.0–7.0.
Conductivity (40% in water): ≤2.5µmhos.

Features:
- **Quality Tested:** Each lot of Molecular Grade Acrylamide is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C. Protect from moisture.

Agarose, LE, Analytical Grade

Product	Size	Cat.#
Agarose, LE, Analytical Grade	100 g	V3121
	500 g	V3125

Description: Agarose, LE, Analytical Grade, is used for the electrophoretic separation of nucleic acids.

Form: White powder.

Properties:
Gel Strength (1%): ≥1,000g/cm^2.
Gelling Point (1.5%): 36–39°C.
Melting Point (1.5%): 87–89°C.
EEO (–mr): 0.09–0.13.
Sulfate: ≤0.14%.
Moisture: ≤7.0%.

Features:
- **Quality Tested:** Each lot of Analytical Grade LE Agarose is tested and certified to be free of DNase and RNase activity.

Storage Conditions: Store at 22–25°C.

Agarose, LMP, Preparative Grade for Large Fragments (>1,000bp)

Product	Size	Cat.#
Agarose, LMP, Preparative Grade for Large Fragments (>1,000bp)	25 g	V2831

Description: Agarose, LMP, Preparative Grade for Large Fragments (>1,000bp), is a premium agarose used for isolating DNA fragments larger than 1,000bp. Each lot is tested and certified for the following applications: 1) restriction digestion, 2) ligation and transformation, and 3) random prime labeling. LMP = low melting point (i.e., ≤65°C).

Form: White powder.

Properties:
Gelling Point (1.5%): 26–30°C.
Melting Point (1.5%): ≤65°C.
Sulfate: ≤0.10%.
EEO (–mr): ≤0.10.
Moisture: ≤10%.
Gel Strength (1%): ≥200g/cm^2.

Features:
- **Quality Tested:** Each lot of Preparative Grade LMP Agarose is tested and certified to be free of DNase and RNase activity.

Storage Conditions: Store at 22–25°C.

Agarose, LMP, Preparative Grade for Small Fragments (10 to 1,000bp)

Product	Size	Cat.#
Agarose, LMP, Preparative Grade for Small Fragments (10 to 1,000bp)	25 g	V3841

Description: Agarose, LMP, Preparative Grade for Small Fragments (10 to 1,000bp), is a premium agarose used for isolating DNA fragments from 10 to 1,000bp. The isolated DNA fragments can be used in various molecular biology applications: 1) restriction digestion, 2) ligation and transformation, 3) random prime labeling. LMP = low melting point (i.e., ≤65°C).

Form: White powder.

Properties:
Gelling Point (4%): ≤35°C.
Melting Point (4%): ≤65°C.
Sulfate: ≤0.15%.
EEO (–mr): ≤0.15.
Moisture: ≤10%.
Gel Strength: ≥500g/cm^2.

Features:
- **Quality Tested:** Each lot of Preparative Grade LMP Agarose is tested and certified to be free of DNase and RNase activity.

Storage Conditions: Store at 22–25°C.

Agarose, Low Melting Point, Analytical Grade

Product	Size	Cat.#
Agarose, Low Melting Point, Analytical Grade	25 g	V2111

Description: Agarose, Low Melting Point, Analytical Grade, is ideal for applications that require recovery of intact DNA fragments after gel electrophoresis.

Form: White powder.

Properties:
Gelling Point (1.5%): 24–28°C.
Melting Point (1.5%): ≤65.5°C.
Sulfate: ≤0.12%.
EEO (–mr): ≤0.11.
Gel Strength (1%): ≥300g/cm^2.

Features:
- **Quality Tested:** Each lot of Analytical Grade LMP Agarose is tested and certified to be free of DNase and RNase activity.

Storage Conditions: Store at 22–25°C.

Ammonium Persulfate, Molecular Grade

Product	Size	Cat.#
Ammonium Persulfate, Molecular Grade	25 g	V3131

Description: Ammonium Persulfate, Molecular Grade, is an oxidizing agent that promotes the polymerization of acrylamide gels.

Formula Weight: 228.20.

Form: White, free-flowing crystals.

Properties:
Purity: ≥98%.
Insolubles: ≤0.005%.
Chloride and Chlorate: ≤10ppm.
Lead: ≤50ppm.
Iron: ≤10ppm.
Manganese: ≤0.5ppm.
Residue After Ignition: ≤0.05%.
Moisture: ≤1.0%.
Titratable Free Acid: ≤0.04meq/g.

Features:
- **Quality Tested:** Each lot of Ammonium Persulfate is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C. Protect from moisture.

Ammonium Sulfate, Molecular Biology Grade

Product	Size	Cat.#
Ammonium Sulfate, Molecular Biology Grade	5 kg	H5252

Description: Ammonium Sulfate, Molecular Biology Grade, is a salt used in the purification of enzymes and other proteins by precipitation.

Formula Weight: 132.13.

Properties:
Purity: ≥99.0%.
Chloride: ≤5ppm.
Copper: ≤5ppm.
Iron: ≤5ppm.
Zinc: ≤5ppm.
Lead: ≤5ppm.
pH at 25°C (1M): 5.0–6.0.
A_{260} at 1M: ≤0.03.
A_{280} at 1M: ≤0.03.

Features:
- **Quality Tested:** Each lot of Ammonium Sulfate is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Antibiotic G-418 Sulfate

Product	Size	Cat.#
Antibiotic G-418 Sulfate	100 mg	V7981
	1 g	V7982
	5 g	V7983
Antibiotic G-418 Sulfate Solution	20 ml	V8091

Description: Antibiotic G-418 Sulfate is an aminoglycosidic antibiotic toxic to both prokaryotic and eukaryotic cells. It acts by interfering with protein synthesis and is used as an agent for selecting cultured cells expressing a gene (i.e., aminoglycoside 3´ phosphotransferase [APH 3´]) that confers resistance to G-418. The liquid form of the product is in distilled water.

Formula Weight: 692.6 (anhydrous).

Form: White powder.

Physical/Chemical Properties of Powder:
Appearance: White powder.
TLC: Single major spot.
Elemental Analysis: %C=28.8–36.07; %H=5.76–7.76; %N=6.72–8.41.
Absorbance: A_{280} (1mg/ml) = 0–0.015; A_{570} (100mg/ml) = 0–0.1.
Specific Rotation: +104° to +121°.

Properties Specific to V7981, V7982, V7983:
Appearance: White powder.
Hydration Waters: 0–6, as determined from Elemental Analysis.
Potency: ≥500µg/mg.

Properties Specific to V8091:
Potency: 40–60mg/ml.
Sterility: Aseptically filtered.

Features:
- **Sterile:** Antibiotic G-418 Sulfate Solution is quality tested for sterility.

Storage Conditions: Store powder at 22–25°C. Store liquid at –20°C.

BCIP/NBT Color Development Substrate (5-bromo-4-chloro-3-indolyl-phosphate/ nitro blue tetrazolium)

Product	Size	Cat.#
BCIP/NBT Color Development Substrate	1.25/2.5 ml	S3771

For Laboratory Use.

Description: BCIP (5-bromo-4-chloro-3-indolyl-phosphate) is used in conjunction with NBT (nitro blue tetrazolium) for the colorimetric detection of alkaline phosphatase activity. Each vial of BCIP is supplied with a vial of NBT.

Preparation of Substrates to Detect Alkaline Phosphatase: For every 5ml of alkaline phosphatase buffer (100mM Tris-HCl [pH 9.0], 150mM NaCl, 1mM $MgCl_2$), add 33µl NBT and 16.5µl BCIP. Add the NBT first, mix, add the BCIP, and mix again. Use within 1 hour, and discard any unused solution.

Concentration: BCIP (50mg/ml) in 100% dimethylformamide; NBT (50mg/ml) in 70% dimethylformamide.

Features:

- **Quality Tested:** Each lot of BCIP/NBT Color Development Substrate is tested and qualified for use in blotting.

Storage Conditions: Store at either 4°C or –20°C.

Bisacrylamide, Molecular Grade (N,N´-Methylenebisacrylamide)

Product	Size	Cat.#
Bisacrylamide, Molecular Grade	25 g	V3141
	125 g	V3143

Description: Bisacrylamide, Molecular Grade, is a cross-linking agent used in the preparation of polyacrylamide gels. This product is tested for its efficiency in gel polymerization.

Formula Weight: 154.20.

Form: White, free-flowing crystals.

Properties:
Purity: ≥99.0%.
Acrylic Acid (CH_2:CHCOOH): ≤0.001%.
A_{290} (1% solution): ≤0.20.
Magnesium: ≤2ppm.
Conductivity (2% in water): ≤10µmhos.

Features:

- **Quality Tested:** Each lot of Bisacrylamide is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Boric Acid, Molecular Biology Grade (orthoboric acid)

Product	Size	Cat.#
Boric Acid, Molecular Biology Grade	500 g	H5001
	1 kg	H5003

Description: Boric Acid, Molecular Biology Grade, in conjunction with Tris, is commonly used in buffers for the preparation of agarose or acrylamide gels and their associated running buffers.

Formula Weight: 61.84.

Properties:
Purity: ≥99.5%.
Iron: ≤5ppm.
Lead: ≤5ppm.
Moisture: ≤0.5%.
A_{260} at 1M: ≤0.015.
A_{280} at 1M: ≤0.010.

Features:

- **Quality Tested:** Each lot of Boric Acid is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Bovine Serum Albumin, Acetylated

Product	Size	Conc.	Cat.#
Bovine Serum Albumin, Acetylated	1 ml	10 mg/ml	R3961
	400 µl	1 µg/µl	R9461

For Laboratory Use.

Description: Bovine Serum Albumin, Acetylated, can be used as an enzyme stabilizer or as a carrier protein. It is prepared by a modification of the method of Gonzalez *et al.* and dialyzed extensively with deionized water to remove impurities.

Features:

- **Quality Tested:** Each lot of BSA is tested and certified to be free of DNase and RNase activity.

Storage Conditions: Store at –20°C.

DTT, Molecular Grade (DL-Dithiothreitol)

Product	Size	Conc.	Cat.#
DTT, Molecular Grade	100 µl	100 mM	P1171
DTT, Molecular Grade (Dry Powder)	5 g		V3151
	25 g		V3155

Cat.# P1171 For Laboratory Use.

Description: DTT, Molecular Grade, is an antioxidant used to stabilize enzymes and other proteins containing sulfhydryl groups. The liquid form of the product is a 100mM solution of DTT in water.

Formula: $C_4H_{10}O_2S_2$.

Formula Weight: 154.25.

Form: White crystals/powder or liquid in deionized water.

Properties:
Purity: ≥99.0%.
Melting Point: 40–44°C.
A_{283} at 20mM: ≤0.04.
% Oxidized: ≤0.50%.

Features:
- **Quality Tested:** Each lot of DTT is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at –20°C.

EDTA, 0.5M (pH 8.0), Molecular Biology Grade

Product	Size	Cat.#
EDTA, 0.5M (pH 8.0), Molecular Biology Grade	100 ml	V4231
	400 ml	V4233

Description: EDTA, 0.5M (pH 8.0), Molecular Biology Grade, is a chelator of divalent cations and is suitable for biochemistry and molecular biology applications. It is supplied as a solution in deionized water.

Form: Clear, colorless liquid.

Properties:
pH at 25°C: 7.9–8.1.
A_{280} at 0.5M: ≤0.25.
RNase Activity at 0.5M: ≤1.0% release of ^{3}H-RNA.
DNase Activity at 0.5M: ≤1.0% release of ^{3}H-DNA.
Protease Assay: None detected.

Storage Conditions: Store at 22–25°C.

EDTA, Disodium Salt (Dihydrate), Molecular Biology Grade

Product	Size	Cat.#
EDTA, Disodium Salt, Molecular Biology Grade	100 g	H5031
	500 g	H5032

Description: EDTA, Disodium Salt, Molecular Biology Grade, is a chelator of divalent metal cations.

Formula Weight: 372.20.

Properties:
Purity: ≥99.0%.
Insolubles: ≤0.005%.
Lead: ≤5ppm.
Iron: ≤10ppm.

Features:
- **Quality Tested:** Each lot of EDTA is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Ethidium Bromide Solution, Molecular Grade

Product	Size	Conc.	Cat.#
Ethidium Bromide Solution, Molecular Grade	10 ml	10 mg/ml	H5041

Description: Ethidium Bromide Solution, Molecular Grade (10mg/ml), is a fluorescent dye suitable for staining nucleic acids after electrophoresis or in cesium chloride gradients. The solution can be used to detect both double-stranded and single-stranded DNA.

Features:
- **Quality Tested:** Each lot of Ethidium Bromide Solution is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Formamide, Molecular Grade

Product	Size	Cat.#
Formamide, Molecular Grade	100 ml	H5051
	500 ml	H5052

Description: Formamide is often used for the denaturation of nucleic acids in applications such as hybridization, sequencing gel electrophoresis and electron microscopy.

Formula Weight: 45.04.

Properties:
Purity: ≥99.5%.
Copper: ≤1ppm.
Iron: ≤1ppm.
Lead: ≤1ppm.
Zinc: ≤1ppm.
Refractive Index at 20°C: 1.446–1.448.
pH at 25°C of 1%: 6.5–7.5.
A_{260} at 10%: ≤0.10.
A_{280} at 10%: ≤0.02.

Features:
- **Quality Tested:** Each lot of Formamide is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Glycerol, Molecular Biology Grade

Product	Size	Cat.#
Glycerol, Molecular Biology Grade	1,000 ml	H5433

Description: Glycerol is used for storage of enzymes at low temperatures. A 50% (w/v) glycerol solution will not freeze at –20°C. Glycerol is often used as a component in electrophoresis loading buffers because of its density (1.26g/ml). In addition, glycerol gradients can be used in the purification of bacteriophage or proteins. Cat.# H5433 is anhydrous glycerol with a purity of ≥99.5%.

Properties:
Purity: ≥99.5%.
Calcium: ≤2ppm.
Magnesium: ≤1ppm.
Lead: ≤5ppm.
Zinc: ≤1ppm.
A_{260} at 10%: ≤0.05.
A_{280} at 10%: ≤0.05.

Features:
- **Quality Tested:** Each lot of glycerol is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Glycine, Molecular Biology Grade

Product	Size	Cat.#
Glycine, Molecular Biology Grade	500 g	H5071
	1 kg	H5073

Description: Glycine is an amino acid used in the preparation of some electrophoresis buffers.

Formula Weight: 75.07.

Properties:
Purity: ≥99.0%.
Iron: ≤10ppm.
A_{260} at 1M: ≤0.05.
A_{280} at 1M: ≤0.05.

Features:
- **Quality Tested:** Each lot of Glycine is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Guanidine Thiocyanate, Molecular Grade (Guanidinium Thiocyanate)

Product	Size	Cat.#
Guanidine Thiocyanate, Molecular Grade	100 g	V2791

Description: Guanidine Thiocyanate, Molecular Grade, at high concentrations, is a protein denaturant used most commonly for the isolation of intact RNA due to its ability to inhibit RNase.

Formula Weight: 118.16.

Form: White, crystalline powder.

Properties:
Purity: ≥99.0%.
Insolubles: None.
A_{280} at 6M: ≤0.8.
A_{300} at 6M: ≤0.1.
A_{320} at 6M: ≤0.1.
A_{410} at 6M: ≤0.1.
Moisture: ≤1%.
Melting Point: 118–121°C.
Potassium: ≤50ppm.
Sodium: ≤0.5%.
Zinc: ≤1.5ppm.
Copper: ≤0.5ppm.
Barium: ≤3ppm.
Iron: ≤5ppm.

Features:
- **Quality Tested:** Each lot of Guanidine Thiocyanate is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Guanidine-HCl, Molecular Biology Grade (Guanidinium Hydrochloride)

Product	Size	Cat.#
Guanidine-HCl, Molecular Biology Grade	100 g	H5381
	500 g	H5383

Description: Guanidine-HCl, Molecular Grade, is commonly used for the isolation of intact mRNA from tissues or cultured cells.

Formula Weight: 95.53.

Form: Fine, colorless or white crystals.

Properties:
Purity: ≥99.5%.
A_{230} at 6M: ≤0.15.
A_{260} at 6M: ≤0.03.
A_{280} at 6M: ≤0.02.
Moisture: ≤0.3%.
Melting Point: 186–188°C.
Lead: ≤5ppm.
Zinc: ≤1ppm.
Copper: ≤1ppm.
Iron: ≤5ppm.

Features:
- **Quality Tested:** Each lot of Guanidine-HCl is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

HEPES, Molecular Biology Grade (free acid)

Product	Size	Cat.#
HEPES, Molecular Biology Grade (free acid)	100 g	H5302
	500 g	H5303

Description: HEPES is a biological buffer that functions over a pH range of 6.8 to 8.2.

Formula Weight: 238.3.

Properties:
Appearance: White, crystalline powder.
Purity: ≥99.5%.
Lead: ≤5ppm.
Iron: ≤5ppm.
Moisture: ≤0.5%.
pH at 25°C (1M): 5.0–6.5.
A_{260} at 0.1M: ≤0.05.
A_{280} at 0.1M: ≤0.04.

Features:
- **Quality Tested:** Each lot of HEPES is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

IPTG, Dioxane-Free

Product	Size	Cat.#
IPTG, Dioxane-Free	1 g	V3955
	5 g	V3951
	50 g	V3953

Cat.# V3951, V3955 For Laboratory Use.

Description: IPTG, Dioxane-Free (isopropyl-β-D-thiogalactopyranoside), is an inducer of β-galactosidase activity in many bacteria. Functioning as a *lac* analog, IPTG induces β-galactosidase activity by binding to and inhibiting the *lac* repressor. This product is used to differentiate recombinants from nonrecombinants in cloning strategies using vectors containing the *lacZ* or *lacZ* α-peptide gene.

Formula Weight: 238.31.

Form: White powder.

Properties:
Purity: ≥99.0%.
Moisture: ≤1%.
pH (5%, H_2O): 5–7.
Dioxane Content: ≤10ppm.

Storage Conditions: Store dry at 4°C or –20°C.

Luciferin-EF™ Endotoxin-Free Luciferin Na

Product	Size	Cat.#
Luciferin-EF™	25 mg	E6551
	250 mg	E6552

Description: Luciferin-EF™ is an endotoxin-free beetle luciferin that can be used for cell-based imaging applications in living systems, where endotoxin may create problems. Luciferin-EF™ is tested to ensure endotoxin is below detectable levels and packaged in amber vials with septa to facilitate easy dilution and use.

Features:
- **Achieve Endotoxin Levels Below Detection Limits:** No potential interference in assay due to the presence of endotoxins.
- **Be Assured of Product Integrity:** Luciferin-EF™ is packaged in amber vials with septa to ensure product integrity as well as offer ease of dilution and use for imaging experiments.
- **Appreciate Flexibility and Convenience:** Luciferin-EF™ is available in two sizes, depending on the number of experiments to be performed.

Storage Conditions: Store at –70°C.

Nuclease-Free Water

Product	Size	Cat.#
Nuclease-Free Water	50ml (2 × 25 ml)	P1193
	150 ml	P1195

For Laboratory Use.

Description: Nuclease-Free Water is an essential component of molecular biology experiments.

Features:
- **Quality Tested:** Each lot of Nuclease-Free Water is tested and certified to be free of DNase and RNase activity.

Storage Conditions: Store at <30°C.

PEG 8000, Molecular Biology Grade (Polyethylene Glycol 8000)

Product	Size	Cat.#
PEG 8000 Powder, Molecular Biology Grade	500 g	V3011

Description: PEG 8000 is used in the precipitation of phage, isolation of plasmid DNA and the enhancement of blunt-ended ligation reactions.

Formula Weight: 7,000–9,000.

Form: White, waxy crystalline flakes.

Properties:
Purity: ≥99.0%.
pH at 25°C (5% water): 5.0–7.0.

Features:
- **Quality Tested:** Each lot of PEG 8000 is tested and certified to be free of DNase and RNase activity.

Storage Conditions: Store at 22–25°C.

SDS Solution, Molecular Biology Grade (10% w/v)

Product	Size	Cat.#
SDS Solution, Molecular Biology Grade (10% w/v)	100 ml	V6551
	500 ml	V6553

Description: SDS Solution (10% w/v) is sodium dodecyl sulphate in distilled, deionized water. SDS is a detergent that is known to denature proteins. It is used in polyacrylamide gel electrophoresis for the determination of protein molecular weight. It is also used in nucleic acid extraction procedures for the disruption of cell walls and dissociation of nucleic acid:protein complexes.

Properties:
A_{260}: ≤0.3.
A_{280}: ≤0.2.

Features:
- **Quality Tested:** Each lot of SDS Solution is tested and certified to be free of DNase and RNase activity.

Storage Conditions: Store at 22–25°C.

Sephacryl® S-400

Product	Size	Cat.#
Sephacryl® S-400	10 ml	V3181
For Laboratory Use.		

Description: Sephacryl® S-400 is a chromatography matrix used for rapid gel filtration. This matrix is useful in experiments involving the incorporation of synthetic linkers and adaptors. After linker ligation and digestion with the appropriate enzyme, unincorporated linkers and linker fragments may be rapidly removed from the DNA sample using spin columns containing Sephacryl® S-400. Such columns may be used to separate small DNA fragments (≤271bp) from longer DNA molecules.

Features:
- **Quality Tested:** Each lot is tested and certified to be free of DNase and RNase activity.
- **Composition:** Suspension in 10mM Tris-HCl (pH 8.0), 100mM NaCl and 1mM EDTA.

Storage Conditions: Store at 4°C.

5M Sodium Chloride, Molecular Biology Grade

Product	Size	Conc.	Cat.#
5M Sodium Chloride, Molecular Biology Grade	1 L	5 M	V4221

Description: 5M Sodium Chloride is commonly used in many molecular biology and forensic applications.

Form: Clear, colorless liquid.

Properties:
pH at 25°C (1M): 5.0–8.0.
A_{260} at 5M: ≤0.02.
A_{280} at 5M: ≤0.01.
Conductivity at 25°C (0.05M): 5,000–7,000µSm.

Features:
- **Quality Tested:** Each lot of NaCl is tested and certified to be free of DNase, RNase and protease activity.
- **Composition:** 292.2g/L NaCl in deionized water.

Storage Conditions: Store at 22–25°C.

Sodium Chloride, Molecular Biology Grade

Product	Size	Cat.#
Sodium Chloride, Molecular Biology Grade	500 g	H5271
	1 kg	H5273

Description: Sodium Chloride, Molecular Biology Grade, is commonly used in many molecular biology and forensic applications.

Formula Weight: 58.45.

Properties:
Purity: ≥99.5%.
Iron: ≤2ppm.
Lead: ≤5ppm.
pH at 25°C of 1M: 5.0–8.0.
A_{260} at 1M: ≤0.02.
A_{280} at 1M: ≤0.01.
Conductivity at 25°C (0.05M): 5,000–7,000µSm.

Features:
- **Quality Tested:** Each lot of Sodium Chloride is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Sodium Dodecyl Sulphate, Molecular Biology Grade (SDS)

Product	Size	Cat.#
Sodium Dodecyl Sulphate, Molecular Biology Grade (SDS)	100 g	H5113
	500 g	H5114
	1 kg	H5115

Description: Sodium Dodecyl Sulphate, Molecular Biology Grade (SDS), is a detergent that is known to denature proteins. It is used in denaturing polyacrylamide gel electrophoresis for the determination of protein molecular weight. It is also used in nucleic acid extraction procedures for the disruption of cell walls and dissociation of nucleic acid:protein complexes.

Formula Weight: 288.38.

Properties:
Purity: ≥99.5%.
pH at 25°C (3% w/v): 6.0–7.5.
A_{230} at 3%: ≤0.40.
A_{260} at 3%: ≤0.30.
A_{280} at 3%: ≤0.05.
A_{405} at 3%: ≤0.01.

Features:
- **Quality Tested:** Each lot is tested and certified to be free of DNase and RNase activity.

Storage Conditions: Store at 22–25°C.

SSC Buffer, 20X, Molecular Grade

Product	Size	Cat.#
SSC Buffer, 20X, Molecular Grade	1,000 ml	V4261

Description: SSC Buffer, 20X, Molecular Grade (pH 7.0), is commonly used in nucleic acid hybridization techniques at concentrations from 0.1X to 20X, depending on the application.

Form: Clear, colorless liquid.

Properties:
pH at 25°C (20X): 6.9–7.1.
Lead: ≤10ppm.
Conductivity at 25°C (2X): 24.4–32.4mmhos.

Features:
- **Quality Tested:** Each lot of SSC Buffer is tested and certified to be free of DNase, RNase and protease activity.
- **Composition:** 3M NaCl, 0.3M sodium citrate (for 20X concentration).

Storage Conditions: Store at 22–25°C.

Streptavidin Alkaline Phosphatase

Product	Size	Cat.#
Streptavidin Alkaline Phosphatase	0.5 ml	V5591
For Laboratory Use.		

Description: Streptavidin Alkaline Phosphatase is used for the detection of biotinylated molecules.

Features:
- **Composition:** Conjugated Streptavidin Alkaline Phosphatase in PBS, 1mg/ml BSA, 1mM $MgCl_2$, 0.1mM $ZnCl_2$ and 0.02% sodium azide.
- **Quality Tested:** Streptavidin Alkaline Phosphatase is quality tested to ensure optimal performance for the detection of biotinylated molecules.

Storage Conditions: Store at 4°C. **Do not freeze!**

TAE Buffer, Molecular Biology Grade (Tris-acetate-EDTA)

Product	Size	Conc.	Cat.#
TAE Buffer, 10X, Molecular Biology Grade	1,000 ml	10X	V4271
TAE Buffer, 40X, Molecular Biology Grade	1,000 ml	40X	V4281

Description: TAE Buffer is the most commonly used buffer for agarose DNA electrophoresis. A 1X solution is obtained by adding 1 part of the concentrated TAE to 9 or 39 parts of deionized water.

Form: Clear, colorless liquid.

Properties:
Composition (10X): 400mM Tris-acetate, 10mM EDTA.
Composition (40X): 1.6M Tris-acetate, 40mM EDTA.
pH at 25°C: 8.2–8.4.
Lead: ≤10ppm.

Features:
- **Quality Tested:** Each lot of TAE Buffer is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

TBE Buffer, 10X, Molecular Biology Grade

Product	Size	Cat.#
TBE Buffer, 10X, Molecular Biology Grade	1,000 ml	V4251

Description: TBE Buffer, 10X (pH 8.3), is used for polyacrylamide and agarose gel electrophoresis. This product has been optimized for use in DNA applications.

Form: Clear, colorless liquid.

Properties:
pH at 25°C (1X): 8.2–8.4.

Features:
- **Quality Tested for DNase Activity:** Each lot of TBE Buffer is tested and demonstrates ≤1% release.
- **Quality Tested for RNase Activity:** Each lot of TBE Buffer is tested and demonstrates ≤1% release.
- **Composition:** 890mM Tris-borate, 890mM boric acid, 20mM EDTA.

Storage Conditions: Store at 22–25°C.

TE Buffer, 1X, Molecular Biology Grade

Product	Size	Cat.#
TE Buffer, 1X, Molecular Biology Grade	100 ml	V6231
	500 ml	V6232

Description: TE Buffer, 1X, Molecular Grade (pH 8.0), is a buffer composed of 10mM Tris-HCl containing 1mM EDTA•Na_2.

Properties:
pH at 25°C: 7.9–8.1.
A_{280}: ≤0.05.

Features:
- **Quality Tested:** Each lot of TE Buffer is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

TMB Stabilized Substrate for Horseradish Peroxidase

Product	Size	Cat.#
TMB Stabilized Substrate for Horseradish Peroxidase	200 ml	W4121

Description: TMB Stabilized Substrate is a stable, ready-to-use TMB (3,3´, 5,5´-tetramethylbenzidine) color development substrate for localization of horseradish peroxidase-conjugated antibodies on dot blots and Western blots. It is easier to use than 4-chloro-1-naphthol (CN), which must be prepared immediately before use. TMB Stabilized Substrate comes premixed and fully diluted in a proprietary buffer containing less than 0.5% organic solvent.

Features:
- **Convenient:** Premixed, ready-to-use; in proprietary buffer containing less than 0.5% organic solvents.
- **Stable:** Stable at room temperature for 12 months.
- **Sensitive:** At least threefold more sensitive than 4-chloro-1-naphthol (CN); as little as 412pg of β-galactosidase detected on TMB blot as compared to 1.12ng on CN blot when detected with a β-galactosidase-specific antibody and HRP-conjugated secondary antibody.
- **Long-Lasting Color:** Color is much more stable than 4-chloro-1-naphthol and photographs more easily.

Storage Conditions: Store at 22–25°C.

Tris Base, Molecular Biology Grade

Product	Size	Cat.#
Tris Base, Molecular Biology Grade	100 g	H5133
	500 g	H5131
	2,500 g	H5135

Description: Tris Base, Molecular Biology Grade, is commonly used for many molecular biology applications.

Formula: $C_4H_{11}NO_3$.

Formula Weight: 121.14.

Form: Crystalized free base.

Properties:
pH at 25°C of 1M: 10.0–11.5.
Purity: ≥99.9%.
A_{260} at 1M: ≤0.05.
A_{280} at 1M: ≤0.05.
Melting Point: 167–172°C.
Moisture: ≤0.2%.
Lead: ≤2ppm.
Magnesium: ≤1ppm.
Calcium: ≤1ppm.
Iron: ≤1ppm.

Features:
- **Quality Tested:** Each lot of Tris Base is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Tris-HCl, Molecular Biology Grade (Tris-Hydrochloride)

Product	Size	Cat.#
Tris-HCl, Molecular Biology Grade	100 g	H5121
	500 g	H5123
	2,500 g	H5125

Description: Tris-HCl, Molecular Biology Grade, is sometimes used in combination with Tris base for preparation of Tris-HCl buffers.

Formula Weight: 157.56.

Properties:
pH at 25°C (0.1M): 4.2–5.0.
Purity: ≥99.0%.
A_{240} at 1M: ≤0.06.
A_{260}, A_{280}, A_{300}, A_{600} at 1M: ≤0.05.
Melting Point: 150–152°C.
Moisture: ≤0.5%.
Calcium: ≤5ppm.
Iron: ≤5ppm.
Lead: ≤1ppm.
Magnesium: ≤1ppm.
Manganese: ≤1ppm.
Copper: ≤1ppm.
Zinc: ≤1ppm.

Features:
- **Quality Tested:** Each lot of Tris-HCl is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Triton® X-100, Molecular Biology Grade

Product	Size	Cat.#
Triton® X-100, Molecular Biology Grade	100 ml	H5142
	500 ml	H5141

Description: Triton® X-100, Molecular Biology Grade, is a widely used nonionic surfactant.

Properties:
Moisture: ≤1.0%.
Lead: ≤5ppm.
Iron: ≤5ppm.
Density at 25°C: 1.0645–1.0655g/ml.

Features:
- **Quality Tested:** Each lot of Triton® X-100 is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Tween® 20, Molecular Biology Grade

Product	Size	Cat.#
Tween® 20, Molecular Biology Grade	100 ml	H5152
	500 ml	H5151

Description: Tween® 20, Molecular Biology Grade, is a nonionic detergent used for many different molecular biology applications.

Properties:
Appearance: Clear, yellow, viscous liquid.
Hydroxyl Number: 96–108.
Lead: ≤10ppm.

Features:
- **Quality Tested:** Each lot of Tween® 20 is tested and certified to be free of DNase, RNase and protease activity.

Storage Conditions: Store at 22–25°C.

Urea

Product	Size	Cat.#
Urea	1 kg	V3171
	5 kg	V3175

Description: Urea is a protein denaturant. Urea is qualified for use as the denaturing component in polyacrylamide gels.

Formula: $(NH_2)_2CO$.

Formula Weight: 60.06.

Form: Fine, white, free-flowing pastilles.

Properties:
Purity: ≥99.0%.
Melting Point: 132–135°C.
A_{280} at 8M in water: ≤0.10.
Chloride: ≤0.0005%.
Heavy Metals: ≤0.001%.
Iron: ≤0.001%.
Cyanate: none detected.

Storage Conditions: Store at 22–25°C. Protect from moisture.

Western Blue® Stabilized Substrate for Alkaline Phosphatase

Product	Size	Cat.#
Western Blue® Stabilized Substrate for Alkaline Phosphatase	100 ml	S3841

Description: Western Blue® Stabilized Substrate for Alkaline Phosphatase is a stable, ready-to-use substrate for Western blots and immunoscreening. Western Blue® Substrate should be used directly and without dilution. It is a mixture of 5-bromo-4-chloro-3-indolyl-phosphate (BCIP) and nitro blue tetrazolium (NBT) in a proprietary stabilizing buffer. This liquid substrate deposits a permanent dark purple stain on membrane sites bearing alkaline phosphatase. Western Blue® Substrate is as sensitive as BCIP/NBT reagents.

Features:

- **Convenient:** Use directly without diluting or mixing.
- **Sensitive:** Substrate is as sensitive as other BCIP/NBT reagents.
- **Stable:** Stable for one year at room temperature.

Storage Conditions: Store at 22–25°C.

X-Gal (5-bromo-4-chloro-3-indolyl-β-D-galactopyranoside)

Product	Size	Conc.	Cat.#
X-Gal	100mg/2 ml	50 mg/ml	V3941
For Laboratory Use.			

Description: X-Gal, in conjunction with IPTG, is used to detect β-galactosidase activity to differentiate recombinants from nonrecombinants in cloning experiments using vectors containing the *lac*Z or *lac*Z α-peptide gene.

Features:

- **Concentration:** 50mg/ml in dimethylformamide, 2.0ml/vial.
- **Quality Tested:** X-Gal is tested for use with the pGEM®-Z Vectors in a chromogenicity assay.

Storage Conditions: Store at 4°C or –20°C.

Cell Line Authentication

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Section Contents

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StemElite™ ID System

Product	Size	Cat.#
StemElite™ ID System	50 reactions	G9530

Description: The StemElite™ ID System provides the reagents required for successful and simple identification and authentication of human cell lines and detection of intra-species cell line cross-contamination. The StemElite™ ID System uses short tandem repeat (STR) analysis of specific loci in the human genome through co-amplification and three-color detection of ten loci (nine STR loci and Amelogenin for gender identification), including D21S11, TH01, TPOX, vWA, Amelogenin, CSF1PO, D16S539, D7S820, D13S317 and D5S818. Additionally, the StemElite™ ID System incorporates a sensitive marker that specifically detects the presence of DNA from mouse (*Mus musculus*). All eleven loci are amplified simultaneously in a single tube. The StemElite™ ID System includes a hot-start *Taq* DNA polymerase system in an enzyme mix for convenient room-temperature reaction assembly. Alleles, along with the allelic ladder and an internal lane standard that assists in defining sizes for each of the loci, are resolved by capillary electrophoresis in a single injection, and CE data are analyzed using allele-calling software to generate a genetic profile of the cell line. These loci collectively provide a genetic profile with a random match probability of 1 in 2.92×10^9.

The StemElite™ ID System is designed specifically for use with the Applied Biosystems 3130 and 3130*xl* Genetic Analyzers, the ABI PRISM® 3100 and 3100-*Avant* Genetic Analyzers and the ABI PRISM® 310 Genetic Analyzer.

Dye matrix generation is required on initial setup and is performed using the PowerPlex® Matrix Standards, 310 (Cat.# DG4640) or PowerPlex® Matrix Standards, 3100/3130 (Cat.# DG4650).

An overview of the StemElite™ ID System protocol.

DNA Sample Preparation

↓

Amplification Setup

↓

Thermal Cycling
GeneAmp® PCR System 9700
GeneAmp® PCR System 9600

↓

Instrument Setup and Sample Preparation
Applied Biosystems 3130 or 3130xl Genetic Analyzer
ABI PRISM® 3100 or 3100-*Avant* Genetic Analyzer with Data Collection Software, Version 2.0
ABI PRISM® 310 Genetic Analyzer

↓

Data Analysis
GeneMapper® ID Software, Versions 3.1 and 3.2GeneMapper® Software, Versions 3.7 and 4.0

9581MA

Features:

- **Improved "Gold Standard" STR Analysis:** The StemElite™ ID System includes all loci used in the original PowerPlex® 1.2 System plus the D21S11 loci for improved power of discrimination.
- **Detects Mouse Contamination:** The StemElite™ ID System includes a mouse locus allowing detection of contamination of human cells with mouse cells.
- **Simplified Data Interpretation:** The StemElite™ ID System includes 10 loci to accurately discriminate between human cell lines without using a full forensic profile and includes Allelic Ladders for all 10 loci to simplify data interpretation.
- **Simplified Amplification Setup:** Inclusion of a hot-start *Taq* DNA polymerase system allows room temperature reaction assembly.
- **Internal Lane Standard 600 Included:** This marker offers the greatest precision available for DNA typing. It is designed for use in each capillary injection to increase the precision of your analyses.
- **Automatic Assignment of Genotypes:** To make genotyping easier and more accurate, panel and bin files are available for download at: **www.promega.com/stemeliteid/** and are required for use with GeneMapper® *ID* software. GeneMapper® 4.0 software requires custom bin generation, and procedures can be obtained at: **www.promega.com/stemeliteid/**

Protocol	Part#
Technical Manual	TM307

Storage Conditions: Store at –20°C. The Primer Pair Mix, Allelic Ladder and Internal Lane Standard are light-sensitive and should be stored protected from light.

Cell ID™ System

Product	Size	Cat.#
Cell ID™ System	50 reactions	G9500

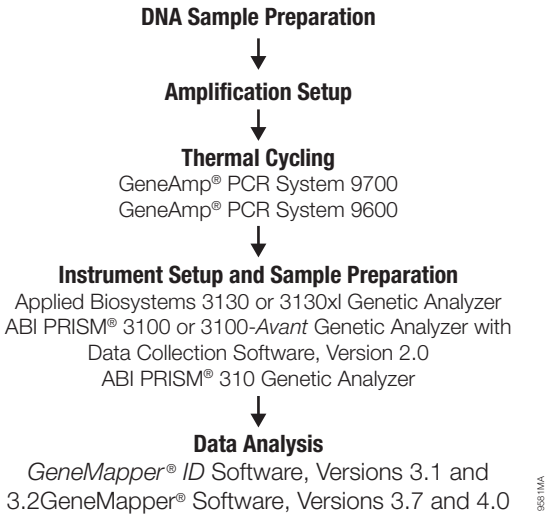
For Research Use Only. Not for use in diagnostic procedures.

Description: The Cell ID™ System provides the reagents required for successful and simple identification and authentication of human cell lines and detection of intra-species cell line cross-contamination. The Cell ID™ System uses short tandem repeat (STR) analysis of specific loci in the human genome through co-amplification and three-color detection of ten loci (nine STR loci and Amelogenin for gender identification), including D21S11, TH01, TPOX, vWA, Amelogenin, CSF1PO, D16S539, D7S820, D13S317 and D5S818. All ten loci are amplified simultaneously in a single tube. The Cell ID™ System includes a hot-start *Taq* DNA polymerase system in an enzyme mix for convenient room-temperature reaction assembly. Alleles, along with the allelic ladder and an internal lane standard that assists in defining sizes for each of the loci, are resolved by capillary electrophoresis in a single injection, and CE data are analyzed using allele-calling software to generate a genetic profile of the cell line. These loci collectively provide a genetic profile with a random match probability of 1 in 2.92×10^9.

The Cell ID™ System is designed specifically for use with the Applied Biosystems 3130 and 3130*xl* Genetic Analyzers, the ABI PRISM® 3100 and 3100-*Avant* Genetic Analyzers and the ABI PRISM® 310 Genetic Analyzer.

Dye matrix generation is required on initial setup and is performed using the PowerPlex® Matrix Standards, 310 (Cat.# DG4640) or PowerPlex® Matrix Standards, 3100/3130 (Cat.# DG4650).

An overview of the Cell ID™ System protocol.



Features:

- **Improved "Gold Standard" STR Analysis:** The Cell ID™ System includes all loci used in the original PowerPlex® 1.2 System plus the D21S11 loci for improved power of discrimination.
- **Simplified Data Interpretation:** The Cell ID™ System includes 10 loci to accurately discriminate between human cell lines without using a full forensic profile and includes Allelic Ladders for all 10 loci to simplify data interpretation.
- **Simplified Amplification Setup:** Inclusion of a hot-start *Taq* DNA polymerase system allows room temperature reaction assembly.
- **Internal Lane Standard 600 Included:** This marker offers the greatest precision available for DNA typing. It is designed for use in each capillary injection to increase the precision of your analyses.
- **Automatic Assignment of Genotypes:** To make genotyping easier and more accurate, panel and bin files are available for download at: **www.promega.com/cellidapps/** and are required for use with GeneMapper® *ID* software. GeneMapper® 4.0 software requires custom bin generation, and procedures can be obtained at: **www.promega.com/cellidapps/**

Protocol	Part#
Technical Manual	TM074

Storage Conditions: Store at –20°C. The Primer Pair Mix, Allelic Ladder and Internal Lane Standard are light-sensitive and should be stored protected from light.

personal
DETECTION

Promega

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Cell Signaling

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For information regarding cell signaling assays that are based on reporter gene activity, please refer to Chapter 8, Genetic Reporters and Transfection Systems.

cAMP-Glo™ Assay

Product	Size	Cat.#
cAMP-Glo™ Assay	300 assays (384-well plate)	V1501
	3,000 assays (384-well plate)	V1502
	30,000 assays (384-well plate)	V1503

For Research Use Only. Not for use in diagnostic procedures. Cat.# V1501 contains sufficient reagents to perform 300 assays at 60µl final volume per well in 384-well plates. Cat.# V1502 contains sufficient reagents to perform 3,000 assays at 60µl final volume per well in 384-well plates. Cat.# V1503 contains sufficient reagents to perform 30,000 assays at 60µl final volume per well in 384-well plates.

Description: The cAMP-Glo™ Assay is a homogeneous, bioluminescent and high-throughput assay for measuring cAMP levels in cells. The cAMP-Glo™ Assay monitors cAMP production in cells in response to the effects of test compounds on G protein-coupled receptors (GPCR). GPCRs that couple with adenylate cyclase will increase or decrease intracellular cAMP. The assay is based on the principle that cyclic AMP (cAMP) stimulates protein kinase A (PKA) holoenzyme activity, decreasing available ATP and leading to decreased light production in a coupled luciferase reaction.

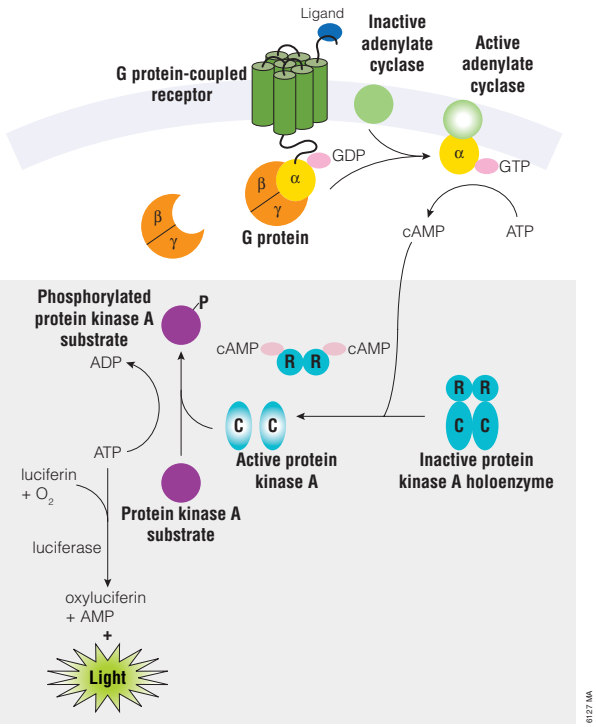
The cAMP-Glo™ Assay can be performed in 96-, 384- or 1536-well plates. The cells are induced with a test compound for an appropriate period of time to modulate cAMP levels. After induction, cells are lysed to release cAMP, then the cAMP detection solution, which contains protein kinase A, is added. The Kinase-Glo® Reagent is then added to terminate the PKA reaction and detect the remaining ATP via a luciferase reaction. Plates are read using a microplate-reading luminometer. Luminescence can be correlated to the cAMP concentrations by using a cAMP standard curve. The half-life for the luminescent signal is greater than 4 hours. This extended signal half-life eliminates the need for luminometers with reagent injectors and allows batch-mode processing of multiple plates.

Features:

- **Fast and Easy to Use**
  - Assay can be completed in approximately 45 minutes.
  - Homogeneous.
  - Two steps following lysis of cells.
- **Excellent Signal-to-Noise Ratios**
  - Best signal:background ratio of all the cAMP assays.
  - Signal:Background >200 (with cAMP), >15 (on cells).
  - Easily scalable to 1536-well plate formats and beyond.
- **Proven Luminescent Technology**
  - Powered by Ultra-Glo™ Recombinant Luciferase.
  - No interference by fluorescent compounds.
  - Non-radioactive.

Protocol	Part#
Technical Bulletin	TB357

Storage Conditions: Store the system at –20°C. Once prepared, the cAMP detection solution (cAMP-Glo™ Reaction Buffer with Protein Kinase A) should not be frozen. Once prepared, the Kinase-Glo® Reagent should be dispensed into aliquots and stored at –20°C. See the product label for the expiration date.



Schematic diagram of cAMP production in cells and the cAMP-Glo™ Assay.

GloSensor™ cAMP Assay

Product	Size	Cat.#
GloSensor™ cAMP HEK293 Cell Line	2 vials	E1261
pGloSensor™-22F cAMP Plasmid	20 µg	E2301
pGloSensor™-20F cAMP Plasmid	20 µg	E1171
GloSensor™ cAMP Reagent	25 mg	E1290
	250 mg	E1291

Description: The GloSensor™ cAMP Assay presents a novel approach to measuring cAMP levels in live cells. cAMP is a key second messenger involved in signal transduction of GPCRs acting through Gα-s and Gα-i proteins. The new assay is based on the GloSensor™ Technology, a genetically modified form of firefly luciferase into which a cAMP-binding protein moiety has been inserted. Upon binding of cAMP, conformational change is induced leading to increased light output. This live-cell assay excels at kinetic and modulation studies of signaling through cAMP.

Researchers can use the GloSensor™ cAMP Assay by transiently expressing a receptor of interest and the biosensor in the cell line of choice. Alternatively, stably transfected cell lines with both the biosensor and the receptor of interest can be made. The protocol is simple: Cells are pre-equilibrated with GloSensor™ cAMP Reagent for approximately 2 hours; then cells are treated with specific agonists/antagonists or compounds, and luminescence is measured after 10–30 minutes. No other reagent additions or manipulations are required. Most any common luminometer with injectors is sufficient to read the assay. GloSensor™ cAMP Reagent is required for use with this assay per the GloSensor™ Limited Use Label License.

Choosing the Appropriate Plasmid

We offer two variants of the biosensor, and we recommend the pGloSensor™-22F cAMP Plasmid as the first choice for most applications.

pGloSensor™-22F cAMP Plasmid. Following cell-free expression in vitro, the version encoded by this construct shows an increased EC_{50} for activation together with increased signal-to-background ratio at cAMP saturation relative to the version encoded by the pGloSensor™-20F cAMP construct. In general, we have observed similar relationships between the two constructs when their performance is compared in living cells.

pGloSensor™-20F cAMP Plasmid. The version encoded by this construct performs well in HEK293 cells at 37°C. Luminescence from the pGloSensor™-22F cAMP Plasmid construct can be more difficult to detect at physiologic temperatures.

For a more thorough explanation of the general performance differences between the two plasmids, please consult Section 3.B, Recommendations on Choice of GloSensor™ Plasmid, in the Technical Manual (#TM076).

Note: For custom assay development services incorporating the GloSensor™ technology, please contact Promega.

For more information on pricing and accessing the technology, please visit: **www.promega.com/glosensor/**

Features:

- **Best-in-Class Performance:** High Z′ and large signal:background ratio values. Ideally suited to HTS/uHTS. Up to 1,000-fold changes in light output obtained.
- **Live-Cell, Non-Lytic Assay Format:** "Zero-step assay" greatly facilitates HTS/uHTS. Easy monitoring of cAMP in live cells enables a more complete analysis of receptor biology.
- **High Sensitivity and Increased Biological Relevance:** Easy detection of low-abundance, endogenous receptors; direct detection of G_i-coupled receptor activation and inverse agonist activity in the absence of added forskolin. PDE inhibitors not needed.

Protocol	Part#
Technical Manual	TM076

Storage Conditions: Store the pGloSensor™ cAMP Plasmid at –20°C and the GloSensor™ cAMP Reagent at –70°C. Store the resuspended GloSensor™ cAMP Reagent at –70°C in single-use aliquots.

PDE-Glo™ Phosphodiesterase Assay

Product	Size	Cat.#
PDE-Glo™ Phosphodiesterase Assay	1,000 assays	V1361
	10,000 assays	V1362

Description: The PDE-Glo™ Phosphodiesterase Assay is a luminescent, high-throughput screening (HTS) method for measuring cyclic nucleotide phosphodiesterase activity from **purified** sources. Cyclic nucleotide phosphodiesterases (PDEs) are involved in a myriad of cellular processes due to their ability to hydrolyze, and thus control, the levels of the second-messenger signaling molecules cAMP and cGMP.

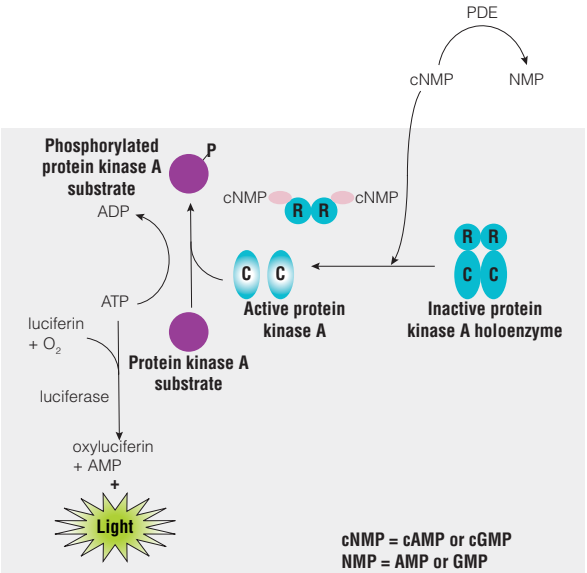
The availability of selective inhibitors for PDEs has facilitated their use as tools to study cyclic nucleotide signaling and paved the way to investigate the role of PDEs in cellular and tissue pathologies. The PDE-Glo™ Phosphodiesterase Assay allows lead candidates to be identified from compound libraries. The assay is designed for 384-well plates, but assay volumes can easily be scaled for 96- or 1536-well plates. The PDE-Glo™ Phosphodiesterase Assay is optimized to work with both cAMP- and cGMP-dependent phosphodiesterases. The total time required for the assay from start to finish is less than 1 hour after the PDE reaction is complete.

Features:

- **Versatile**
  - Works with **both** cAMP and cGMP PDEs.
- **Sensitive**
  - Excellent signal:background ratios
  - Scalable to 1536-well plate formats.
- **Fast and Easy to Use**
  - Assay can be completed in Homogeneous.
- **Proven Luminescent Technology**
  - Powered by Ultra-Glo™ Luciferase.
  - Non-radioactive.
- **No Interference by Fluorescent Compounds.**

Protocol	Part#
Technical Bulletin	TB353

Storage Conditions: Store the system at –20°C. See the product label for the expiration date.



The PDE-Glo™ Phosphodiesterase Assay.

ADP-Glo™ Kinase Assay

Product	Size	Cat.#
ADP-Glo™ Kinase Assay	1,000 assays	V9101
	10,000 assays	V9102
	100,000 assays	V9103

The ADP-Glo™ Kinase Assays contain enough reagent to perform the indicated number of reactions in 384-well format using 5µl, 5µl and 10µl of a kinase reaction, ADP-Glo™ Reagent and Kinase Detection Reagent, respectively, per sample.

Description: ADP-Glo™ Kinase Assay is a luminescent kinase assay that measures ADP formed from a kinase reaction; ADP is converted into ATP, which is converted into light by Ultra-Glo™ Luciferase. The luminescent signal positively correlates with kinase activity. The assay is well suited for measuring the effects of chemical compounds on the activity of a broad range of purified kinases, making it ideal for both primary screening and kinase selectivity profiling. The ADP-Glo™ Kinase Assay can be used to monitor the activity of virtually any ADP-generating enzyme (e.g., kinase or ATPase) using up to 1mM ATP.

The assay is performed in two steps: 1) After the kinase reaction, an equal volume of ADP-Glo™ Reagent is added to terminate the reaction and deplete the remaining ATP; 2) Kinase Detection Reagent is added, which converts ADP to ATP and allows measurement of the newly synthesized ATP using a coupled luciferase/luciferin reaction.

The ADP-Glo™ Kinase Assay has a high dynamic range and produces a strong signal at low ATP to ADP conversion. The assay produces minimal false hits and Z′ values of greater than 0.8.

Features:

- **High Signal Strength at Low ATP Conversion:** Measure kinase activity that more closely mimics physiological conditions. Well suited for low-activity kinases such as receptor tyrosine kinases.
- **Sensitive:** Sensitive to low concentrations of ADP, requiring less enzyme than other assays; cost savings.
- **Universal:** Use with virtually with any kinase—screen a wider range of kinases in-house, reducing dependency on costly outsourcing of kinase selectivity profiling.
- **Accurate:** Measures ADP levels at a wide range of starting ATP concentrations; activity measured truly reflects kinase activity and produces IC_{50}s comparable to radioactivity-based assays.
- **Accommodate Wide Range of ATP Levels:** Use at ATP concentrations up to 1mM, important for kinases with high K_m values for ATP.
- **Stable Luminescent Signal:** Perform batch-plate processing without need for strictly timed incubations.

Protocol	Part#
Technical Manual	TM313

Storage Conditions: Store at –20°C.

Kinase Enzyme Systems

Product	Cat.#	Size	Kinase	Molecular Weight	Substrate	Other
ABL1 Kinase Enzyme System	V1901	10µg	ABL1, 10µg (Human, recombinant; amino acids 27-end)	~135 kDa	Abltide (EAIYAAPFAKKK); derived from the C-terminus of Abl	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + ABL1 Kinase Enzyme System	V9051	1 each				
AKT1 Kinase Enzyme System	V1911	10µg	AKT1, 10µg (Human, recombinant full-length)	~85 kDa	Akt (PKB) substrate (CKRPRAASFAE); derived from the N-terminus of GSK3.	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + AKT1 Kinase Enzyme System	V9061	1 each				
AXL Kinase Enzyme System	V3961	10µg	AXL, 10µg (Human, recombinant; amino acids 473-end)	~55 kDa	Axltide (KKSRGDYMTMQIG); derived from the mouse Insulin receptor substrate 1 (amino acid 979-989).	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + AXL Kinase Enzyme System	V9171	1 each				
AMPK (A1/B1/G1) Kinase Enzyme System	V1921	10µg	AMPK (A1/B1/G1), 10µg (Human, recombinant full-length)	~68kDa (A1), ~38kDa (B1), ~40kDa (G1)	SAMStide (HMRSAMSGLHLVKRR); derived from mouse acetyl-Coenzyme A carboxylase alpha (amino acid 73-85).	Reaction Buffer; DTT; AMP
ADP-Glo™ Kinase Assay + AMPK (A1/B1/G1) Kinase Enzyme System	V9021	1 each				
BTK Kinase Enzyme System	V2941	10µg	BTK, 10µg (Human, recombinant full-length)	~78 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + BTK Kinase Enzyme System	V9071	1 each				
CAMK2 Kinase Enzyme System	V3531	10µg	CAMK2 , 10µg (Human, recombinant; c-terminal truncation)	~60 kDa	Autocamtide-2 (KKALRRQETVDAL-amide) ;derived from the autophosphorylation site (amino acid 283-290) on CaMKII.	Reaction Buffer; DTT; Ca^{2+}/Calmodulin solution
ADP-Glo™ Kinase Assay + CAMK2 Kinase Enzyme System	V9201	1 each				
CAMK4 Kinase Enzyme System	V2951	10µg	CAMK4, 10µg (Human, recombinant full-length)	~79 kDa	Autocamtide-2 (KKALRRQETVDAL-amide) ;derived from the autophosphorylation site (amino acid 283-290) on CaMKII.	Reaction Buffer; DTT; Ca^{2+}/Calmodulin solution
ADP-Glo™ Kinase Assay + CAMK4 Kinase Enzyme System	V9091	1 each				
CDK1/CyclinA2 Kinase Enzyme System	V2961	10µg	CDK1/CyclinA2, 10µg (Human, recombinant full-length)	CDK1 ~59 kDa and CyclinA2 ~78 kDa	Histone H1- Native histone H1 was purified from calf thymus tissues.	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + CDK1/CyclinA2 Kinase Enzyme System	V9211	1 each				
CDK2/CyclinA2 Kinase Enzyme System	V2971	10µg	CDK2/CyclinA2, 10µg (Human, recombinant full-length)	CDK2 ~58 kDa and CyclinA2 ~78 kDa	Histone H1- Native histone H1 was purified from calf thymus tissues.	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + CDK2/CyclinA2 Kinase Enzyme System	V9221	1 each				
CHK1 Kinase Enzyme System	V1941	10µg	CHK1, 10µg (Human, recombinant full-length)	~82 kDa	CHKtide (KKKVSRSGLYRSPSMPENLNRPR); derived from the human CDC25C protein isoform A (amino acid 205-225).	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + CHK1 Kinase Enzyme System	V9241	1 each				
CSK Kinase Enzyme System	V2981	10µg	CSK, 10µg (Human, recombinant full-length)	~78 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + CSK Kinase Enzyme System	V9251	1 each				

9526LA

Kinase Enzyme Systems-continued

Product	Cat.#	Size	Kinase	Molecular Weight	Substrate	Other
EGFR Kinase Enzyme System	V3831	10µg	EGFR, 10µg (Human, recombinant; amino acids 695-end)	~89 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + EGFR Kinase Enzyme System	V9261	1 each				
EPHA1 Kinase Enzyme System	V3561	10µg	EPHA1, 10µg (Human, recombinant; amino acids 569-end)	~71 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + EPHA1 Kinase Enzyme System	V9271	1 each				
FAK Kinase Enzyme System	V1971	10µg	FAK, 10µg (Human, recombinant; amino acids 393-698)	~35 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + FAK Kinase Enzyme System	V9301	1 each				
FGFR1 Kinase Enzyme System	V2991	10µg	FGFR1, 10µg (Human, recombinant; amino acids 399-822)	~73 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + FGFR1 Kinase Enzyme System	V9321	1 each				
FLT1 Kinase Enzyme System	V3001	10µg	FLT1, 10µg (Human, recombinant; amino acids 784-end)	~94 kDa	IGF1Rtide (KKKSPGEYVNIEFG); derived from human IRS-1 protein residues 891-902.	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + FLT1 Kinase Enzyme System	V9331	1 each				
FYN A Kinase Enzyme System	V3571	10µg	FYN A, 10µg (Human, recombinant full-length)	~85kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + FYN A Kinase Enzyme System	V9341	1 each				
GSK3 Kinase Enzyme System	V3051	10µg	GSK3 , 10µg (Human, recombinant full-length)	~81 kDa	GSK3 Substrate (YRRAAVPPSPSLSRHSSPHQ(pS)EDEEE); derived from human muscle glycogen synthase 1 (amino acid 636-661).	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + GSK3 Kinase Enzyme System	V9361	1 each				
GSK3 Kinase Enzyme System	V1991	10µg	GSK3 , 10µg (Human, recombinant full-length)	~48 kDa	GSK3 Substrate (YRRAAVPPSPSLSRHSSPHQ(pS)EDEEE); derived from human muscle glycogen synthase 1 (amino acid 636-661).	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + GSK3 Kinase Enzyme System	V9371	1 each				
HER2 Kinase Enzyme System	V3891	10µg	HER2, 10µg (Human, recombinant; amino acids 676-end)	~116 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + HER2 Kinase Enzyme System	V9381	1 each				
HER4 Kinase Enzyme System	V3101	10µg	HER4, 10µg (Human, recombinant; amino acids 682-993)	~57 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + HER4 Kinase Enzyme System	V9391	1 each				
IGF1R Kinase Enzyme System	V3581	10µg	IGF1R, 10µg (Human, recombinant; amino acids 960-end)	~53 kDa	IGF1Rtide (KKKSPGEYVNIEFG); derived from human IRS-1 protein residues 891-902.	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + IGF1R Kinase Enzyme System	V9401	1 each				
InsR Kinase Enzyme System	V3901	10µg	InsR, 10µg (Human, recombinant; amino acids 1011-end)	~70 kDa	Axltide (KKSRGDYMTMQIG); derived from the mouse Insulin receptor substrate 1 (amino acid 979-989).	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + InsR Kinase Enzyme System	V9411	1 each				
ITK Kinase Enzyme System	V3191	10µg	ITK, 10µg (Human, recombinant; amino acids 352-end)	~53 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + ITK Kinase Enzyme System	V9431	1 each				
JAK3 Kinase Enzyme System	V3701	10µg	JAK3, 10µg (Human, recombinant; amino acids 781-end)	~64 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + JAK3 Kinase Enzyme System	V9441	1 each				
KDR Kinase Enzyme System	V2681	10µg	KDR, 10µg (Human, recombinant; amino acids 789-end)	~110 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + KDR Kinase Enzyme System	V9471	1 each				
LCK Kinase Enzyme System	V2691	10µg	LCK, 10µg (Human, recombinant full-length)	~84 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + LCK Kinase Enzyme System	V9491	1 each				
LYN B Kinase Enzyme System	V3711	10µg	LYN B, 10µg (Human, recombinant full-length)	~85 kDa	SRC substrate (KVEKIGEGTYGVVYK-amide); derived from human p34cdc2 (amino acid 6-20).	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + LYN B Kinase Enzyme System	V9501	1 each				
c-MER Kinase Enzyme System	V3541	10µg	c-MER, 10µg (Human, recombinant; amino acids 578-872)	~58 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + c-MER Kinase Enzyme System	V9561	1 each				
MET Kinase Enzyme System	V3361	10µg	MET, 10µg (Human, recombinant; amino acids 956-end)	~81kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + MET Kinase Enzyme System	V9571	1 each				

9526LB

Kinase Enzyme Systems-continued

Product	Cat.#	Size	Kinase	Molecular Weight	Substrate	Other
p70S6K Kinase Enzyme System	V2741	10µg	p70S6K, 10µg (Human, recombinant full-length)	~76 kDa	S6K substrate (KRRRLASLR); derived from human 40S ribosomal protein S6 (amino acid 230-238).	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + p70S6K Kinase Enzyme System	V9611	1 each				
PDGFR Kinase Enzyme System	V3721	10µg	PDGFR , 10µg (Human, recombinant; amino acids 550-end)	~95 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + PDGFR Kinase Enzyme System	V8011	1 each				
PDGFR Kinase Enzyme System	V3731	10µg	PDGFR , 10µg (Human, recombinant; amino acids 557-end)	~104kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + PDGFR Kinase Enzyme System	V8021	1 each				
PDK1 Kinase Enzyme System	V2761	10µg	PDK1, 10µg (Human, recombinant full-length)	~67 kDa	PDKtide (KTFCGTPEYLAPEVRREPRILSEE-EQEMFRDFDYIADWC); derived from two human proteins: residues 1-14 are based on AKT1 (307-320) and residues 16-39 are based on PKN2/PRK2 (961-984).	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + PDK1 Kinase Enzyme System	V9681	1 each				
PKC Kinase Enzyme System	V3381	10µg	PKC , 10µg (Human, recombinant full-length)	~103 kDa	CREBtide (KRREILSRRPSYR); derived from human CREB1 isoform A (amino acid 109-121).	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + PKC Kinase Enzyme System	V9691	1 each				
PKC II Kinase Enzyme System	V3741	10µg	PKC II, 10µg (Human, recombinant full-length)	~105 kDa	CREBtide (KRREILSRRPSYR); derived from human CREB1 isoform A (amino acid 109-121).	Reaction Buffer; DTT; Lipid solution
ADP-Glo™ Kinase Assay + PKC II Kinase Enzyme System	V9701	1 each				
PKC Kinase Enzyme System	V3391	10µg	PKC , 10µg (Human, recombinant full-length)	~105 kDa	PKCtide (ERMRPRKRQGSVRRRV); derived from protein kinase C epsilon (amino acid 149-164).	Reaction Buffer; DTT; Lipid solution
ADP-Glo™ Kinase Assay + PKC Kinase Enzyme System	V9711	1 each				
PKC Kinase Enzyme System	V3401	10µg	PKC , 10µg (Human, recombinant full-length)	~104 kDa	CREBtide (KRREILSRRPSYR); derived from human CREB1 isoform A (amino acid 109-121).	Reaction Buffer; DTT; Lipid solution
ADP-Glo™ Kinase Assay + PKC Kinase Enzyme System	V9721	1 each				
PKC Kinase Enzyme System	V2781	10µg	PKC , 10µg (Human, recombinant full-length)	~93 kDa	CREBtide (KRREILSRRPSYR); derived from human CREB1 isoform A (amino acid 109-121).	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + PKC Kinase Enzyme System	V9731	1 each				
PKC Kinase Enzyme System	V3751	10µg	PKC , 10µg (Human, recombinant full-length)	~98 kDa	CREBtide (KRREILSRRPSYR); derived from human CREB1 isoform A (amino acid 109-121).	Reaction Buffer; DTT; Lipid solution
ADP-Glo™ Kinase Assay + PKC Kinase Enzyme System	V9751	1 each				
RET Kinase Enzyme System	V3761	10µg	RET, 10µg (Human, recombinant; amino acids 658-end)	~74 kDa	IGF1Rtide (KKKSPGEYVNIEFG); derived from human IRS-1 protein residues 891-902.	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + RET Kinase Enzyme System	V8061	1 each				
ROCK1 Kinase Enzyme System	V3411	10µg	ROCK1, 10µg (Human, recombinant; amino acids 17-535)	~85 kDa	S6K substrate (KRRRLASLR); derived from human 40S ribosomal protein S6 (amino acid 230-238).	Reaction Buffer; DTT; Lipid solution
ADP-Glo™ Kinase Assay + ROCK1 Kinase Enzyme System	V9581	1 each				
RON Kinase Enzyme System	V3921	10µg	RON, 10µg (Human, recombinant; amino acids 983-end)	~71 kDa	Axltide (KKSRGDYMTMQIG); derived from the mouse Insulin receptor substrate 1 (amino acid 979-989).	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + RON Kinase Enzyme System	V8071	1 each				
RSK2 Kinase Enzyme System	V3501	10µg	RSK2, 10µg (Human, recombinant full-length)	~112 kDa	RSK Substrate (KRRRLSSLRA); Derived from human 40S ribosomal protein S6 (amino acid 230-239).	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + RSK2 Kinase Enzyme System	V9651	1 each				
SGK1 Kinase Enzyme System	V2911	10µg	SGK1, 10µg (Human, recombinant; amino acids 60-end)	~73 kDa	Akt (PKB) substrate (CKRPRAASFAE)	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + SGK1 Kinase Enzyme System	V9671	1 each				
SRC Kinase Enzyme System	V2921	10µg	SRC, 10µg (Human, recombinant full-length)	~83 kDa	SRC substrate (KVEKIGEGTYGVVYK-amide); derived from human p34cdc2 (amino acid 6-20).	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + SRC Kinase Enzyme System	V9741	1 each				
SYK Kinase Enzyme System	V3801	10µg	SYK, 10µg (Human, recombinant full-length)	~100kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + SYK Kinase Enzyme System	V8271	1 each				
TRKA Kinase Enzyme System	V2931	10µg	TRKA, 10µg (Human, recombinant; amino acids 440-end)	~66 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT
ADP-Glo™ Kinase Assay + TRKA Kinase Enzyme System	V9761	1 each				
ZAP70 Kinase Enzyme System	V3811	10µg	ZAP70, 10µg (Human, recombinant full-length)	~96 kDa	Poly (4:1 Glu, Tyr) Peptide	Reaction Buffer; DTT; $MnCl_2$
ADP-Glo™ Kinase Assay + ZAP70 Kinase Enzyme System	V8311	1 each				

9526LC

Kinase-Glo® Luminescent Kinase Assays

Product	Size	Cat.#
Kinase-Glo® Luminescent Kinase Assay	10 ml	V6711
	10 × 10 ml	V6712
	100 ml	V6713
	10 × 100 ml	V6714
Kinase-Glo® Plus Luminescent Kinase Assay	10 ml	V3771
	10 × 10 ml	V3772
	100 ml	V3773
	10 × 100 ml	V3774
Kinase-Glo® Max Luminescent Kinase Assay	10 ml	V6071
	10 × 10 ml	V6072
	100 ml	V6073
	10 × 100 ml	V6074

Description: The Kinase-Glo® Luminescent Kinase Assays are homogeneous non-radioactive methods for determining the activity of purified kinases by quantifying the amount of ATP remaining in solution following a kinase reaction. The assays are designed for use with multiwell plate formats, making them ideal for automated high-throughput screening (HTS), and they can be used to assay protein, lipid and sugar kinases. The assay procedure involves addition of a single reagent directly to a completed kinase reaction. This addition results in the generation of a luminescent signal correlated with the amount of ATP present and inversely proportional to the amount of kinase activity. The Kinase-Glo® Assays generate a "glow-type" luminescent signal produced using a patented stabilized luciferase (Ultra-Glo™ Luciferase) coupled with a proprietary buffer system. When assayed in the presence of kinase reaction buffers, such as the reaction buffer for PKA, the half-life of the luminescent output is greater than five hours, eliminating the need for luminometers with injectors and allowing batch plate processing. The assay produces excellent Z′-factor values of greater than 0.7 in 96- and 384-well formats, easily detects known kinase inhibitors and provides IC_{50} values comparable to those reported in the literature.

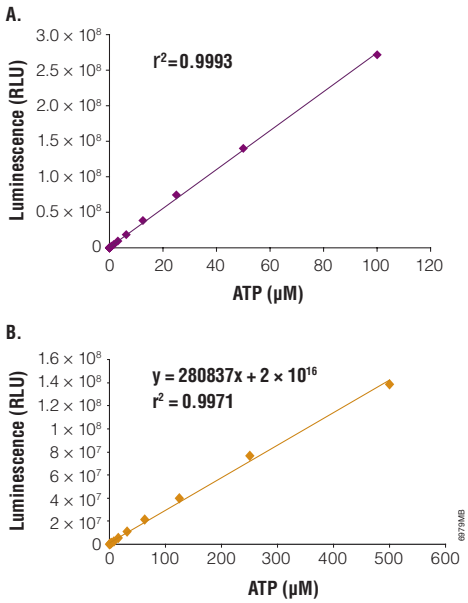
The Kinase-Glo® Assay systems are differentiated by their linear response to ATP (see figure below). The original Kinase-Glo® Assay is linear to 10µM ATP, while Kinase-Glo® Plus Assay is linear to 100µM ATP. The newest assay, Kinase-Glo® Max, is linear to 500µM ATP, making it well suited for use with kinases with high K_m for ATP as well as for screening for kinase inhibitors that do not compete at the ATP binding site.

Features:

- **Assay a Variety of Kinases:** Can be used for a wide range of kinases (including lipid, sugar and alcohol kinases) and substrates (peptides, proteins, lipids, sugars and alcohols).
- **Obtain Reliable Results:** Luminescence is much less susceptible to interference from library compounds than other luciferase-based ATP detection reagents. Z′-factor values greater than 0.7 in either 96- or 384-well plate formats.
- **Simplify Your Assay:** Homogeneous—everything is performed in a single well.
- **Non-Radioactive:** No radioactive waste disposal and safety issues.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**
- **Screen for Non-ATP Binding Site Inhibitors:** Use ATP concentrations as high as 500µM (Kinase-Glo® Max Assay).

Protocol	Part#
Technical Bulletin	TB372

Storage Conditions: Store at –20°C. Avoid multiple freeze-thaw cycles and exposure to frequent temperature changes. These fluctuations can greatly alter product stability.



Luminescent output correlates with amount of ATP. A direct relationship exists between the luminescence measured with the Kinase-Glo® Assay systems and the amount of ATP in the reaction. **Panel A.** Kinase-Glo® Plus Assay. **Panel B.** Kinase-Glo® Max Assay.The Kinase-Glo® Assay is linear to 10µM (data not shown).

ProFluor® PKA Assay

Product	Size	Cat.#
ProFluor® PKA Assay	4 plate	V1240
	8 plate	V1241

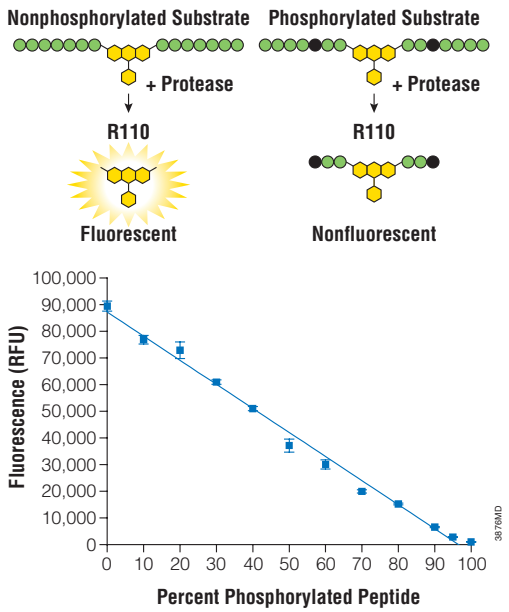
Description: The ProFluor® PKA Assay measures protein kinase A activity using purified kinase in a multiwell plate format and involves "add-mix-read" steps only—ideal for high-throughput applications. The assay begins with a standard kinase reaction performed with a provided PKA bisamide rhodamine 110 peptide substrate. Following the kinase reaction, a termination buffer containing a protease reagent is added, which simultaneously stops the kinase reaction and removes amino acids specifically from the nonphosphorylated PKA substrate, liberating highly fluorescent rhodamine 110. Phosphorylated PKA substrate, however, is resistant to digestion by the protease reagent and remains nonfluorescent. Thus, fluorescence intensity measured in this assay is inversely correlated with kinase activity. The assay produces excellent Z′-factor values (>0.8) in either 96- or 384-well plate formats and easily distinguishes known PKA inhibitors from other compounds.

Features:

- **Achieve Highly Predictive Results:** Robust Z′ values greater than 0.7 in either 96- or 384-well plate formats.
- **Observe Minimal Test Compound Interference:** Rhodamine 110 fluorescent signal produced is much higher than the fluorescent signal given off by test compounds.
- **Homogeneous:** Add-mix-read format reduces the number of steps.
- **Non-Radioactive:** No radioactive waste disposal and safety issues.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB315

Storage Conditions: Store the entire system at –20°C. Protect the PKA R110 Substrate from light. For best results, make solutions fresh and use immediately. System components should be thawed on ice and returned to –20°C as soon as possible. The PKA R110 Substrate is provided in 100% DMSO and therefore requires thawing at room temperature.



Schematic and graph demonstrating that the presence of a phosphorylated amino acid (black circles) blocks the removal of amino acids from the PKA peptide substrate by the protease.

ProFluor® Src-Family Kinase Assay

Product	Size	Cat.#
ProFluor® Src-Family Kinase Assay	4 plate	V1270
	8 plate	V1271

Description: The ProFluor® Src-Family Kinase Assay measures the activity of purified Src-family tyrosine kinases (Src, Lck, Lyn, Fyn, and Hck tested) in a multiwell plate format and involves "add-mix-read" steps only—ideal for high-throughput applications. The assay begins with a standard kinase reaction performed with a provided Src-family kinase bisamide rhodamine 110 peptide substrate. Following the kinase reaction, a termination buffer containing a protease reagent is added, which simultaneously stops the kinase reaction and removes amino acids specifically from the nonphosphorylated substrate, liberating highly fluorescent rhodamine 110. Phosphorylated substrate, however, is resistant to digestion by the protease reagent and remains nonfluorescent. Thus, fluorescence intensity measured in this assay is inversely correlated with kinase activity. A control peptide (AAF-AMC) is included to control for compounds that may inhibit the protease. The assay produces excellent Z′ values (>0.7) in either 96- or 384-well plate formats and easily distinguishes known Src-family kinase inhibitors from other compounds.

Features:

- **Achieve Highly Predictive Results:** Robust Z′ values greater than 0.7 in either 96- or 384-well plate formats.
- **Observe Minimal Test Compound Interference:** Rhodamine 110 fluorescent signal produced is much higher than the fluorescent signal given off by test compounds.
- **Control Peptide Included:** Use AAF-AMC control peptide to monitor protease activity and reduce false-positive hits.
- **Homogeneous:** Add-mix-read format reduces the number of plate-handling steps.
- **Non-Radioactive:** No radioactive waste disposal and safety issues.

Protocol	Part#
Technical Bulletin	TB331

Storage Conditions: For long-term storage, store the system at –20°C. Protect the Src-Family Kinase R110 Substrate and Control AMC Substrate from light. Avoid multiple freeze-thaw cycles or exposure to frequent temperature changes. These fluctuations can greatly alter product stability.

SAM²® Biotin Capture Membrane

Product	Size	Cat.#
SAM²® Biotin Capture Membrane	96 samples	V2861
	7.6 × 10.9 cm	V7861

For Laboratory Use.

Description: The SAM²® Biotin Capture Membrane binds biotinylated molecules based on their affinity for streptavidin. The proprietary process by which the SAM²® Membrane is produced results in a high density of streptavidin on the filter, providing rapid, quantitative substrate binding in the nmol/cm^2 range, depending upon the substrate used. In addition, the membrane is designed to minimize nonspecific binding. The membrane is available either as a large, prenumbered, partially cut sheet (approximately 10.5 × 15.0cm; Cat.# V2861) or as a smaller, uncut sheet (approximately 7.6 × 10.9cm; Cat.# V7861). The partially cut membrane allows easy separation into 96 individual squares and is designed for small-scale experiments where high binding capacity is required. The uncut sheet can be analyzed as a whole membrane or may be cut to the size desired. The uncut membrane allows sample application using a multichannel pipettor. Both membranes may be analyzed using phosphorimaging analysis, autoradiography or scintillation counting to quantitate results. The membranes also have been used successfully with chemiluminescence detection techniques. The use of fluorescence for detection of captured molecules is not recommended at this time.

Features:

- **Use a Variety of Substrates:** Analysis of biotinylated substrates can be applied to a wide variety of substrate types without optimizing each substrate for binding to a matrix. The user can perform experiments with a wide array of sample numbers and sizes without changing the analysis technique, since the membrane is available in 96-square (partially cut) and solid sheet (uncut) formats.
- **Minimize Nonspecific Binding:** The combination of protein denaturant and high-salt washes minimizes nonspecific binding to the membrane without interfering with the high-affinity interaction between streptavidin and biotin.
- **Obtain High Signal-to-Noise Ratios:** The stringent washing conditions employed assist in attaining very low background counts.
- **Perform Kinetic Studies:** Membrane can linearly bind biotinylated substrates up to the nmol/cm^2 range. Allows for kinetic studies.
- **Strong Binding Reaction:** Membrane retains the biotin conjugate over 8 logs of pH (pH 2–10), changes in temperature, organic solvents, ionic and nonionic detergents (SDS, CHAPS, Triton® X-100, Tween® 20 and Tween® 80) and denaturing agents (5M guanidine-HCl and 2M urea).
- **Rapid:** Binds within 1 minute.
- **Convenient:** Compatible with enzyme assays using radioactive detection. Membranes manufactured by this method have been shown to allow chemiluminescent detection.

Protocol	Part#
Technical Bulletin	TB547

Storage Conditions: Store membranes at –20°C in resealable bag.

SAM²® 96 Biotin Capture Plate

Product	Size	Cat.#
SAM²® 96 Biotin Capture Plate	96 -well plate	V7541
	5 × 96 -well plate	V7542

For Laboratory Use.

Description: The SAM²® 96 Biotin Capture Plate contains the SAM²® Biotin Capture Membrane in the wells of a microfiltration plate. The membrane binds biotinylated molecules based on their strong affinity for streptavidin. The proprietary process by which the SAM²® Biotin Capture Membrane is produced results in a high density of streptavidin on the membrane filter matrix, promoting rapid, quantitative capture of biotinylated substrates. In addition, the membrane is designed to minimize nonspecific binding. The 96-well plate configuration allows use of a vacuum manifold or a commercially available plate washer for washes. The plate is supplied with transparent top and bottom seals, allowing data quantitation using a microplate liquid scintillation counter.

Features:

- **Use a Variety of Substrates:** Analyze a wide variety of biotinylated substrates without optimizing each substrate for binding to a matrix.
- **Quantitative Binding:** Nanomole levels of biotinylated substrate/cm^2; 400pmol of biotin per well in the SAM²® Biotin Capture Plate format compares favorably to the approximately 5pmol per well binding capacity of streptavidin-coated plates provided by other suppliers.
- **Obtain High Signal-to-Noise Ratios:** The stringent washing conditions employed assist in attaining very low background counts.
- **Rapid:** Binds within 1 minute.
- **Perform Kinetic Studies:** Membrane can linearly bind biotinylated substrates up to the nmol/cm^2 range.
- **Obtain Reliable Results:** Coefficient of variation within plates and between plates less than or equal to 15%.
- **Convenient:** Washes easily performed with vacuum manifold or commercial plate washer.

Protocol	Part#
Technical Bulletin	TB249

Storage Conditions: Store plates at –20°C.

SignaTECT® Protein Kinase Assay Systems

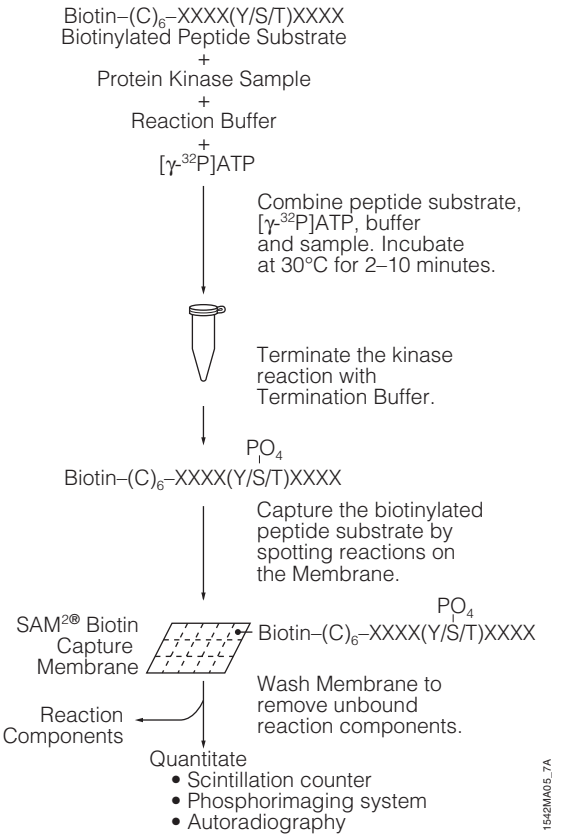
Product	Size	Cat.#
SignaTECT® cAMP-Dependent Protein Kinase (PKA) Assay System	96 reactions	V7480
SignaTECT® Protein Kinase C (PKC) Assay System	96 reactions	V7470
SignaTECT® Protein Tyrosine Kinase (PTK) Assay System	96 reactions	V6480
SignaTECT® Calcium/Calmodulin-Dependent Protein Kinase (CaM KII) Assay System	96 reactions	V8161
SignaTECT® DNA-Dependent Protein Kinase Assay System	96 reactions	V7870
SignaTECT® cdc2 Protein Kinase Assay System	96 reactions	V6430

Description: The SignaTECT® Protein Kinase Assay Systems contain the proprietary SAM$^{2®}$ Biotin Capture Membrane. The streptavidin-coated SAM$^{2®}$ Membranes possess high binding capacity and high specificity characteristics, which produce lower backgrounds and higher signal-to-noise ratios compared to the traditional P81 phosphocellulose method of capture and measurement. The perforated and numbered membrane allows researchers to measure from 1 up to 96 kinase reactions. Following the kinase reaction, samples are spotted onto the SAM$^{2®}$ Membrane, and a series of short wash steps are performed to remove nonspecific label. The process is complete in less than 1 hour. In addition, the nature of the SAM$^{2®}$ Membrane allows it to be used under a variety of buffer/reaction conditions (e.g., cell extracts), which many other methods do not allow. The high binding capacity allows use of the SignaTECT® Systems for kinetic studies.

Each system contains highly specific biotinylated peptide substrates for the appropriate kinase as well as the necessary reaction components. The researcher must supply [γ-^{32}P]ATP.

Protocol	Part#
SignaTECT® PTK Technical Bulletin	TB211
SignaTECT® cdc2 PK Technical Bulletin	TB227
SignaTECT® PKA Technical Bulletin	TB241
SignaTECT® PKC Technical Bulletin	TB242
SignaTECT® DNA-PK Technical Bulletin	TB250
SignaTECT® CaM KII Technical Bulletin	TB279

Storage Conditions: Store all SignaTECT® Systems except V7470 at –20°C. Store Cat.# V7470 at –70°C.



Schematic diagram of the SignaTECT® Protein Kinase Assay protocol. Protocol steps to prepare, run and analyze a specific protein kinase activity using any of the SignaTECT® Protein Kinase Assay Systems.

PepTag® Non-Radioactive Protein Kinase Assays

Product	Size	Cat.#
PepTag® Non-Radioactive PKC Assay	120 reactions	V5330
PepTag® Non-Radioactive cAMP-Dependent Protein Kinase Assay	120 reactions	V5340

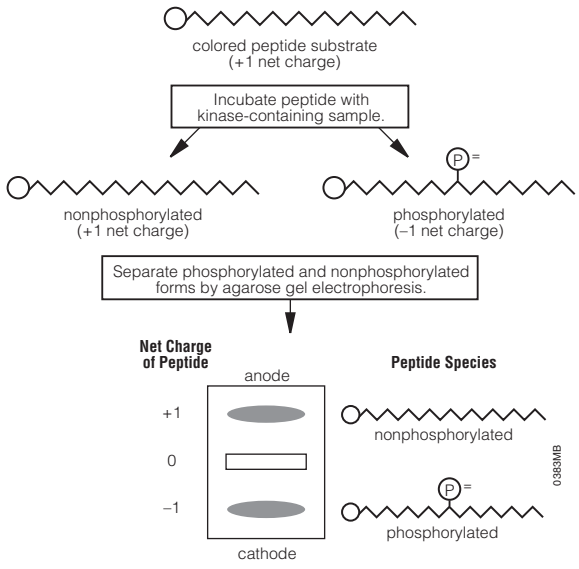
Description: The PepTag® Non-Radioactive Protein Kinase Assay Systems provide a rapid, sensitive and non-radioactive method to detect either Protein Kinase C (PKC) or Protein Kinase A (PKA) activity. The PepTag® Assays use brightly colored, fluorescent peptide substrates that are highly specific for PKC (PepTag® C1 Peptide-PLSRTLSVAAK) and PKA (PepTag® A1 Peptide-LRRASLG). Phosphorylation of the peptide alters the net charge from +1 to –1. This change in the net charge allows the phosphorylated and nonphosphorylated versions of the substrate to be rapidly separated on an agarose gel at neutral pH. Using fluorescent detection, less than 2ng of purified kinase can be detected in less than 2 hours. The PepTag® Non-Radioactive Protein Kinase Assay Systems can detect kinase activity in partially purified samples as well as purified preparations of enzymes, making it a good choice for the rapid screening of column fractions or the screening of kinase activators and inhibitors. In addition to the assay components, each system includes purified kinase for use as a positive control.

Features:

- **Non-Radioactive:** The fluorescent tag on the peptide substrate facilitates quantitation of the phosphorylation reaction without the use of radioactivity.
- **Low Background:** Because the phosphorylation of the colored peptide supplied with the system is used to measure kinase activity, phosphorylation of other substrates occurring naturally in the sample does not add to the kinase activity measured.
- **Convenient:** Quantitation of the phosphorylated peptide can be accomplished using a densitometer, spectrophotometer, 96-well plate reader, or fluorometer.

Protocol	Part#
Technical Bulletin	TB132

Storage Conditions: Store at –70°C.



Schematic diagram of PepTag® Non-Radioactive Protein Kinase Assay procedure.

cAMP-Dependent Protein Kinase, Catalytic Subunit

Product	Size	Conc.	Cat.#
cAMP-Dependent Protein Kinase, Catalytic Subunit	2,500 u	1.5–3 mg/ml	V5161

Description: The purified 40kDa cAMP-Dependent Protein Kinase (PKA), Catalytic Subunit, may be used to phosphorylate target proteins or for in vitro enzymological studies of neural and hormonal signal transduction. Intracellular targets include ion channels, transcriptional activator proteins, and regulatory enzymes of glycogen metabolism.

Features:

- **Highly Pure:** The PKA Catalytic Subunit has been purified from a recombinant *E. coli* strain expressing the catalytic subunit of bovine PKA and is 90% pure.

Protocol	Part#
Promega Product Information	9PIV516

Storage Conditions: Store at –70°C.

DNA-Dependent Protein Kinase

Product	Size	Cat.#
DNA-Dependent Protein Kinase	2,500 u	V5811

Description: DNA-Dependent Protein Kinase (DNA-PK) phosphorylates several DNA-binding substrates in vitro, including the tumor suppressor protein p53, the SV40 large T antigen and several transcription factors. DNA-PK is thought to play a role in controlling gene regulation and cell growth.

DNA-PK is isolated from HeLa nuclear extracts as a complex consisting of a 400kDa catalytic subunit and a 155kDa heterodimeric DNA-binding component named Ku, which itself consists of subunits of approximately 85kDa and 70kDa.

Protocol	Part#
Promega Product Information	9PIV581

Storage Conditions: Store at –70°C.

cGMP-Dependent Protein Kinase

Product	Size	Cat.#
cGMP-Dependent Protein Kinase (α-Isozyme)	6,000 u	V5171

Description: cGMP-Dependent Protein Kinase is a serine/threonine protein kinase present in smooth muscle and a variety of other tissues. The kinase is a 78kDa polypeptide composed of a regulatory domain and a catalytic domain and is active as a homodimer.

Specific Activity: >1,000u/µg (kinase activity).

Features:

- **Highly Pure:** cGMP-Dependent Protein Kinase has been purified by the method of Corbin and Doskeland and is >90% pure as determined by SDS-PAGE (single band).

Protocol	Part#
Promega Product Information	9PIV517

Storage Conditions: Store at –70°C.

Casein Kinase I

Product	Size	Cat.#
Casein Kinase I	100 u	V5631

Description: Casein Kinase I (CKI or CK-1) is a ubiquitous and highly conserved serine/threonine protein kinase found in eukaryotic cells. CKI exists in multiple forms in mammalian tissue and is present in the nucleus, cytosol, plasma membrane and microsomes. CKI isolated from most species is a 35–37kDa monomer. In contrast to Casein Kinase II, CKI primarily uses Mg^{2+}/ATP as the phosphate donor and is not sensitive to heparin inhibition.

Protocol	Part#
Promega Product Information	9PIV563

Storage Conditions: Store at –20°C.

Casein Kinase II

Product	Size	Cat.#
Casein Kinase II	100 u	V5621

Description: Casein Kinase II (CKII or CK-2) is a ubiquitous serine/threonine protein kinase found in eukaryotic cells and is also known as phosvitin kinase, glycogen synthase 5 kinase, troponin T kinase and casein kinase G. CKII is a multifunctional protein kinase implicated in a variety of cellular processes and functions, including mitosis and cellular transformation.

CKII isolated from most species is composed of α and α´ subunits (37–44kDa) and β subunits (24–28kDa). The holoenzyme exists as an $\alpha\alpha'\beta_2$ tetramer. The α subunit contains the catalytic domain, whereas the β subunits presumably regulate the catalytic activity of the holoenzyme. Casein Kinase II is purified from rat liver.

Protocol	Part#
Promega Product Information	9PIV562

Storage Conditions: Store at –70°C.

EGF Receptor

Product	Size	Cat.#
EGF Receptor	10 u	V5551

Description: Epidermal Growth Factor Receptor (EGF Receptor) is a cell-surface glycoprotein composed of a single polypeptide chain that binds the peptide Epidermal Growth Factor (EGF). The EGF Receptor consists of an extracellular ligand binding domain, a single transmembrane region and a cytoplasmic intrinsic tyrosine kinase domain. Upon ligand binding, the EGF Receptor autophosphorylates, activating the tyrosine kinase domain of the EGF Receptor.

EGF Receptor is immunopurified from the A431 cell line following a procedure detailed by Weber *et al.* The purified EGF Receptor does possess tyrosine kinase activity due to the bound EGF; however, the EGF Receptor has not been autophosphorylated.

Protocol	Part#
Promega Product Information	9PIV555

Storage Conditions: Store at –70°C.

Protein Kinase C

Product	Size Conc.	Cat.#
Protein Kinase C	1µg (2 × 0.5 µg) 25 µg/ml	V5261

Description: Protein Kinase C is an 82kDa monomeric enzyme consisting of a C-terminal catalytic domain and a cysteine-rich N-terminal regulatory domain. The regulatory domain contains the sites for calcium and phospholipid binding and a pseudosubstrate subdomain, the target for PKC autophosphorylation. PKC is isolated from rat brain following the procedure of Walton and colleagues. The purified PKC consists primarily of α, β and γ isoforms with lesser amounts of δ and ζ isoforms.

Features:

- **Highly Pure:** PKC is greater than 90% pure as determined by SDS-PAGE.

Protocol	Part#
Promega Product Information	9PIV526

Storage Conditions: Store at –70°C.

MEK Inhibitor U0126

Product	Size	Cat.#
MEK Inhibitor U0126	5mg (5 × 1 mg)	V1121

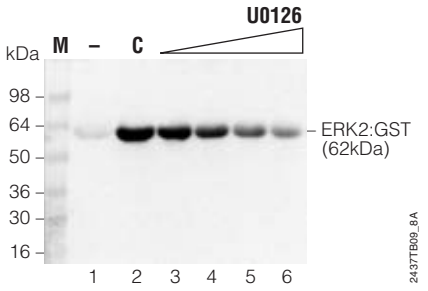
Description: MEK Inhibitor U0126 is a chemically synthesized organic compound that inhibits activation of MAPK (ERK 1/2) by inhibiting the kinase activity of MAP Kinase Kinase (MAPKK or MEK 1/2). U0126 inhibits MEK1 with an IC_{50} of 0.5µM (in vitro). It has been used in both in vivo and in vitro studies of MEK. The chemical identity of MEK Inhibitor U0126 has been confirmed by proton NMR spectroscopy.

Features:

- **Potent:** U0126 inhibits Raf-activated MEK1/2 in vitro with an IC_{50} of approximately 10–20µM.

Protocol	Part#
Promega Product Information	9PIV112

Storage Conditions: Store dried material at –20°C.



MEK1 inhibition by U0126 measured in vitro.Inhibition of Raf-1 kinase-activated wildtype hMEK1 by MEK1 inhibitors in vitro.Lanes 3–6: 10, 25, 50 and 100µM U0126; (Cat.# V1121); lane 1 is a negative control (without NGF); lane C is protein plus no inhibitor.Proteins were resolved by SDS-PAGE and immunoblotted with Anti-ACTIVE® MAPK pAb (Cat.# V8031).

Myristoylated Protein Kinase C Peptide Inhibitor

Product	Size	Cat.#
Myristoylated Protein Kinase C Peptide Inhibitor	1 mg	V5691

Description: Myristoylated Protein Kinase C (PKC) Peptide Inhibitor, Myr. RFARKGALRQKNV (MW = 1,754 daltons), specifically inhibits calcium- and phospholipid-dependent PKC. It is based on the pseudosubstrate region of PKC-α and PKC-β. It is supplied ready for use in kinase reactions. Greater than 75% inhibition of PKC activity is usually obtained with 50µM Myristoylated PKC Peptide Inhibitor.

Properties:

Concentration: 10mg/ml (peptide).
Molarity: 4.3mM.
Peptide Content: ≥ 70%. The mass of the peptide is verified by FAB/MS.
Molecular Weight: 1,754Da.

Features:

- **Cell Permeable:** The Myristoylated Protein Kinase C Peptide Inhibitor is taken up by intact cells, allowing in vivo PKC inhibition studies.

Storage Conditions: Store at –20°C.

InCELLect™ AKAP St-Ht31 Inhibitor Peptide

Product	Size	Cat.#
InCELLect™ AKAP St-Ht31 Inhibitor Peptide	150 µl	V8211

Description: InCELLect™ AKAP St-Ht31 Inhibitor Peptide is a stearated (St) form of the peptide Ht-31 derived from the human thyroid AKAP (A-kinase anchoring protein). This peptide has been shown to inhibit the interaction between the RII subunits of cAMP-Dependent Protein Kinase (PKA) and AKAP in cell extracts. The presence of the hydrophobic stearated moiety enhances the cellular uptake of the peptide through the lipophilic microenvironment of the plasma membrane. InCELLect™ St-Ht31P Control Peptide, which has a very similar sequence but does not inhibit the interaction between protein kinase A and AKAP, has been used as a control peptide for studies with InCELLect™ AKAP St-Ht31 Inhibitor Peptide.

Properties:

Concentration: 10mM in 50mM Tris-HCl (pH 7.5).
Purity: >80% by HPLC.
Molecular Weight: 2,797±3 a.m.u.

Features:

- **Cell Permeable:** The InCELLect™ AKAP St-Ht31 Inhibitor Peptide and Control Peptide are taken up by intact cells, allowing in vivo studies on PKA localization.

Storage Conditions: Dispense into aliquots and store (as a stock solution) at –20°C.

InCELLect™ St-Ht31P Control Peptide

Product	Size	Cat.#
InCELLect™ St-Ht31P Control Peptide	150 µl	V8221

Description: InCELLect™ St-Ht31P Control Peptide is a stearated form of the peptide Ht-31P. This peptide has been used as a negative control peptide for the study of InCELLect™ AKAP St-Ht31 Inhibitor Peptide.

Properties:

Concentration: 10mM.
Purity: >80% by HPLC.
Molecular Weight: 2,765 ±3 a.m.u.

Storage Conditions: Dispense into aliquots and store (as a stock solution) at –20°C. Avoid multiple freeze-thaw cycles.

cAMP-Dependent Protein Kinase Peptide Inhibitor

Product	Size	Cat.#
cAMP-Dependent Protein Kinase Peptide Inhibitor	1 mg	V5681

Description: The cAMP-Dependent Protein Kinase (PKA) Peptide Inhibitor (also known as PKI), TTYADFIASGRTGRRNAIHD, inhibits phosphorylation of target proteins by binding to the protein-substrate site of the catalytic subunit of PKA. It corresponds to the region 5–24 of the naturally occurring protein kinase A inhibitor.

Properties:

Concentration: 10mg/ml in water at neutral pH.
Molecular Weight: 2,221Da.

Storage Conditions: Store at –20°C.

Olomoucine (cdc2 Protein Kinase Inhibitor)

Product	Size	Cat.#
Olomoucine (cdc2 Protein Kinase Inhibitor)	0.5 mg	V2372
	10 mg	V2373

Description: Olomoucine is a chemically synthesized inhibitor that is specific for $p34^{cdc2}$ and related protein kinases. Its molecular weight is 298, and its molecular formula is $C_{15}H_{18}N_{60}$. The chemical identity has been confirmed by proton NMR spectroscopy.

Storage Conditions: Store desiccated at –20°C.

PD 98059

Product	Size	Cat.#
PD 98059	5 mg	V1191

Description: PD 98059 (2′-amino-3′-methoxyflavone) is a potent, cell-permeable and selective inhibitor of MAPK/ERK kinase 1 (MAP kinase kinase 1 or MEK1). It blocks the activation of MEK1, therefore inhibiting the subsequent phosphorylation and activation of MAP kinase. IC_{50} values are in the 1–20µM range. The chemical identity and purity of PD 98059 is confirmed by proton NMR and elemental analysis.

Storage Conditions: Store dried material at –20°C.

SB 203580

Product	Size	Cat.#
SB 203580	1 mg	V1161

Description: SB 203580 [4-(4′-fluorophenyl)-2-(4′-methylsulfinylphenyl)-5-(4′-pyridyl) imidazole] is a specific, cell-permeable inhibitor of the stress- and inflammatory cytokine-activated MAP kinase homologues p38α, p38β and p38β2. SB 203580 acts as a competitive inhibitor of ATP binding. Reported IC_{50} values for p38 activity range from 21nM to 1µM. SB 203580 has no significant effect on the activities of ERKs, JNKs, p38γ or p38δ. The chemical identity and purity of SB 203580 is confirmed by proton NMR and elemental analysis.

Storage Conditions: Store dried material at –20°C.

LY 294002

Product	Size	Cat.#
LY 294002	5 mg	V1201

Description: LY 294002 [2-(4-Morpholinyl)-8-phenyl-4 H-1-benzopyran-4-one] is a potent and specific cell-permeable inhibitor of phosphatidylinositol 3-kinases (PI 3-kinases) with IC_{50} values in the 1–50µM range. LY 294002 competitively inhibits ATP binding to the catalytic subunit of PI 3-kinases and does not inhibit PI 4-kinase, DAG-kinase, PKC, PKA, MAPK, S6 kinase, EGFR or c-src tyrosine kinases and rabbit kidney ATPase. The chemical identity and purity of LY 294002 is confirmed by proton NMR and elemental analysis.

Storage Conditions: Store dried material at –20°C in the dark, ideally in a nonfrost-free freezer.

Kemptide (PKA) Peptide Substrate

Product	Size	Conc.	Cat.#
Kemptide (PKA) Peptide Substrate	1 mg	10 mg/ml	V5601

Description: Kemptide is a peptide substrate for cAMP-Dependent Protein Kinase with the sequence Leu-Arg-Arg-Ala-**Ser**-Leu-Gly.

Properties:
Concentration: 10mg/ml in water.
Molecular Weight: 772Da.
Peptide Content: ≥ 70%. The mass of the peptide is verified by FAB/MS.

Storage Conditions: Store at –20°C.

$Neurogranin_{(28–43)}$ (PKC) Peptide Substrate

Product	Size	Conc.	Cat.#
$Neurogranin_{(28–43)}$ (PKC) Peptide Substrate	1 mg	10 mg/ml	V5611

Description: $Neurogranin_{(28–43)}$ serves as a specific and potent substrate for calcium/phospholipid-dependent protein kinase (PKC) having the sequence Ala-Ala-Lys-Ile-Gln-Ala-**Ser**-Phe-Arg-Gly-His-Met-Ala-Arg-Lys-Lys.

Properties:
Concentration: 10mg/ml in water at neutral pH.
Molecular Weight: 1,800Da.
Peptide Content: ≥ 70%. The mass of the peptide is verified by FAB/MS.

Storage Conditions: Store at –20°C.

DNA-Dependent Protein Kinase Peptide Substrate

Product	Size	Conc.	Cat.#
DNA-Dependent Protein Kinase Peptide Substrate	1 mg	10 mg/ml	V5671

Description: DNA-Dependent Protein Kinase (DNA-PK) Peptide Substrate is a highly specific substrate for DNA-PK, with the sequence Glu-Pro-Pro-Leu-**Ser**-Gln-Glu-Ala-Phe-Ala-Asp-Leu-Trp-Lys-Lys. The K_m value for this peptide is 760µM. DNA-PK is a nuclear serine/threonine protein kinase that, when activated by DNA, phosphorylates several DNA-binding substrates, including the tumor suppressor protein p53, the simian virus 40 (SV40) large T antigen and several transcription factors (e.g., SP1, OCT-1, c-Fos and serum response factor).

Properties:
Concentration: 10mg/ml in water at neutral pH.
Molecular Weight: 1,759Da.
Peptide Content: ≥ 70%. The mass of the peptide is verified by FAB/MS.

Storage Conditions: Store at –20°C.

cdc2 Protein Kinase Peptide Substrate

Product	Size	Conc.	Cat.#
cdc2 Protein Kinase Peptide Substrate	1 mg	10 mg/ml	V2211

Description: cdc2 Protein Kinase Peptide Substrate is a synthetic peptide with the sequence Pro-Lys-**Thr**-Pro-Lys-Lys-Ala-Lys-Lys-Leu. This sequence was derived from the $p34^{cdc2}$ in vitro phosphorylation sites of histone H1. The K_m value for this peptide substrate is 5μM. cdc2 kinase is a serine/threonine kinase that is known to be involved in the G2/M transition of the cell cycle. cdc2 requires a regulatory factor, cyclin, for kinase activity.

Properties:

Concentration: 10mg/ml in water at neutral pH.
Molecular Weight: 1,137Da.
Peptide Content: ≥ 70%. The mass of the peptide is verified by FAB/MS.

Storage Conditions: Store at –20°C.

cGMP-Dependent Protein Kinase (PKG) Peptide Substrate

Product	Size	Conc.	Cat.#
cGMP-Dependent Protein Kinase Peptide Substrate	1 mg	10 mg/ml	V7451

Description: cGMP-Dependent Protein Kinase (PKG) Peptide Substrate is a synthetic peptide having the sequence Arg-Lys-Ile-**Ser**-Ala-Ser-Glu-Phe. This sequence was derived from bovine lung cGMP-binding phosphodiesterase, a relatively specific substrate for cGMP-Dependent Protein Kinase. The K_m value for this substrate is 68μM. cGMP-Dependent Protein Kinase is a serine/threonine kinase present at highest levels in smooth muscle and at lower levels in a variety of other tissues, including lung, heart and Purkinje cells of the cerebellum. This kinase has been implicated in the regulation of smooth muscle relaxation, platelet function, sperm metabolism, cell division and nucleic acid synthesis.

Properties:

Concentration: 10mg/ml in water at neutral pH.
Molecular Weight: 937Da.
Peptide Content: ≥ 70%. The mass of the peptide is verified by FAB/MS.

Storage Conditions: Store at –20°C.

Casein Kinase I Peptide Substrate

Product	Size	Conc.	Cat.#
Casein Kinase I Peptide Substrate	1 mg	10 mg/ml	V7441

Description: Casein Kinase I Peptide Substrate is a specific substrate for Casein Kinase I, with the sequence Asp-Asp-Asp-Glu-Glu-**Ser**-Ile-Thr-Arg-Arg. The K_m value of the CKI Peptide Substrate was determined to be in the range of 0.5–1mM. A number of other protein kinases, including Casein Kinase II, show no significant phosphorylation of this peptide substrate.

Properties:

Concentration: 10mg/ml in water at neutral pH.
Molecular Weight: 1,235Da.
Peptide Content: The mass of the peptide is verified by FAB/MS.

Storage Conditions: Store at –20°C.

Casein Kinase II Peptide Substrate

Product	Size	Conc.	Cat.#
Casein Kinase II Peptide Substrate	1 mg	10 mg/ml	V5661

Description: Casein Kinase II Peptide Substrate is a selective substrate for Casein Kinase II, with the sequence Arg-Arg-Arg-Glu-Glu-Glu-**Thr**-Glu-Glu-Glu. The K_m value of the CKII Peptide Substrate was determined to be 0.5mM. A number of other protein kinases, including PKA, PKC, phosphorylase kinase, MLCK and Casein Kinase I show no significant phosphorylation of this peptide substrate.

Properties:

Concentration: 10mg/ml in water at neutral pH.
Molecular Weight: 1,362Da.
Peptide Content: ≥ 70%. The mass of the peptide is verified by FAB/MS.

Storage Conditions: Store at –20°C.

PMA

Product	Size	Cat.#
PMA	5 mg	V1171

Description: Phorbol 12-myristate 13-acetate (PMA) is a specific activator of group A (α, βI, βII, γ) and group B (δ, ε, η, θ) Protein Kinase Cs (PKCs) (in the 1–100nM range). Phorbol esters, such as PMA, affect PKCs by mimicking diacylglycerol, a natural ligand and activator of PKCs. PMA is an effective skin irritant and potent tumor promoter in mice. A common alternative name for PMA is 12-O-tetradecanoylphorbol 13-acetate (TPA). The chemical identity and purity of PMA are confirmed by proton NMR.

Storage Conditions: Store dried material at –20°C.

4α-PMA

Product	Size	Cat.#
4α-PMA	1 mg	V1181

Description: 4α-phorbol 12-myristate 13-acetate (4α-PMA) is an inactive analogue of and negative control for phorbol 12-myristate 13-acetate (PMA), which is also known as 12-O-tetradecanoylphorbol 13-acetate (TPA). Effects of PMA in a given system are thought to be specific if 4α-PMA is not active in the same system. The chemical identity and purity of 4α-PMA are confirmed by proton NMR.

Storage Conditions: Store dried material at –20°C.

cGMP and cAMP

Product	Size	Cat.#
cGMP, 1mM	500 µl	V6411
cAMP, 1mM	500 µl	V6421
For Laboratory Use.		

Description: cAMP (adenosine-3´,5´-cyclic monophosphate) is an activator of cAMP-Dependent Protein Kinase, Catalytic Subunit (Cat.# V5161). cGMP (guanosine-3´,5´-cyclic monophosphate) is an activator of cGMP-Dependent Protein Kinase (Cat.# V5171).

Storage Conditions: Store at –20°C.

PPase-2A

Product	Size	Cat.#
PPase-2A	25 u	V6311

Description: Protein Phosphatase-2A (PPase-2A) is a serine/threonine phosphatase isolated from human red blood cells. It is isolated as the heterodimer of 60kDa (A) and 36kDa (C) subunits. It has the ability to dephosphorylate the α-subunit of phosphorylase kinase. With its 36–38kDa catalytic subunit, PPase-2A has broad substrate specificity and may play a regulatory role in DNA replication, transcription, protein synthesis, mitosis and glycogen metabolism. PPase-2A is stimulated in vitro by basic proteins such as protamine, histones and polylysine. The enzyme is inhibited by several environmental toxins and tumor promoters such as okadaic acid and microcystin-LR. The chemically synthesized phosphopeptide, RRA(pT)VA (available in the Ser/Thr Phosphatase Assay System, Cat.# V2460), is an excellent substrate for PPase-2A.

Protocol	Part#
Promega Product Information	9PIV631

Storage Conditions: Store at –20°C.

PPase-2B

Product	Size	Cat.#
PPase-2B	10 u	V6361

Description: PPase-2B is a heterodimeric enzyme composed of a 19kDa calcium-binding subunit and a catalytic subunit (61kDa) that binds calmodulin. PPase-2B was originally identified based on its calcium- and calmodulin-dependent activity toward phosphorylase kinase and inhibitor-1. PPase-2B is identical to the brain protein calcineurin, which constitutes up to 1% of total brain protein. The immunosuppressive drugs FK-506 and cyclosporin A inhibit PPase-2B activity in immune cells, implicating a role for this enzyme in regulation of the immune system. PPase-2B also plays a major role in regulating secretory functions of a variety of cells.

PPase-2B is less sensitive to okadaic acid than PPase-2A and PPase-1, requiring micromolar concentrations of okadaic acid for inhibition. It is not inhibited by Inhibitor-1 or Inhibitor-2. Promega PPase-2B is isolated from bovine brain.

Protocol	Part#
Promega Product Information	9PIV636

Storage Conditions: Store at –70°C.

Non-Radioactive Phosphatase Assay Systems

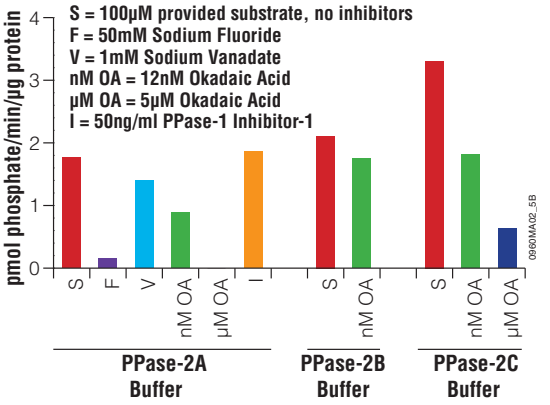
Product	Size	Cat.#
Serine/Threonine Phosphatase Assay System	96 reactions	V2460
Tyrosine Phosphatase Assay System	96 reactions	V2471

Description: The Non-Radioactive Phosphatase Assay Systems provide a fast, convenient and flexible alternative for measuring protein phosphatase activity. These systems determine the amount of free phosphate generated in a reaction by measuring the absorbance of a molybdate:malachite green:phosphate complex. These systems allow the use of a variety of buffer conditions and substrates, including naturally phosphorylated proteins or synthetic phosphopeptides. The Serine/Threonine Phosphatase Assay System contains the chemically synthesized phosphopetide, RRA(pT)VA, a peptide substrate that is compatible with several serine/threonine phosphatases such as the Protein Phosphatases 2A, 2B, and 2C. **However the supplied phosphopeptide is a poor substrate for Protein Phosphatase 1 because of its more stringent structural requirements.**

The Tyrosine Phosphatase Assay System contains two chemically synthesized phosphopeptides, END(pY)INASL and DADE(pY)LIPQQG, that serve as substrates for many protein tyrosine phosphatases. The effective range for the detection of phosphate released during an assay using the Phosphatase Assay Systems is 100–4,000pmol of phosphate. In addition to measuring phosphatase activity in partially fractionated and purified samples, the Phosphatase Assay Systems can also measure phosphatase activity in crude cell or tissue extracts. For this application, the high concentration of phosphate in these preparations is eliminated prior to performing the assay using the supplied Spin Columns, which rapidly remove free phosphate and other low-molecular-weight inhibitors from the sample. In addition, a unique Molybdate Dye Additive that is combined with the Molybdate Dye Solution aids in the solubilization of proteins exposed to the acid conditions of the Molybdate Dye Solution, which alone could potentially cause precipitation of the proteins.

Protocol	Part#
Serine/Threonine Phosphatase Assay System Technical Bulletin	TB218
Tyrosine Phosphatase Assay System Technical Bulletin	TB212

Storage Conditions: Store the entire kit at 4°C.



Serine/Threonine phosphatase activity in HeLa cell extract using the Serine/Threonine Phosphatase Assay System.

ProFluor® Ser/Thr PPase Assay

Product	Size	Cat.#
ProFluor® Ser/Thr PPase Assay	4 plate	V1260
	8 plate	V1261

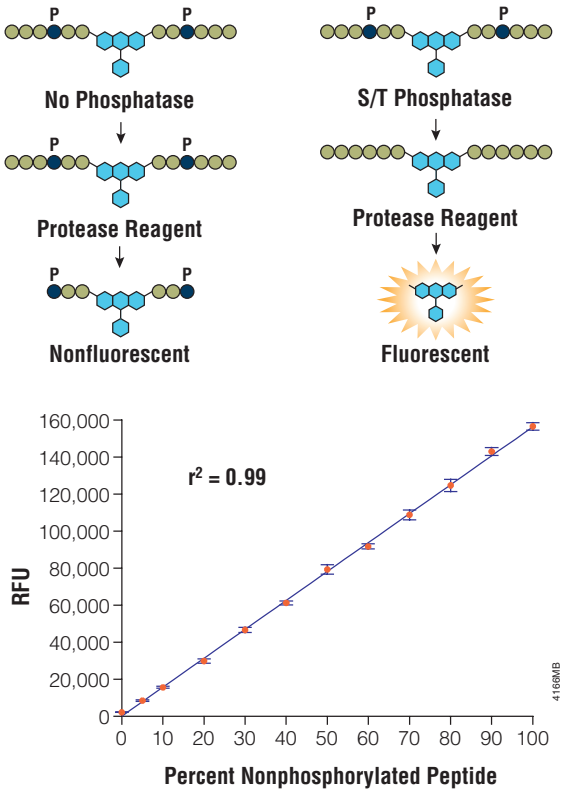
Description: The ProFluor® Ser/Thr PPase Assay measures purified serine/threonine protein phosphatase activity in a multiwell plate format and involves "add-mix-read" steps only—ideal for high-throughput applications. The assay works with protein phosphatase 1 (PP1), PP2A, PP2B and PP2C. The assay begins with a standard phosphatase reaction performed with a provided phosphorylated bisamide rhodamine 110 peptide substrate (S/T PPase R110 Substrate) and Control AMC Substrate that serves as a control for compounds that may inhibit the protease reaction. Following the phosphatase reaction, a termination buffer containing a protease reagent is added, which simultaneously stops the phosphatase reaction and removes amino acids specifically from the nonphosphorylated substrate, liberating highly fluorescent rhodamine 110. Phosphorylated substrate, however, is resistant to digestion by the protease reagent and remains nonfluorescent. Thus, fluorescence intensity measured in this assay is directly correlated with phosphatase activity. The assay produces excellent Z′ values (>0.8) in either 96- or 384-well plate formats and easily distinguishes known phosphatase inhibitors from other compounds.

Features:

- **Achieve Highly Predictive Results:** Robust Z′ values greater than 0.7 in either 96- or 384-well plate formats.
- **Observe Minimal Test Compound Interference:** Rhodamine 110 fluorescent signal produced is much higher than the fluorescent signal given off by test compounds.
- **Control Peptide Included:** Use AAF-AMC control peptide to monitor protease activity and reduce false-positive hits.
- **Simplify Your Assays:** Add-mix-read format reduces the number of steps.
- **Non-Radioactive:** No radioactive waste disposal and safety issues.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB324

Storage Conditions: Store the entire system at –20°C. Protect the S/T PPase R110 Substrate and Control AMC Substrate from light.



Effect of phosphopeptide content on fluorescence intensity. The graph shows the average RFU obtained after a 90-minute digestion of mixtures of nonphosphorylated S/T PPase R110 Substrate and phosphorylated substrate as indicated to mimic a phosphatase titration.

ProFluor® Tyrosine Phosphatase Assay

Product	Size	Cat.#
ProFluor® Tyrosine Phosphatase Assay	4 plate	V1280
	8 plate	V1281

Description: The ProFluor® Tyrosine Phosphatase Assay measures purified tyrosine phosphatase enzyme activity in a multiwell plate format and involves "add-mix-read" steps only—ideal for high-throughput applications. Tyrosine phosphatases tested with the assay include PTP-1B, CD45, LAR PTPase and YOP-51. The assay begins with a standard phosphatase reaction performed with a provided phosphorylated bisamide rhodamine 110 peptide substrate (PTPase R110 Substrate) and Control AMC Substrate that serves as a control for compounds that may inhibit the protease. Following the phosphatase reaction, a termination buffer containing a protease reagent is added, which simultaneously stops the phosphatase reaction and removes amino acids specifically from the nonphosphorylated substrate, liberating highly fluorescent rhodamine 110. Phosphorylated substrate, however, is resistant to digestion by the protease reagent and remains nonfluorescent. Thus, fluorescence intensity measured in this assay is directly correlated with phosphatase activity. The assay produces excellent Z´ values (>0.7) in either 96- or 384-well plate formats and easily distinguishes known phosphatase inhibitors from other compounds.

Features:

- **Achieve Highly Predictive Results:** Robust Z´ values greater than 0.8 in either 96- or 384-well plate formats.
- **Observe Minimal Test Compound Interference:** Substrate used at micromolar concentration. Rhodamine 110 fluorescent signal produced is much higher than the fluorescent signal given off by test compounds.
- **Control Peptide Included:** Control peptide (AAF-AMC) included that is used to monitor protease activity. Reduces false positive hits.
- **Simplify Your Assays:** Simple add-mix-read format reduces the number of plate-handling steps to fewer than that required for other phosphatase assays.
- **Save Time:** Minimal throughput time compared to the multiple steps and lengthy incubations with other phoshatase assays.
- **Non-Radioactive:** No radioactive waste disposal and safety issues.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB334

Storage Conditions: Store the entire system at –20°C. Protect the PTPase R110 Substrate and Control AMC Substrate from light.

Growth Factors

Product	Size	Cat.#
rhBDNF	5 µg	G1491
rhEGF	100 µg	G5021
rhFGF, Basic	25 µg	G5071
rhGDNF	5 µg	G2781
rhIGF-I	25 µg	G5111
mNGF, 2.5S	100 µg	G5141
rhTNFα	10 µg	G5241

Description: Growth factors, cytokines and neurotrophic factors are used in a wide variety of applications. Growth factors typically act as signaling molecules between cells. They are capable of stimulating cellular proliferation and differentiation and are important for regulating a variety of cellular processes. When binding to its receptor a growth factor initiates, or sometimes blocks, the ability of cells to divide and multiply. Besides their application in the areas of cell growth and differentiation, growth factors are used in the study of signal transduction, embryonic development, immunology, tumorigenesis, and clinical medicine. Growth factors are also used for maintenance of cells in culture.

Protocol	Part#
rhBDNF Promega Product Information	9PIG149
rhGDNF Promega Product Information	9PIG278
rhEGF Promega Product Information	9PIG502
rhFGF, Basic Promega Product Information	9PIG507
rhIGF-I Promega Product Information	9PIG511
NGF, 25S, Murine Promega Product Information	9PIG514
rhTNF-αPromega Product Information	9PIG524

Vitronectin, Human

Product	Size	Cat.#
Vitronectin, Human	100 µg	G5381

Description: Human Vitronectin is purified from plasma. Vitronectin belongs to the group of structurally and functionally homologous adhesive proteins (fibrinogen, fibronectin, Von Willebrand factor) that interact with platelets and the vessel wall in the early stages of blood clotting. When coated on surfaces, very low concentrations of Vitronectin promote endothelial cell attachment and induce spreading and migration of cells in a time- and concentration-dependent fashion.

Activity: When coated onto tissue culture plastic, Vitronectin promotes one-half maximal attachment of BALB/3T3 fibroblasts in serum-free medium below 0.1µg/cm^2. Maximal attachment occurs at approximately 0.2µg/cm^2.

Protocol	Part#
Promega Product Information	9PIG538

Storage Conditions: Store at –70°C.

Griess Reagent System

Product	Size	Cat.#
Griess Reagent System	1,000 assays	G2930

Description: The Griess Reagent System measures nitrite (NO_2^-), which is one of two primary stable and nonvolatile breakdown products of nitric oxide (NO). Nitric oxide is an important physiological messenger and effector molecule in many biological systems, including immunological, neuronal and cardiovascular tissues. This assay relies on a diazotization reaction that was originally described by Griess in 1879. Through the years, many modifications to the original reaction have been described.

The Griess Reagent System is based on a chemical reaction that uses sulfanilamide and N-1-naphthylethylenediamine dihydrochloride (NED) under acidic (phosphoric acid) conditions. This system detects NO_2^- in a variety of biological and experimental liquid matrices such as plasma, serum, urine and tissue culture medium. The nitrite sensitivity is dependent on the matrix. The limit of detection is 2.5µM (125pmol) nitrite (in ultrapure, deionized, distilled water) using the protocol described in Technical Bulletin #TB229.

Protocol	Part#
Technical Bulletin	TB229

Storage Conditions: Store at 4°C. Keep all solutions in their original light-protective plastic bottles.

psiCHECK™-1 and psiCHECK™-2 Vectors

Product	Size	Cat.#
psiCHECK™-1 Vector	20 µg	C8011
psiCHECK™-2 Vector	20 µg	C8021

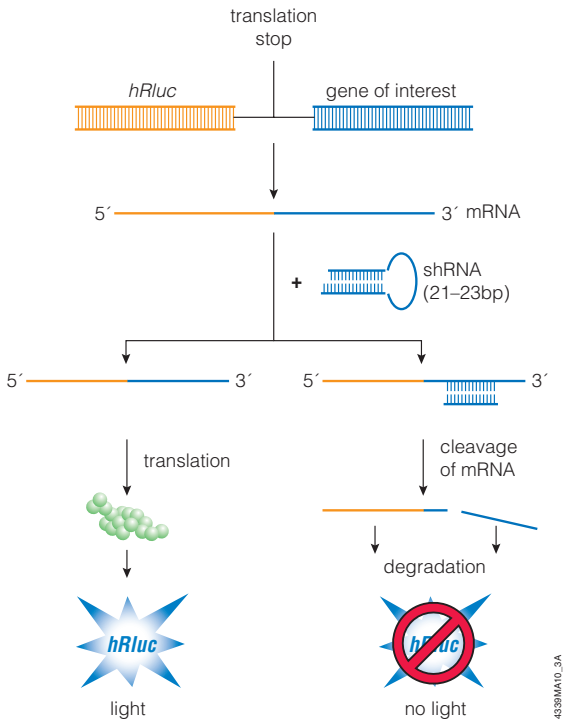
Description: The psiCHECK™-1 and psiCHECK™-2 Vectors are designed to provide a quantitative and rapid approach for initial optimization of RNA interference (RNAi). The vectors enable monitoring of changes in expression of a target gene fused to a reporter gene. In both vectors *Renilla* luciferase is used as the primary reporter gene, and the gene of interest is cloned into a multiple cloning region located downstream of the *Renilla* translational stop codon. Initiation of the RNAi process by synthetic siRNAs or in vivo-expressed shRNAs toward a gene of interest results in cleavage and subsequent degradation of the fusion mRNA. Measuring decreases in *Renilla* activity provides a convenient way of monitoring the RNAi effect. In comparison with other fusion approaches (e.g., GFP or flag-tags), the *Renilla* luciferase approach offers more convenient and rapid quantitation with higher sensitivity. The psiCHECK™-1 Vector is recommended for use in monitoring RNAi effects in live cells. The changes in *Renilla* luciferase activity are measured with the EnduRen™ Live Cell Substrate (Cat.# E6481), which allows continuous monitoring of intracellular *Renilla* luminescence. The psiCHECK™-2 Vector contains a second reporter gene, firefly luciferase, and is designed for endpoint lytic assays. Introduction of firefly luciferase in the psiCHECK™-2 Vector allows normalization of *Renilla* luciferase expression, achieving robust and reproducible results.

Features:

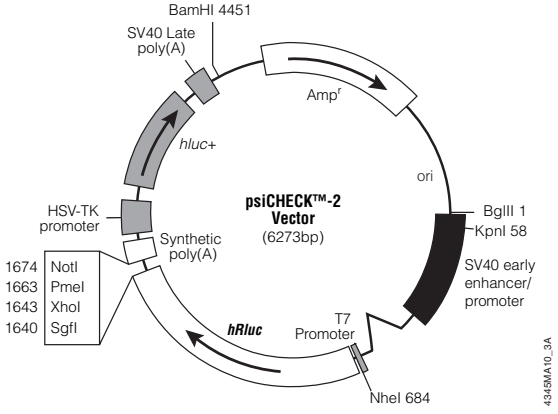
- **Save Money:** Quantitation is performed with a common luminometer; no need to purchase expensive equipment.
- **Choose Your Format:** Protocols allow measurements in live cells or crude cell lysates.
- **Save Time:** No requirement for labor-intensive, time-consuming assays or waiting for phenotypic changes.
- **Convenient:** No requirement for transfection normalization when using the psiCHECK™-2 Vector.

Protocol	Part#
Technical Bulletin	TB329

Storage Conditions: Store at –20°C.



Mechanism of action of the psiCHECK™ Vectors.



GeneClip™ U1 Hairpin Cloning Systems

Product	Size	Cat.#
GeneClip™ U1 Hairpin Cloning System—Basic	1 system	C8750
GeneClip™ U1 Hairpin Cloning System—Puromycin	1 system	C8760
GeneClip™ U1 Hairpin Cloning System—Hygromycin	1 system	C8770
GeneClip™ U1 Hairpin Cloning System—Neomycin	1 system	C8780
GeneClip™ U1 Hairpin Cloning System—hMGFP	1 system	C8790

Description: The GeneClip™ U1 Hairpin Cloning Systems consist of linearized plasmids designed for fast and easy cloning of human target sequences to express short hairpin RNAs (shRNAs) in human cells. After transfection into human cells, in vivo expression of short interfering RNAs (siRNAs) can be effectively achieved from DNA constructs that contain a U1 RNA polymerase promoter and a siRNA template. The U1 promoter has been used successfully to generate hairpin siRNAs in vivo.

To insert hairpin siRNAs into the pGeneClip™ Vectors, two short DNA oligonucleotides are annealed to form a DNA insert that contains the hairpin siRNA target sequence. After annealing, the oligonucleotides form overhangs that are compatible with the pGeneClip™ Vector ends and facilitate sticky-end ligation. Once transfected, RNA polymerase II transcribes the hairpin insert sequences to generate hairpin siRNAs in vivo.

The siRNA Target Designer program (**www.promega.com/siRNADesigner/**) designs oligonucleotides for use with Promega RNA interference systems. The program analyzes input DNA or RNA sequences for regions that fit siRNA design requirements. siRNAs that could target these regions are displayed along with the oligonucleotides required for use with the chosen system.

Features:

- **More Vector Choices:** These systems provide vectors containing a variety of eukaryotic antibiotic-selectable markers for stable transfection or hMGFP for determination of transfection efficiency.
- **Time Savings:** Vectors are supplied predigested to eliminate time-consuming vector preparation.
- **Convenience:** Each system includes T4 DNA Ligase, 2X Rapid Ligation Buffer, Oligo Annealing Buffer and the pGeneClip™ Vector.
- **Easier Identification of Desired Clones:** A PstI digestion quickly identifies positive recombinants.

Protocol	Part#
Technical Manual	TM256

Storage Conditions: Store at –20°C.

T7 RiboMAX™ Express RNAi System

Product	Size	Cat.#
T7 RiboMAX™ Express RNAi System	50 × 20µl reactions	P1700

Description: The T7 RiboMAX™ Express RNAi System is an in vitro transcription system designed for producing milligram amounts of double-stranded RNA (dsRNA) in a short amount of time. The dsRNA is free of protein and other contaminants and is suitable for use in RNA interference (RNAi) in both mammalian and nonmammalian systems.

The T7 RiboMAX™ Express RNAi System can be used to synthesize short interfering RNAs (siRNAs) of 21bp for use in mammalian systems. siRNAs synthesized in vitro have been demonstrated to be as effective as chemically synthesized siRNAs for inducing RNAi in mammalian cells.

In addition, the T7 RiboMAX™ Express RNAi System can be used for the synthesis of dsRNA molecules of approximately 200bp or greater, which can be applied to nonmammalian systems. Two complementary RNA strands are synthesized from DNA template (either plasmid or PCR product). The resulting RNA strands are annealed after the transcription reaction to form dsRNA. Any remaining single-stranded RNA and DNA template are removed with a nuclease digestion step. The dsRNA is then purified by isopropanol precipitation and can be introduced into the organism of choice for RNAi applications.

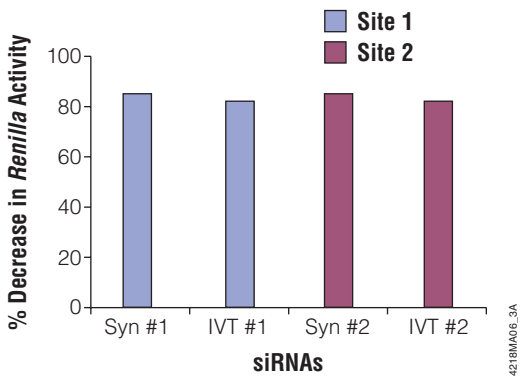
The siRNA Target Designer program (**www.promega.com/siRNADesigner/**) designs oligonucleotides for use with Promega RNA interference systems. The program analyzes input DNA or RNA sequences for regions that fit siRNA design requirements. siRNAs that could target these regions are displayed along with the oligonucleotides required for use with the chosen system.

Features:

- **Save Time:** The T7 RiboMAX™ Express RNAi System produces milligram amounts of RNA in as little as 30 minutes.
- **Minimize Pipetting Errors:** The four rNTPs and 2X transcription buffer have been combined, thus minimizing pipetting errors and setup time.

Protocol	Part#
Technical Bulletin	TB316

Storage Conditions: Store all components at –20°C, except RNase A, which should be stored at 22–25°C after the initial thaw.

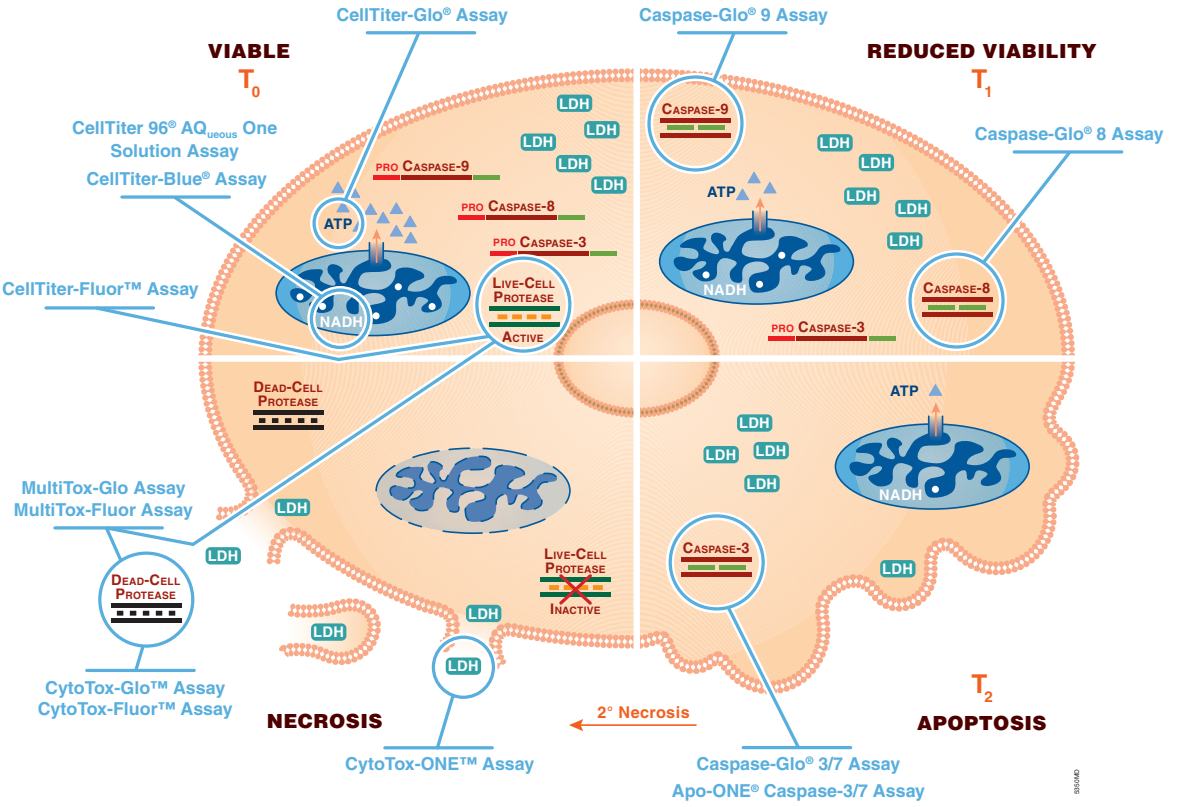


Comparison of RNA interference induced by siRNAs synthesized chemically and by in vitro transcription. Two different target luciferase sequences were synthesized by in vitro transcription using the T7 RiboMAX™ Express RNAi System (IVT #1 and #2) and synthesized chemically (Syn #1 and #2). After transfection using CodeBreaker™ Transfection Reagent, these siRNAs were evaluated for RNA interference in CHO cells stably expressing luciferase.

Cell Viability, Apoptosis and ADME/Tox Assays

Cell Proliferation and Cytotoxicity Assays	**58**
Oxidative Stress Assay	**68**
Apoptosis Detection Systems and Reagents	**69**
Proteases and Protease Assays	**75**
ADME Assays	**80**

Cell Proliferation and Cytotoxicity Assays



Assay Type	Parameter/Biomarker Measured	Time to Results	Sensitivity (*384 well)	Plate Format	Instrument
CellTiter-Glo® Assay	Viable cell ATP	10 minutes	10 viable cells*	96/384/1536	Luminometer/CCD
CellTiter-Fluor™ Assay	Live-cell protease	0.5–3 hours	40 viable cells	96/384/1536	Fluorometer AFC 400nm$_{Ex}$/505nm$_{Em}$
CellTiter-Blue® Assay	Resazurin reduction by NADH	1–4 hours	50 cells*	96/384/1536	Fluorometer, Resorufin 560nm$_{Ex}$/590nm$_{Em}$
CellTiter 96® AQueous One Solution Assay	MTS reduction by NADH	1–4 hours	200 cells*	96/384	Spectrophotometer Abs 490nm
MultiTox-Glo Assay	Viability and cytotoxicity by live- and dead-cell proteases	0.5 hour	40 viable cells, 10 dead cells	96/384/1536	Fluorometer AFC 400nm$_{Ex}$/505nm$_{Em}$ Luminometer
MultiTox-Fluor Assay	Viability and cytotoxicity by live- and dead-cell proteases	0.5–3 hours	40 live cells, 10 dead cells	96/384/1536	Fluorometer AFC 400nm$_{Ex}$/505nm$_{Em}$ R110 485nm$_{Ex}$/520nm$_{Em}$
CytoTox-Glo™ Assay	Dead-cell protease release	15 minutes	10 dead cells	96/384/1536	Luminometer
CytoTox-Fluor™ Assay	Dead-cell protease release	0.5–3 hours	10 dead cells	96/384	Fluorometer R110 485nm$_{Ex}$/520nm$_{Em}$
CytoTox-ONE™ Assay	LDH release	10 minutes	200 cells*	96/384	Fluorometer, Resorufin 560nm$_{Ex}$/590nm$_{Em}$
Caspase-Glo® 3/7 Assay	Caspase-3/7 activity	0.5 hour	20 cells*	96/384/1536	Luminometer
Apo-ONE® Caspase 3/7 Assay	Caspase-3/7 activity	1–18 hours	200 cells*	96/384/1536	Fluorometer R110 499nm$_{Ex}$/521nm$_{Em}$
Caspase-Glo® 8 Assay	Caspase-8 activity	0.5 hour	~1000 cells	96	Luminometer
Caspase-Glo® 9 Assay	Caspase-9 activity	0.5 hour	~1500 cells	96	Luminometer
Caspase-Glo® 2 Assay	Caspase-2 activity	0.5 hour	Purified enzyme	96/384/1536	Luminometer
Caspase-Glo® 6 Assay	Caspase-6 activity	0.5 hour	Purified enzyme	96/384/1536	Luminometer

9481LA

ApoTox-Glo™ Triplex Assay

Product	Size	Cat.#
ApoTox-Glo™ Triplex Assay	10 ml	G6320
	5 × 10 ml	G6321

For Laboratory Use. For in vitro use only. Cat.# G6320 contains sufficient reagents for 100 assays in 96-well format or 400 assays in 384-well format. Cat.# G6321 contains sufficient reagents for 500 assays in 96-well format or 2,000 assays in 384-well format.

Description: The ApoTox-Glo™ Triplex Assay combines three assay chemistries to easily assess viability, cytotoxicity and apoptosis events in the same cell-based assay well. First, viability and cytotoxicity are determined by measuring two differential protease biomarkers simultaneously with the addition of a single nonlytic reagent containing two peptide substrates. The live-cell protease activity is restricted to intact viable cells and is measured using a fluorogenic, cell-permeant peptide substrate (GF-AFC Substrate). The substrate enters intact cells, where it is cleaved to generate a fluorescent signal proportional to the number of living cells. This live-cell protease activity marker becomes inactive upon loss of membrane integrity and leakage into the surrounding culture medium. A second, cell-impermeant, fluorogenic peptide substrate (bis-AAF-R110 Substrate) is used simultaneously to measure dead-cell protease activity that has been released from cells that have lost membrane integrity. This results in ratiometric, inversely correlated measures of cell viability and cytotoxicity. A second reagent containing luminogenic DEVD-peptide substrate for caspase-3/7 and Ultra-Glo™ Recombinant Thermostable Luciferase is added. Caspase-3/7 cleavage of the substrate releases luciferin, which is a substrate for luciferase and generates light. The light output, measured with a luminometer, correlates with caspase-3/7 activation as a key indicator of apoptosis.

Features:

- **Measure Viability, Cytotoxicity and Apoptosis in the Same Well:** Determine mechanism of cell death for cells in the same well.
- **Easily Implement:** Assay follows a simple "add-mix-measure" format.
- **Normalize Data with a Built-In Control:** The ratio of live cells/dead cells is independent of cell number and normalizes data.
- **Flexible and Easily Automated:** The volume of each assay component can be scaled to meet throughput needs, and the assay is amenable to automation in 96- and 384-well plates.
- **Improves Efficiency and Saves on Lab Budget:** Reduces cell culture and labor costs by performing three assays in a single well.

Protocol	Part#
Technical Manual	TM322

Storage Conditions: Store all components at –20°C protected from light.

Mechanism of Toxicity Determination

Product	Size	Cat.#
ApoLive-Glo™ Multiplex Assay	10 ml	G6410
	5 × 10 ml	G6411

For Laboratory Use.

Description: The ApoLive-Glo™ Multiplex Assay measures both the number of viable cells as a marker of cytotoxicity and caspase activation as a marker of apoptosis within a single assay well. The first part of the assay measures the activity of a protease marker of cell viability. The live-cell protease activity is restricted to intact viable cells and is measured using a fluorogenic, cell-permeant, peptide substrate (glycyl-phenylalanyl-amino fluorocoumarin; GF-AFC). The substrate enters intact cells, where it is cleaved by the live-cell protease activity to generate a fluorescent signal proportional to the number of living cells. This live-cell protease becomes inactive upon loss of cell membrane integrity and leakage into the surrounding culture medium. The second part of the assay uses the Caspase-Glo® Assay technology to detect caspase activation, a key biomarker of apoptosis. The Caspase-Glo® Assay provides a luminogenic caspase-3/7 substrate, which contains the tetrapeptide sequence DEVD, in a reagent optimized for caspase activity, luciferase activity and cell lysis. Adding the Caspase-Glo® 3/7 Reagent results in cell lysis, followed by caspase cleavage of the substrate and generation of a "glow-type" luminescent signal. Luminescence is proportional to the amount of caspase activity present.

Features:

- **Measure Viability and Apoptosis in the Same Sample Well:** Accurately determine the mechanism of cell death in less time with less sample.
- **Easy to Implement:** The assay uses a simple sequential "add-mix-measure" format.
- **Normalize Caspase Data with Viability Control:** The ratio of caspase activity to viable cell is useful for determining the extent of caspase activation and for normalizing cell numbers.
- **Flexible and Easily Automated:** The volumes of each assay component can be scaled to meet throughput needs, and the assay is amenable to automation in 96- and 384-well plates.
- Reveal cell death even if the window of caspase activity is missed.
- **Multiplex with Other Assays:** The nonlytic nature of the first step of the assay allows further multiplexing with spectrally distinct fluorescent assay chemistries.

Protocol	Part#
Technical Manual	TM325

Storage Conditions: Store all components at –20°C protected from light.

Multiplexed Viability and Cytotoxicity Assays

Product	Size	Cat.#
MultiTox-Glo Multiplex Cytotoxicity Assay	10 ml	G9270
	5 × 10 ml	G9271
	2 × 50 ml	G9272

For Laboratory Use. Using 100µl/assay in 96-well format, Cat.# G9270 is sufficient for 100 assays; Cat.# G9271, 500 assays; Cat.# G9272, 1,000 assays. Using 25µl per well in 384-well format, Cat.# G9270 is sufficient for 400 assays; Cat.# G9271, 2,000 assays; Cat.# G9272, 4,000 assays.

Description: The MultiTox-Glo Multiplex Cytotoxicity Assay is a sequential-reagent-addition fluorescent and luminescent assay that measures the relative number of live and dead cells in cell populations. The assay sequentially measures two protease activities. The live-cell protease activity restricted to intact viable cells and is measured using a fluorogenic, cell-permeant, peptide substrate (GF-AFC). This substrate enters intact cells, where it is cleaved by the live cell protease activity to release AFC and generate a fluorescent signal proportional to the number of viable cells. The live-cell protease becomes inactive upon loss of membrane integrity and leakage into the surrounding culture medium. A second, luminogenic cell-impermeant peptide substrate (AAF-aminoluciferin) is used to measure dead-cell protease activity, which is released from cells that have lost membrane integrity. The liberated aminoluciferin product is measured as "glow type" luminescence generated by Ultra-Glo™ Recombinant Luciferase provided in the assay reagent.

Features:

- **Measure Live and Dead Cells in Culture:** Sequential-reagent-addition assay with a homogeneous "add-mix-measure" protocol.
- **Normalize Data with a Built-In Internal Control:** The ratio of live cells/dead cells is independent of cell number and can be used to normalize data.
- **Immediately Identify More False-Positives and False-Negatives:** Independent cell viability and cytotoxicity measurements serve as controls for each other. If test compounds interfere with one assay chemistry, the other serves as an internal control.
- **Improve your Data:** Reduce statistical probability of false-positives (or false-negatives), and eliminate fluorescence interference issues.

Protocol	Part#
Technical Bulletin	TB358

Storage Conditions: Store at –20°C, protected from light.

MultiTox-Fluor Multiplex Cytotoxicity Assay

Product	Size	Cat.#
MultiTox-Fluor Multiplex Cytotoxicity Assay	10 ml	G9200
	5 × 10 ml	G9201
	2 × 50 ml	G9202

For Laboratory Use. G9200 contains sufficient reagents for 100 assays at 100µl/assay in a 96-well format or 400 assays at 25µl/assay in a 384-well format. G9201 contains sufficient reagents for 500 assays at 100µl/assay in a 96-well format or 2,000 assays at 25µl/assay in a 384-well format. G9202 contains sufficient reagents for 1,000 assays at 100µl/assay in a 96-well format or 4,000 assays at 25µl/assay in a 384-well format.

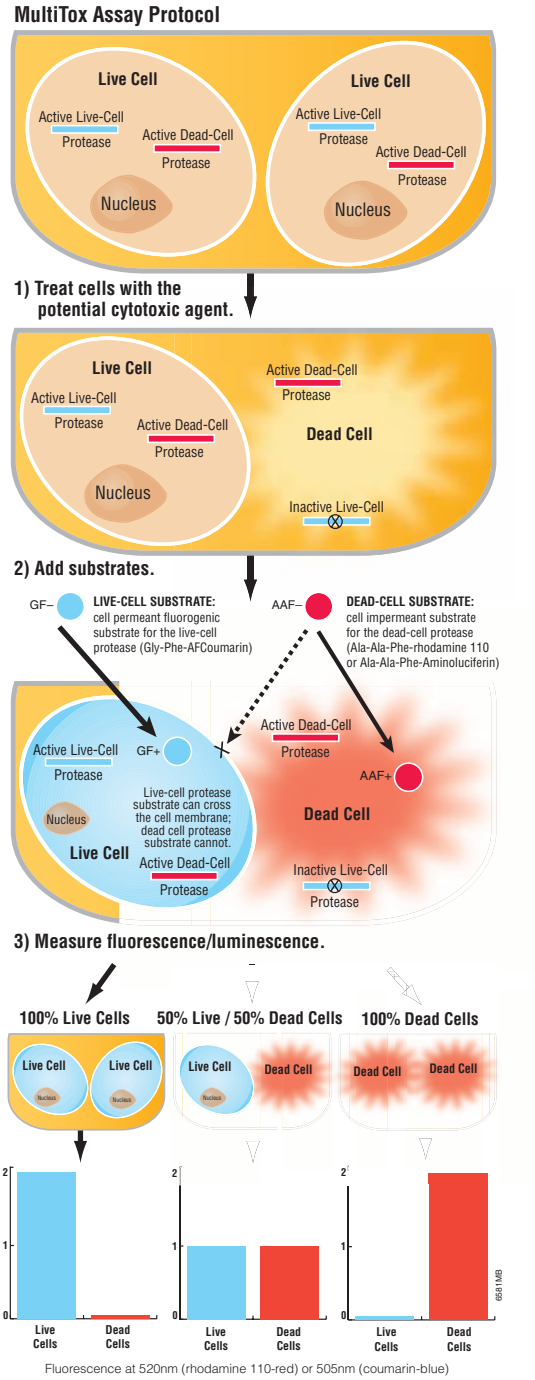
Description: The MultiTox-Fluor Multiplex Cytotoxicity Assay is a single-reagent-addition, homogeneous, fluorescent assay that measures the number of live and dead cells simultaneously in culture wells. The assay simultaneously measures cell viability and cytotoxicity by detecting two distinct protease activities. The live-cell protease activity is restricted to intact viable cells and is measured using a fluorogenic, cell-permeant peptide substrate (GF-AFC Substrate). The substrate enters intact cells where it is cleaved to generate a fluorescent signal proportional to the number of living cells. This live-cell protease activity marker becomes inactive upon loss of membrane integrity and leakage into the surrounding culture medium. A second, cell-impermeant, fluorogenic peptide substrate (bis-AAF-R110 Substrate) is used to measure dead-cell protease activity that has been released from cells that have lost membrane integrity.

Features:

- **Measure the Number of Live and Dead Cells in Culture:** Homogeneous, "add-mix-measure" protocol eliminates parallel plate processing and reduces cell culture costs.
- **Normalize Data for Cell Number:** The ratio of live to dead cells is independent of cell number and normalizes data. Data normalization for cell number makes results more comparable well-to-well, plate-to-plate, day-to-day.
- **Reduce False-Positive and -Negative Results:** Complementary live- and dead-cell measures with independent chemistries serve as internal controls for each other.
- **Get More Data from Every Well:** Multiplex the MultiTox-Fluor Assay with most Promega bioluminescent cell-based apoptosis or genetic reporter assays.
- **Reduce Assay Variability:** The homogeneous "add-mix-measure" protocol avoids the cumulative error associated with multistep protocols.

Protocol	Part#
Technical Bulletin	TB348

Storage Conditions: Store at –20°C.



Overview of the MultiTox-Fluor Multiplex Cytotoxicity Assay protocol.

Bioluminescent Cytotoxicity Assay

Product	Size	Cat.#
CytoTox-Glo™ Cytotoxicity Assay	10 ml	G9290
	5 × 10 ml	G9291
	2 × 50 ml	G9292

For Laboratory Use. Using 100µl/assay in a 96-well plate format, Cat.# G9290 is sufficient to perform 100 assays; Cat.# G9291, 500 assays; Cat.# G9292, 1,000 assays. Using 25µl per well in a 384-well plate format, Cat.# G9290 is sufficient to perform 400 assays; Cat.# G9291, 2,000 assays; Cat.# G9292, 4,000 assays.

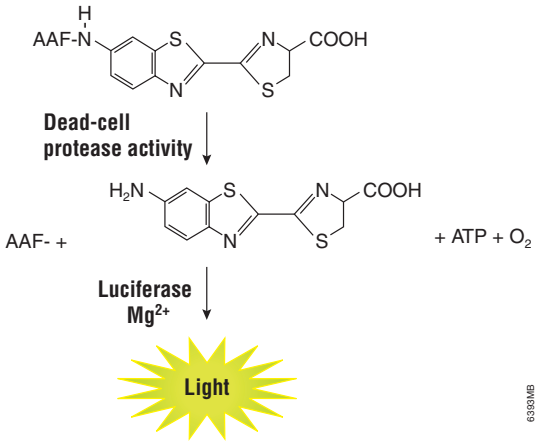
Description: The CytoTox-Glo™ Assay is a luminescent cytotoxicity assay that measures the relative number of dead cells in cell populations. The CytoTox-Glo™ Assay measures the extracellular activity of a distinct intracellular protease activity (dead-cell protease) when the protease is released from membrane-compromised cells. A luminogenic cell-impermeant peptide substrate (AAF-aminoluciferin) is used to measure dead-cell protease activity. The liberated aminoluciferin product is measured as "glow type" luminescence generated by Ultra-Glo™ Recombinant Luciferase provided in the assay reagent. The AAF-aminoluciferin substrate cannot cross the intact membrane of viable cells and does not generate any appreciable signal from the live-cell population. The amount of luminescence directly correlates with the percentage of cells undergoing cytotoxic stress. With the addition of a lysis reagent (provided), the CytoTox-Glo™ Assay also can deliver the luminescent signal associated with the total number of cells in each assay well. Viability can be calculated by subtracting the luminescent dead-cell signal from the total luminescent value, thus allowing you to normalize assay data to cell number and mitigate assay interferences that may lead to erroneous conclusions. The cytotoxicity protease biomarker is constitutive and conserved across cell lines, and the CytoTox-Glo™ Assay demonstrates excellent correlation with other methods of assessing cell viability.

Features:

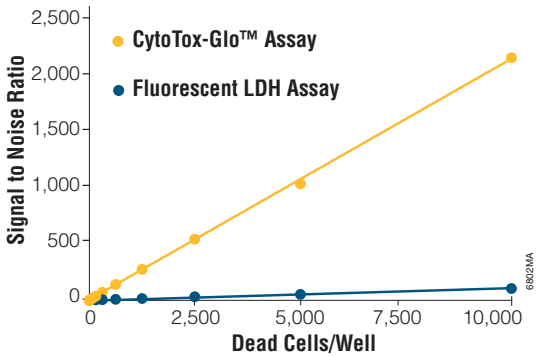
- **Measure the Relative Number of Dead Cells in Culture:** Measure cytotoxicity by adding a single reagent with the homogeneous "add-mix-measure" protocol.
- **Distinguish Between Small Differences in Viability:** The assay provides a linear response and can distinguish between small differences in viability across the entire spectrum of cytotoxicity, from modest cytotoxicity (100 to 95% viability) to profound cytotoxicity (5 to 0% viability).
- **Normalize Data for Cytotoxicity:** Data normalization for dead-cell number makes results more comparable well-to-well, plate-to-plate and day-to-day.
- **Measure the Relative Number of Remaining Viable Cells Using a Total Lysis Protocol:** Correlate increased cytotoxicity with a reduction in viable cells.
- **Improve your Data:** Reduce statistical probability of false-positives (or false-negatives), and eliminate fluorescence interference issues with a stable luminescence readout.

Protocol	Part#
Technical Bulletin	TB359

Storage Conditions: Store at –20°C, protected from light.



Cleavage of the luminogenic AAF-Glo™ Substrate by dead-cell protease activity.



Superior sensitivity and dynamic range of the CytoTox-Glo™ Assay compared to fluorescent LDH Assay.

CytoTox-Fluor™ Cytotoxicity Assay

Product	Size	Cat.#
CytoTox-Fluor™ Cytotoxicity Assay	10 ml	G9260
	5 × 10 ml	G9261
	2 × 50 ml	G9262

For Laboratory Use. G9260 contains sufficient reagents for 100 assays at 100µl/assay in a 96-well format or 400 assays at 25µl/assay in a 384-well format. G9261 contains sufficient reagents for 500 assays at 100µl/assay in a 96-well format or 2,000 assays at 25µl/assay in 384-well format. G9262 contains sufficient reagents for 1,000 assays at 100µl/assay in a 96-well format or 4,000 assays at 25µl/assay in a 384-well format.

Description: The CytoTox-Fluor™ Cytotoxicity Assay is a single-reagent-addition, homogeneous, fluorescent assay that measures the relative number of dead cells in cell populations. The assay measures a distinct protease activity associated with cytotoxicity and uses a fluorogenic peptide substrate (bis-alanyl-alanyl-phenylanlanyl-rhodamine 110; bis-AAF-R110) to measure "dead-cell activity," which has been released from cells that have lost membrane intergrity. The bis-AAF-R110 substrate cannot cross the intact membrane of live cells and therefore gives no signal from live cells. The assay is designed to accommodate downstream multiplexing with several Promega luminescent assays or spectrally distinct fluorescent assay methods, such as assays to measure caspase activation, reporter gene expression or orthogonal measures of viability.

Features:

- **Measure the Relative Number of Dead Cells in Culture:** Homogeneous, "add-mix-measure" protocol eliminates parallel plate processing and reduces cell culture costs.
- **Get More Data from Every Well:** Multiplex the CytoTox-Fluor™ Assay with several Promega luminescent cell-based assays.
- **Normalize Downstream Multiplex Data for Cytotoxicity:** Data normalization for dead-cell number makes results more comparable well-to-well, plate-to-plate, day-to-day.
- **Reduce Assay Variability:** The homogeneous "add-mix-measure" protocol avoids the cumulative error associated with multistep protocols.

Protocol	Part#
Technical Bulletin	TB350

Storage Conditions: Store at –20°C.

Fluorescent Cell Viability Assay

Product	Size	Cat.#
CellTiter-Fluor™ Cell Viability Assay	10 ml	G6080
	5 × 10 ml	G6081
	2 × 50 ml	G6082

For Laboratory Use.

Description: The CellTiter-Fluor™ Cell Viability Assay is a nonlytic, single-reagent-addition fluorescence assay that measures the relative number of viable cells in a population. The assay is based on measurement of a conserved and constitutive protease activity within live cells that serves as a biomarker of cell viability. The live-cell protease activity is restricted to intact viable cells and is measured using a fluorogenic, cell-permeant, peptide substrate (Gly-Phe-AFC). The substrate enters intact cells, where it is cleaved by the live-cell protease activity to generate a fluorescent signal proportional to the number of living cells. The live-cell protease becomes inactive upon loss of membrane integrity and leakage into the surrounding culture medium.

The CellTiter-Fluor™ Assay also can be used in a single-well, sequential, multiplex format with other downstream assay chemistries to normalize data by cell number. Data from the assay can serve as an internal control and allow identification of errors resulting from cell clumping or compound cytotoxicity. The assay is compatible with many Promega luminescence assays or spectrally distinct fluorescence assay methods, such as measuring caspase activation, reporter gene expression or orthogonal measures of viability.

Features:

- **Obtain Better Data from Every Well:** The assay can be performed in multiplex with many Promega luminescence assays or spectrally distinct fluorescence assays.
- **Normalize Data for Cell Number:** Normalizing data for live-cell number makes results more comparable well-to-well, plate-to-plate, day-to-day.
- **Save on Cell Culture Costs:** Multiplexing assays in the same well eliminates parallel plate processing, thus reducing cell culture costs.

Protocol	Part#
Technical Bulletin	TB371

Storage Conditions: Store at –20°C.

CellTiter-Glo® Luminescent Cell Viability Assay

Product	Size	Cat.#
CellTiter-Glo® Luminescent Cell Viability Assay	10 ml	G7570
	10 × 10 ml	G7571
	100 ml	G7572
	10 × 100 ml	G7573

For Laboratory Use. Using 100µl of CellTiter-Glo® Reagent per assay in a 96-well format, Cat.# G7570 is sufficient to perform 100 assays; Cat.# G7571 and G7572, 1,000 assays; Cat.# G7573, 10,000 assays. Using 25µl of CellTiter-Glo® Reagent per assay in a 384-well format, Cat.# G7570 is sufficient to perform 400 assays; Cat.# G7571 and G7572, 4,000 assays; Cat.# G7573, 40,000 assays.

Description: The CellTiter-Glo® Luminescent Cell Viability Assay is a homogeneous method of determining the number of viable cells in culture based on quantitation of the ATP present, an indicator of metabolically active cells. The CellTiter-Glo® Assay is designed for use with multiwell formats, making it ideal for automated high-throughput screening (HTS), cell proliferation and cytotoxicity assays. The homogeneous assay procedure involves adding the single reagent (CellTiter-Glo® Reagent) directly to cells cultured in serum-supplemented medium. Cell washing, removal of medium and multiple pipetting steps are not required. The system detects as few as 15 cells/well in a 384-well format in 10 minutes after adding reagent and mixing.

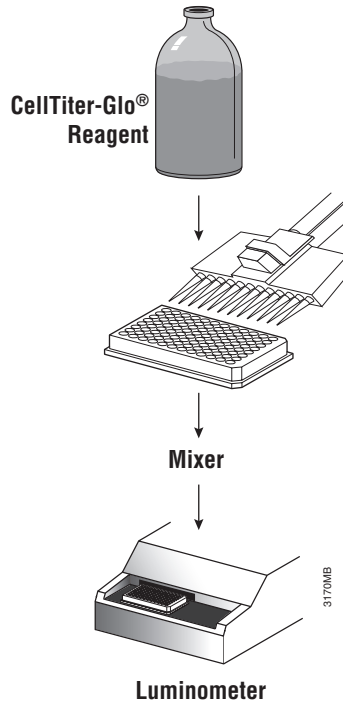
The homogeneous "add-mix-measure" format results in cell lysis and generation of a luminescent signal proportional to the amount of ATP present. The amount of ATP is directly proportional to the number of cells present in culture. The CellTiter-Glo® Assay generates a "glow-type" luminescent signal, which has a half-life generally greater than five hours, depending on cell type and medium used. The extended half-life eliminates the need to use reagent injectors and provides flexibility for continuous or batch mode processing of multiple plates. The unique homogeneous format avoids errors that may be introduced by other ATP measurement methods that require multiple steps.

Features:

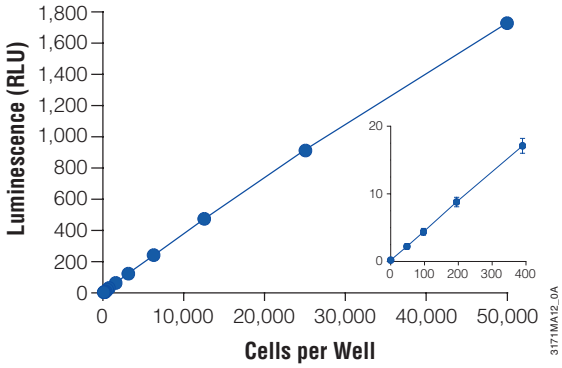
- **Simplify Cell Viability Assays:** Homogeneous "add-mix-measure" format dramatically reduces the number of plate handling steps required for similar assays.
- **Use Fewer Cells:** Detects as few as 15 cells/well in a 384-well format or 50 cells/well in a 96-well format. Accurately measures cells at numbers below the detection limits of standard colorimetric and fluorometric assays. Reduces the number of cells required per assay.
- **Get Results Quickly:** Data can be recorded 10 minutes after adding reagent.
- **Choose Your Format:** Can be used with various multiwell formats. Data can be recorded by luminometer or CCD camera imaging device.
- **Process Plates Consecutively:** Luminescent signal is very stable, with a half-life generally >5 hours, dependent on cell type and medium used, allowing batch processing; delivers excellent Z′-factor values for screening applications.
- **Get More Information:** Multiplex with other cell-based assays from Promega.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB288

Storage Conditions: For long-term storage, the lyophilized CellTiter-Glo® Substrate and CellTiter-Glo® Buffer should be stored at –20°C. Reconstituted CellTiter-Glo® Reagent can be stored at 4°C for 48 hours with ~5% loss of activity or at 4°C for 4 days with ~20% loss of activity.



Flow diagram showing preparation and use of CellTiter-Glo® Reagent.



Excellent sensitivity and extended linearity. Serial twofold dilutions of Jurkat cells were made in RPMI 1640 and 10% PBS in a 96-well plate. The assay was performed as described in Technical Bulletin #TB288. Values represent the mean ± S.D.of four replicates for each cell number.

BacTiter-Glo™ Microbial Cell Viability Assay

Product	Size	Cat.#
BacTiter-Glo™ Microbial Cell Viability Assay	10 ml	G8230
	10 × 10 ml	G8231
	100 ml	G8232
	10 × 100 ml	G8233

For Laboratory Use. Using 100µl of BacTiter-Glo™ Reagent per assay in a 96-well format, Cat.# G8230 provides sufficient reagents to perform 100 assays; Cat.# G8231 and G8232, 1,000 assays; Cat.# G8233, 10,000 assays. Using 25µl of BacTiter-Glo™ Reagent per assay in a 384-well format, Cat.# G8230 provides sufficient reagents to perform 400 assays; Cat.# G8231 and G8232, 4,000 assays; Cat.# G8233, 40,000 assays.

Description: The BacTiter-Glo™ Microbial Cell Viability Assay is a homogeneous method for determining the number of viable microbial cells in culture based on quantitation of the ATP present. ATP is an indicator of metabolically active cells. The homogeneous assay procedure involves adding a single reagent (BacTiter-Glo™ Reagent) directly to bacterial cells cultured in medium and measuring luminescence. The homogeneous format reduces pipetting errors that may be introduced during the multiple steps required by other methods of ATP measurement. The formulation of the reagent supports bacterial cell lysis and generation of a luminescent signal in a homogeneous "add-mix-measure" format. The luminescent signal is proportional to the amount of ATP present, which is directly proportional to the number of viable cells in culture. The assay relies on the properties of a proprietary thermostable luciferase (Ultra-Glo™ Recombinant Luciferase) and a proprietary buffer formulation for extracting ATP from bacteria. The assay has been shown to detect a variety of bacteria and fungi.

Features:

- **Simplify Microbial Detection:** The "add-mix-measure" format reduces the number of handling steps to fewer than that required for similar ATP assays, with no separate lysis step, and no injectors required, allowing easy automation.
- **Get Results Quickly:** Data can be recorded in 5 minutes or less after adding reagent and mixing. Superior sensitivity allows you to detect growth or toxicity quickly after inoculation.
- **Increase Your Sensitivity:** Measure ATP from as few as 10 bacterial cells, 1,000-fold more sensitive than absorbance (O.D.) readings.
- **Choose Your Format:** Can be used with various multiwell-plate or single-use formats. Data can be recorded by luminometer or CCD camera.
- **Process Plates Consecutively:** The "glow-type" luminescent signal is stable, with a half-life generally over 30 minutes.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB337

Storage Conditions: For long-term storage, the lyophilized BacTiter-Glo™ Substrate and BacTiter-Glo™ Buffer should be stored at –20°C.

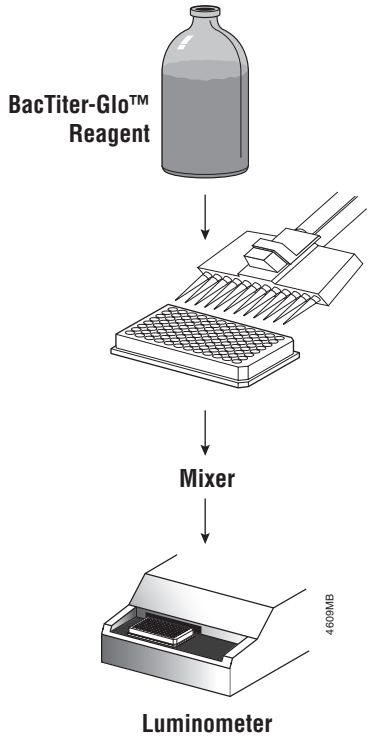
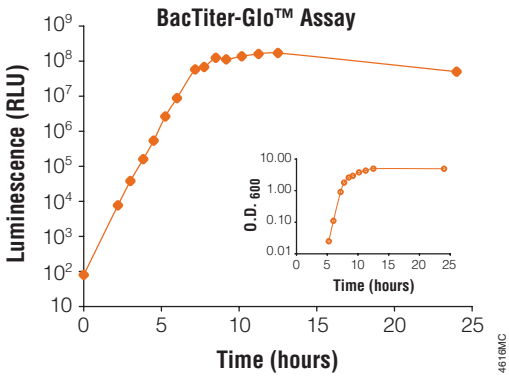


Diagram of the BacTiter-Glo™ Microbial Cell Viability Assay protocol.



Evaluate bacterial growth immediately after inoculation using the BacTiter-Glo™ Assay. When measuring growth by O.D., the first significant measurement (0.25 O.D.with *E.coli*) did not occur until 5 hours after inoculation.

CellTiter 96® AQ_{ueous} One Solution Cell Proliferation Assay (MTS)

Product	Size	Cat.#
CellTiter 96® AQ_{ueous} One Solution Cell Proliferation Assay	200 assays	G3582
	1,000 assays	G3580
	5,000 assays	G3581

For Laboratory Use.

Description: The CellTiter 96® AQ_{ueous} One Solution Cell Proliferation Assay is a colorimetric method for determining the number of viable cells in proliferation, cytotoxicity or chemosensitivity assays. The CellTiter 96® AQ_{ueous} One Solution Reagent contains a tetrazolium compound [3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium, inner salt; MTS] and an electron coupling reagent (phenazine ethosulfate; PES). PES has enhanced chemical stability, which allows it to be combined with MTS to form a stable solution. The CellTiter 96® AQ_{ueous} Assay uses phenazine methosulfate (PMS) as the electron coupling reagent, and PMS Solution and MTS Solution are supplied separately.

Assays are performed by adding a small amount of the CellTiter 96® AQ_{ueous} One Solution Reagent directly to culture wells, incubating for 1–4 hours and then recording absorbance at 490nm with a 96-well plate reader. The quantity of formazan product as measured by the amount of 490nm absorbance is directly proportional to the number of living cells in culture.

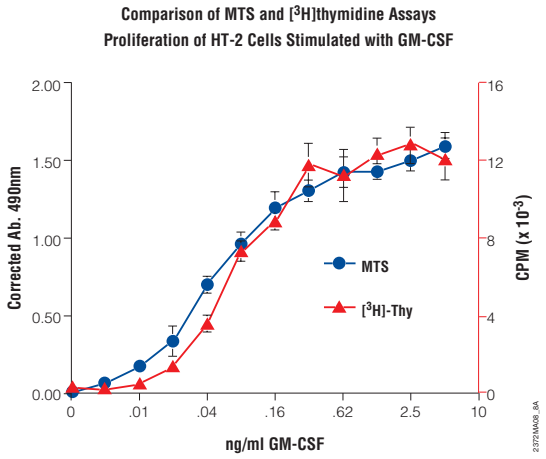
If you currently use a [^{3}H]-thymidine incorporation assay, addition of the CellTiter 96® AQ_{ueous} One Solution Reagent can be substituted for the pulse of [^{3}H]-thymidine at the time point in the assay when the pulse of radioactive thymidine is usually added. Previous bioassay data comparing [^{3}H]-thymidine incorporation to the MTS-based CellTiter 96® AQ_{ueous} Assay and the original MTT-based CellTiter 96® Assay demonstrate that tetrazolium reagents can be substituted for [^{3}H]-thymidine incorporation.

Features:

- **Simplify Colorimetric Viability Assays:** "Add-incubate-measure" format (single-step reagent addition) enables design of homogeneous high-throughput screening assays.
- **Use a Single Solution:** Use as a single solution, filter sterilized and ready to add to assay plates (unlike MTT).
- **Perform Fewer Steps:** Perform the assay in 96-well plates with no washing or cell harvesting. Also eliminates solubilization steps normally required for MTT assays.
- **Gain Flexibility:** Plates can be read and returned to incubator for further color development (unlike MTT).
- **Avoid Organic Solvents:** Requires no volatile organic solvent to solubilize the formazan product (unlike MTT).
- **Non-Radioactive:** Requires no scintillation cocktail or radioactive waste disposal (unlike [^{3}H]-thymidine incorporation assays).
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB245

Storage Conditions: Store at –20°C, protected from light.



Measurement of GM-CSF-stimulated proliferation in HT-2 cells using the CellTiter 96® AQ_{ueous} Cell Proliferation Assay and a [^{3}H]thymidine incorporation assay. Similar results were obtained with both assays.

CellTiter 96® AQ_{ueous} Non-Radioactive Cell Proliferation Assay (MTS)

Product	Size	Cat.#
CellTiter 96® AQ_{ueous} Non-Radioactive Cell Proliferation Assay	1,000 assays	G5421
	5,000 assays	G5430
	50,000 assays	G5440
Available Separately	**Size**	**Cat.#**
CellTiter 96® AQ_{ueous} MTS Reagent Powder	1 g	G1111
	250 mg	G1112

For Laboratory Use.

Description: The CellTiter 96® AQ_{ueous} Non-Radioactive Cell Proliferation Assay is a homogeneous, colorimetric method for determining the number of viable cells in proliferation, cytotoxicity or chemosensitivity assays. The CellTiter 96® AQ_{ueous} Assay is composed of solutions of a novel tetrazolium compound [3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium, inner salt; MTS] and an electron coupling reagent (phenazine methosulfate) PMS. MTS is bioreduced by cells into a formazan product that is soluble in tissue culture medium. The absorbance of the formazan product at 490nm can be measured directly from 96-well assay plates without additional processing. The conversion of MTS into the aqueous soluble formazan product is accomplished by dehydrogenase enzymes found in metabolically active cells. The quantity of formazan product as measured by the amount of 490nm absorbance is directly proportional to the number of living cells in culture.

If you currently use a [^{3}H]-thymidine incorporation assay, addition of the combined MTS/PMS solution can be substituted for [^{3}H]-thymidine at the time point in the assay when the pulse of radioactive thymidine is usually added. Data from proliferation bioassays comparing the CellTiter 96® AQ_{ueous} Assay and [^{3}H]-thymidine incorporation show similar results. This is in agreement with similar radioactivity incorporation studies performed using the original CellTiter 96® Assay.

CellTiter 96® AQ_{ueous} MTS Reagent Powder is a novel tetrazolium compound for use in colorimetric assays for determining the number of viable cells in proliferation, cytotoxicity or chemosensitivity assays. It is provided in powdered form.

Features:

- **Easy to Use:** Combine provided MTS and PMS solutions, add to cells, incubate and read absorbance.
- **Fast:** Perform the assay in a 96-well plate with no washing or cell harvesting. Also eliminates solubilization steps because the MTS formazan product is soluble in tissue culture medium.
- **Non-Radioactive:** Requires no scintillation cocktail or radioactive waste disposal (unlike [^{3}H]-thymidine).
- **Flexible:** Plates can be read and returned to incubator for further color development (unlike MTT).
- **Safe:** Requires no volatile organic solvent to solubilize the formazan product (unlike MTT).

Protocol	Part#
Technical Bulletin	TB169

Storage Conditions: For long-term storage, store MTS and PMS Solutions at –20°C, protected from light.

CellTiter 96® Non-Radioactive Cell Proliferation Assay (MTT)

Product	Size	Cat.#
CellTiter 96® Non-Radioactive Cell Proliferation Assay	1,000 assays	G4000
	5,000 assays	G4100

For Laboratory Use.

Description: The CellTiter 96® Assay is a collection of qualified reagents that provide a convenient method of determining viable cell number. The CellTiter 96® Assay is a modification of the MTT assay method described by Mosmann and incorporates several improvements to the method that address previous technical problems including: 1) serum protein precipitation caused by adding organic solvent; 2) interference by phenol red; 3) incomplete solubilization of the formazan crystals resulting in lower sensitivity; and 4) stability of the colored product.

The CellTiter 96® Assay is performed by adding a premixed, optimized Dye Solution to culture wells of a 96-well plate, usually containing various concentrations of growth factor or test substance. During a 4-hour incubation, living cells convert the MTT tetrazolium component of the Dye Solution into a formazan product. If you currently use a [^{3}H]-thymidine incorporation assay, the addition of Dye Solution can be substituted for the pulse of radioactive thymidine at the time point in the assay when the pulse of [^{3}H]-thymidine is usually added. The Solubilization/Stop Solution is then added to the culture wells to solubilize the formazan product, and the absorbance at 570nm is recorded using a 96-well plate reader. In addition, direct comparison between [^{3}H]-thymidine incorporation and tetrazolium conversion have demonstrated less than a 5% difference between the two assays for determination of growth factor content of several samples.

Features:

- **Gain Sensitivity:** Detect as few as 1,000 cells/well with a 96-well plate reader. Greater sensitivity than the neutral red assay procedure.
- **Use a Variety of Cells:** Assay mammalian, plant and yeast cells.
- **Non-Radioactive:** Requires no scintillation cocktail or radioactive waste disposal.
- **Save Time:** Perform the assay in a 96-well plate with no washing steps, no cell harvesting and no scintillation counting.
- **Adapt to Your Needs:** Follow either a 4-hour or overnight protocol.
- **Convenient:** Requires no weighing or mixing of dye components.

Protocol	Part#
Technical Bulletin	TB112

Storage Conditions: Store Dye Solution at –20°C and Solubilization/Stop Solution at room temperature.

CellTiter-Blue® Cell Viability Assay

Product	Size	Cat.#
CellTiter-Blue® Cell Viability Assay	20 ml	G8080
	100 ml	G8081
	10 × 100 ml	G8082

Each G8080 system provides sufficient reagents to perform 1,000 assays in a 96-well format or 4,000 assays in a 384-well format when the recommended volumes are used. Likewise, each G8081 system provides sufficient reagents to perform 5,000 and 20,000 assays; each G8082 system provides sufficient reagents to perform 50,000 and 200,000 assays.

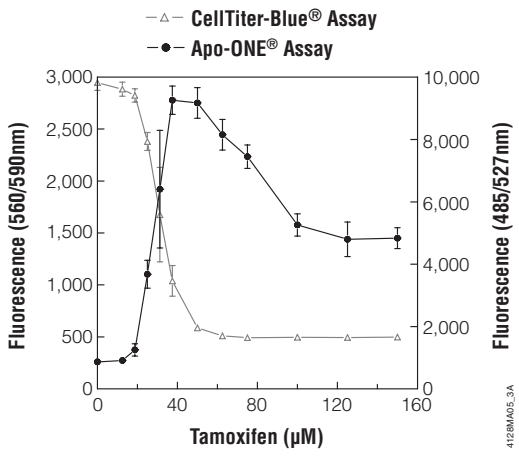
Description: The CellTiter-Blue® Cell Viability Assay provides a homogeneous, fluorescent method for monitoring cell viability. The assay is based on the ability of living cells to convert a redox dye (resazurin) into a fluorescent end product (resorufin). Nonviable cells rapidly lose metabolic capacity and do not generate a fluorescent signal. The homogeneous assay procedure involves adding the single reagent directly to cells cultured in serum-supplemented medium. After an incubation step, data are recorded using either a plate-reading fluorometer (preferred) or spectrophotometer.

Features:

- **Save Time:** The add-incubate-measure format reduces handling steps.
- **Perform More Than One Assay on the Same Sample:** The system can be multiplexed with other assays such as the Apo-ONE® Homogeneous Caspase-3/7 Assay (Cat.# G7790) or the Caspase-Glo® Assays (Cat.# G8090, G8200, G8210) for detecting apoptosis.
- **Gain Flexibility:** The CellTiter-Blue® Assay has an excellent Z′ factor value and offers more flexibility in assay incubation times compared to other resazurin-based assays.
- **Safe:** The reagent is generally nontoxic to cells, allowing extended incubation periods in some situations. Requires no scintillation cocktail, radioactive waste disposal (unlike [^{3}H]-thymidine incorporation assays) or hazardous solvents (as required for MTT tetrazolium-based assays).
- **Adapt to Your Throughput Needs:** The reagent is designed to provide sufficient volumes for accurate pipetting into 96- or 384-well formats. Convenient product sizes available for high-throughput screening.
- **Automate This Assay:** Validated automated methods available at: **www. promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB317

Storage Conditions: Store frozen at –20°C protected from light.



Multiplexing cell-based assays. Collecting viability data (CellTiter-Blue® Assay) and apoptosis data (Apo-ONE® Caspase-3/7 Assay) from the same wells.

CytoTox-ONE™ Homogeneous Membrane Integrity Assay

Product	Size	Cat.#
CytoTox-ONE™ Homogeneous Membrane Integrity Assay	200–800 assays	G7890
	1,000–4,000 assays	G7891
CytoTox-ONE™ Homogeneous Membrane Integrity Assay, HTP	1,000–4,000 assays	G7892

Description: The CytoTox-ONE™ Homogeneous Membrane Integrity Assay is a fluorometric method for estimating the number of nonviable cells present in multiwell plates by rapidly measuring the release of lactate dehydrogenase (LDH) from cells with a damaged membrane. Released LDH is measured with a 10-minute coupled enzymatic assay that results in the conversion of resazurin into a fluorescent resorufin product. The amount of fluorescence produced is proportional to the number of lysed cells. The CytoTox-ONE™ Reagent does not damage normal healthy cells; therefore the reactions to measure released LDH can be performed directly in assay wells containing a mixed population of viable and damaged cells.

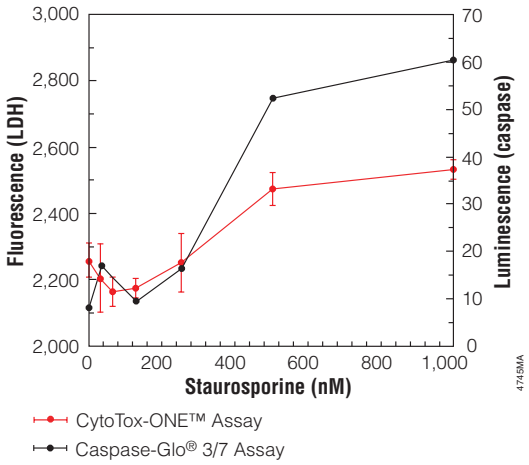
The CytoTox-ONE™ Homogeneous Membrane Integrity Assay, HTP (Cat.# G7892), offers alternative packaging for processing multiple plates. Each bottle of reagent supplied with the system is sufficient to perform 500 assays in a 96-well format or 2,000 assays in a 384-well format when the recommended volumes are used.

Features:

- **Save Time:** Complete the assay in the cell culture plate; the plates are incubated for 10 minutes, compared to 30 minutes or more with classic LDH assays.
- **Multiplex This Assay:** Perform multiple assays on one sample.
- **Adapt Protocol to Your Needs:** Completed assays can be read over several hours after the provided stop solution has been added.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB306

Storage Conditions: Store at –20°C protected from light.



Multiplexing the CytoTox-ONE™ Assay and the Caspase-Glo® 3/7 Assay. With most in vitro apoptosis assays, LDH release occurs relatively late during the process. The duration of drug exposure here was carefully chosen to demonstrate the early stages of cell lysis, while still retaining caspase activity.

CytoTox 96® Non-Radioactive Cytotoxicity Assay

Product	Size	Cat.#
CytoTox 96® Non-Radioactive Cytotoxicity Assay	1,000 assays	G1780

Description: The CytoTox 96® Non-Radioactive Cytotoxicity Assay is a colorimetric alternative to radioactive cytotoxicity assays. The CytoTox 96® Assay quantitatively measures lactate dehydrogenase (LDH), a stable cytosolic enzyme that is released upon cell lysis, in much the same way as [^{51}Cr] is released in radioactive assays. Released LDH in culture supernatants is measured with a 30-minute coupled enzymatic assay that results in the conversion of a tetrazolium salt (INT) into a red formazan product. The amount of color formed is proportional to the number of lysed cells. Visible wavelength absorbance data are collected using a standard 96-well plate reader. The assay can be used to measure membrane integrity for cell-mediated cytotoxicity assays in which a target cell is lysed by an effector cell or to measure lysis of target cells by bacteria, viruses, proteins, chemicals, etc.

Features:

- **Non-Radioactive:** Requires no radioactive waste disposal or [^{51}Cr].
- **Save Time:** Eliminates labeling of target cells prior to experiment.
- **Use Standard Equipment:** Collect absorbance (visible wavelength) data with a standard 96-well plate reader.
- **Adapt to Your Needs:** Used for a variety of applications including measurement of: 1) cell-mediated cytotoxicity; 2) chemical-mediated cytotoxicity; and 3) total cell number.
- **Gain Sensitivity:** Can reveal early, low-level damage to cell membranes that is often missed with other methodologies.

Protocol	Part#
Technical Bulletin	TB163

Storage Conditions: Store Substrate Mix and Assay Buffer at –20°C. Store LDH Positive Control, Lysis Solution (10X) and Stop Solution at 4°C.

Glutathione Quantitation

Product	Size	Cat.#
GSH-Glo™ Glutathione Assay	10 ml	V6911
	50 ml	V6912

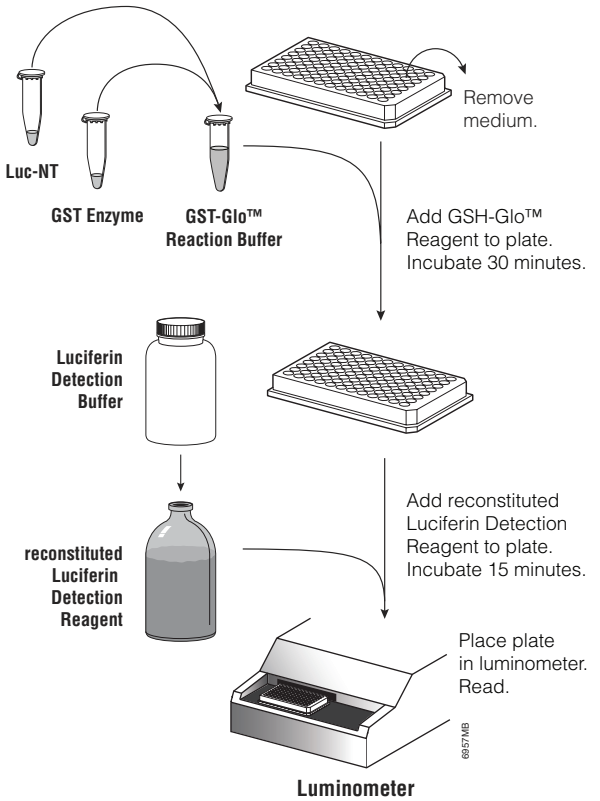
Description: The GSH-Glo™ Assay is a luminescent-based assay for the detection and quantification of glutathione (GSH) in cells or in various biological samples. A change in GSH levels is important in assessment of toxicological responses and is an indicator of oxidative stress, potentially leading to apoptosis or cell death. The assay is based on the conversion of a luciferin derivative into luciferin in the presence of GSH. The reaction is catalyzed by a glutathione S-transferase (GST) enzyme supplied in the kit. The luciferin formed is detected in a coupled reaction using Ultra-Glo™ Recombinant Luciferase that generates a glow type luminescence that is proportional to the amount of glutathione present in cells. The assay provides a simple, fast and sensitive alternative to colorimetric and fluorescent methods and can be adapted easily to high-throughput applications.

Features:

- **Fast:** Results in as little as 30 minutes.
- **Simplified Method:** The simple two-reagent-addition assay minimizes the number of assay steps compared to conventional GSH assays and is adapted easily to higher throughput applications. No deproteination step required!
- **Greater Sensitivity:** The luminescent method avoids inherent background fluorescence associated with other methods thereby providing excellent signal-to-background ratios.
- **Stable Signal:** Half-life greater than 5 hours.

Protocol	Part#
Technical Bulletin	TB369

Storage Conditions: Store at –20°C protected from light.



Schematc showing GSH-Glo™ Assay procedure.

Caspase-Glo® 2 Assay Systems

Product	Size	Cat.#
Caspase-Glo® 2 Assay	10 ml	G0940
	50 ml	G0941

For Laboratory Use. Cat.# G0940 provides sufficient reagents for 100 assays at 100µl/assay or 200 assays at 50µl/assay in 96-well plates or 400 assays at 25µl/assay in 384-well plates. Cat.# G0941 provides sufficient reagents for 500 assays at 100µl/assay or 1,000 assays at 50µl/assay in 96-well plates or 2,000 assays at 25µl/assay in 384-well plates.

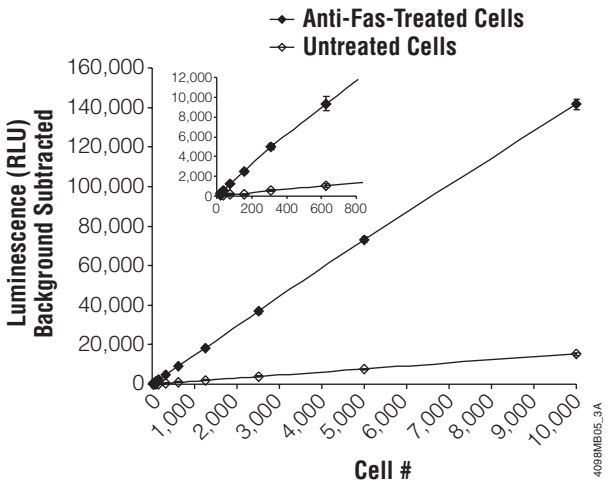
Description: The Caspase-Glo® 2 Assay is a homogeneous, luminescent assay that measures caspase-2 activity. Caspase-2 is a member of the cysteine aspartic acid-specific protease family. The Caspase-Glo® 2 Assay provides a luminogenic substrate (Z-VDVAD-aminoluciferin) in a reagent optimized for caspase-2 and luciferase activity. A single reagent is added to test samples, resulting in caspase cleavage of the substrate and generation of a glow-type luminescent signal produced by luciferase. Luminescence is proportional to the amount of caspase activity present. The assay system may be used with purified enzyme preparations and is ideal for automated high-throughput screening of inhibitors.

Features:

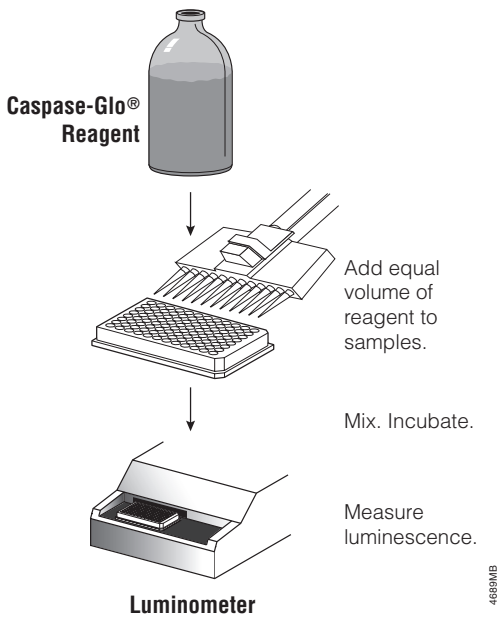
- **Broad Dynamic Range:** The assay is linear over four logs of caspase-2 concentration and can detect caspase-2 activity at concentrations as low as 0.2mU/ml.
- **High-Quality Assay:** The assay demonstrates an excellent Z′-factor value of 0.85 in 384-well plates using 0.05U/ml of enzyme.
- **Increased Accuracy:** The superior sensitivity over fluorescence-based caspase assays allows inhibitor studies to be performed below the K_m.
- **Batch Processing Capability:** The coupled-enzyme, homogeneous format results in a continuous signal, providing excellent stability and allowing plates to be read over an extended period of time. Luminometers with reagent injectors are not required.

Protocol	Part#
Technical Bulletin	TB365

Storage Conditions: Store at –20°C.



The Caspase-Glo® 3/7 Assay produces luminescence that is linear over a broad range of cell numbers. Jurkat cells were treated with anti-Fas mAb for 4.5 hours to induce apoptosis or were left untreated.Caspase-Glo® 3/7 Reagent was added directly to the cells in 96-well plates and incubated for 1 hour before recording luminescence. Each point represents the average of 4 wells. The "no cell" blank control value has been subtracted from each.



Schematic diagram of the Caspase-Glo® Assay protocol.

Caspase-Glo® 6 Assay Systems

Product	Size	Cat.#
Caspase-Glo® 6 Assay	10 ml	G0970
	50 ml	G0971

For Laboratory Use. Cat.# G0970 provides sufficient reagents for 100 assays at 100µl/assay or 200 assays at 50µl/assay in 96-well plates or 400 assays at 25µl/assay in 384-well plates. Cat.# G0971 provides sufficient reagents for 500 assays at 100µl/assay or 1,000 assays at 50µl/assay in 96-well plates or 2,000 assays at 25µl/assay in 384-well plates.

Description: The Caspase-Glo® 6 Assay is a homogeneous, luminescent assay that measures caspase-6 acitivity. Caspase-6 is a member of the cysteine aspartic acid-specific protease family and has a key effector role in the cleavage of specific target proteins during apoptosis. The Caspase-Glo® 6 Assay provides a luminogenic substrate, Z-VEID-aminoluciferin, in a buffer optimized for caspase-6 and luciferase activity. The addition of a single Caspase-Glo® 6 Reagent in an add-mix-measure format results in cleavage of the substrate, releasing aminoluciferin, and generation of a glow-type luminescent signal in the presence of Ultra-Glo™ Recombinant Luciferase. The luminescent signal is proportional to the amount of caspase-6 activity present. The homogeneous Caspase-Glo® 6 Assay is designed for use with purified enzyme preparations in multiwell plate formats, making it ideal for automated high-throughput screening for caspase-6 activity and inhibitors of caspase-6 activity.

Features:

- **Simplified Method:** The homogeneous "add-mix-measure" protocol makes the assay highly amenable to automation.
- **Greater Sensitivity:** The assay is more sensitive than fluorescence-based caspase-6 assays. This bioluminescent assay avoids inherent fluorescent background signals, providing excellent signal-to-noise ratios. The assay is linear over 3 logs of caspase-6 concentration and can detect 0.002U/ml.
- **Increased Accuracy:** The superior sensitivity over fluorescence-based caspase assays allows inhibitor studies at concentrations below the K_m.
- **Faster Results:** The maximum signal (and maximum sensitivity) of the assay is reached in as little as 30 minutes after reagent addition.
- **High-Quality Assay:** The assay demonstrates an excellent Z´-factor value of 0.86 when using 0.1U/ml of caspase-6 for assays in 384-well plates.
- **Batch Processing Capability:** The coupled-enzyme, homogeneous format results in a continuous signal, providing excellent stability and allowing plates to be read over an extended period of time.

Protocol	Part#
Technical Bulletin	TB366

Storage Conditions: Store at –20°C.

Caspase-Glo® 3/7 Assay Systems

Product	Size	Cat.#
Caspase-Glo® 3/7 Assay	2.5 ml	G8090
	10 ml	G8091
	10 × 10 ml	G8093
	100 ml	G8092

For Laboratory Use. Using 100µl of Caspase-Glo® Reagent per assay in a 96-well format, Cat.# G8090 provides sufficient reagents to perform 25 assays; Cat.# G8091, 100 assays; Cat.# G8092 and G8093, 1,000 assays. Using 25µl of Caspase-Glo® Reagent per assay in a 384-well format, Cat.# G8090 provides sufficient reagents to perform 100 assays; Cat.# G8091, 400 assays; Cat.# G8092 and G8093, 4,000 assays.

Description: The Caspase-Glo® 3/7 Assay provides a homogeneous luminescent assay that measures caspase-3/7 activities. The assay provides a proluminescent caspase-3/7 DEVD-aminoluciferin substrate and a proprietary thermostable luciferase in a reagent optimized for caspase-3/7 activity, luciferase activity and cell lysis. Adding the single Caspase-Glo® 3/7 Reagent in an "add-mix-measure" format results in cell lysis, followed by caspase cleavage of the substrate. This liberates free aminoluciferin, which is consumed by the luciferase, generating a "glow-type" luminescent signal. The signal is proportional to caspase-3/7 activity. The stabilized luciferase and proprietary buffer system improve assay performance across a wide range of assay conditions, and the assay is less likely to be affected by compound interference unlike fluorescent- or colorimetric-based assays. The Caspase-Glo® 3/7 Assay is designed for use with multiwell plate formats using either purified enzyme or cells in culture.

Features:

- **Simplify Apoptosis or Caspase Detection:** The "add-mix-measure" protocol makes the assay easy to automate; simply add an equal volume of reagent to sample volume.
- **Use Less Enzyme or Fewer Cells:** The low background luminescence results in excellent signal-to-noise ratios and superior sensitivity not achieved by other caspase formats, allowing assays to be performed in 96- or 384-well formats.
- **Decrease Assay Time:** No sample preparation or manipulation required, and no extended incubation times are necessary, as with fluorescence-based assays. Maximum sensitivity is achieved in as little as 0.25–1 hour.
- **Rely on a Performance-Tested Assay:** The assay delivers excellent Z´-factor values in cell and purified enzyme models.
- **Process Plates in Batch Mode:** The extended-glow signal allows the plates to be read over a 3-hour period of time for batch processing; no injectors required.
- **Get More Information:** Multiplex with other cell-based assays from Promega.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB323

Storage Conditions: Store at –20°C.

Caspase-Glo® 8 Assay Systems

Product	Size	Cat.#
Caspase-Glo® 8 Assay	2.5 ml	G8200
	10 ml	G8201
	100 ml	G8202

For Laboratory Use. Using 100µl of Caspase-Glo® Reagent per assay in a 96-well format, Cat.# G8200 provides sufficient reagents to perform 25 assays; Cat.# G8201, 100 assays; Cat.# G8202, 1,000 assays. Using 25µl of Caspase-Glo® Reagent per assay in a 384-well format, Cat.# G8200 provides sufficient reagents to perform 100 assays; Cat.# G8201, 400 assays; Cat.# G8202, 4,000 assays.

Description: The Caspase-Glo® 8 Assay is a homogeneous luminescent assay that measures caspase-8 activity. The assay provides a proluminogenic caspase-8 substrate in a buffer system optimized for caspase activity, luciferase activity and cell lysis. The addition of a single Caspase-Glo® 8 Reagent in an "add-mix-read" format results in cell lysis, followed by caspase cleavage of the substrate and generation of a "glow-type" luminescent signal. The signal generated is proportional to the amount of caspase activity present. The Caspase-Glo® Reagent relies on the properties of a proprietary thermostable luciferase (Ultra-Glo™ Recombinant Luciferase), which generates the stable "glow-type" luminescent signal and improves performance across a wide range of assay conditions.

The system now includes a separate vial of a protease inhibitor, MG-132 Inhibitor, which may be used to reduce background, thus improving the performance of the Caspase-Glo® 8 Assay in cell-based applications.

Features:

- **Simplify Apoptosis or Caspase Detection:** The homogeneous "add-mix-read" protocol makes the assay easy to automate; simply add an equal volume of reagent to sample volume.
- **Use Less Enzyme:** The low background luminescence results in excellent signal-to-noise ratios and superior sensitivity not achieved by other caspase formats, allowing assays to be performed in 96- or 384-well formats.
- **Decrease Assay Time:** No sample preparation or manipulation required, and no extended incubation times are necessary as with fluorescent-based assays. Maximum sensitivity is achieved in as little as 0.5–1 hour.
- **Rely on a Performance-Tested Assay:** The assay delivers excellent Z′ factors in cell and purified enzyme models.
- **Get More Information:** Multiplex with other cell-based assays from Promega.
- **Experience Improved Caspase-8 Selectivity:** The Caspase-Glo® 8 Assay uses a luminogenic substrate containing the LETD sequence, which has been shown to be selective for caspase-8. The assay includes an optional proteasome inhibitor (MG-132), which when added to the Caspase-Glo® 8 Reagent significantly reduces nonspecific background in cell-based assays.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB332

Storage Conditions: Store at –20°C protected from light.

Caspase-Glo® 9 Assay Systems

Product	Size	Cat.#
Caspase-Glo® 9 Assay	2.5 ml	G8210
	10 ml	G8211
	100 ml	G8212

For Laboratory Use. Using 100µl of Caspase-Glo® Reagent per assay in a 96-well format, Cat.# G8210 contains sufficient reagents for 25 assays; Cat.# G8211, 100 assays; Cat.# G8212, 1,000 assays, respectively. Using 25µl of Caspase-Glo® Reagent per assay in a 384-well format, Cat.# G8210 contains sufficient reagents for 100 assays; Cat.# G8211, 400 assays; Cat.# G8212, 4,000 assays, respectively.

Description: The Caspase-Glo® 9 Assay is a homogeneous luminescent assay that measures caspase-9 activity. The assay provides a proluminogenic caspase-9 substrate in a buffer system optimized for caspase activity, luciferase activity and cell lysis. The addition of a single Caspase-Glo® 9 Reagent in an "add-mix-read" format results in cell lysis, followed by caspase cleavage of the substrate and generation of a "glow-type" luminescent signal. The signal generated is proportional to the amount of caspase activity present. The Caspase-Glo® Reagent relies on the properties of a proprietary thermostable luciferase (Ultra-Glo™ Recombinant Luciferase), which generates the stable "glow-type" luminescent signal and improves performance across a wide range of assay conditions.

The system now includes a separate vial of a protease inhibitor, MG-132 Inhibitor, which may be used to reduce background, thus improving the performance of the Caspase-Glo® 9 Assay in cell-based applications.

Features:

- **Simplify Apoptosis or Caspase Detection:** The homogeneous "add-mix-read" protocol makes the assay easy to automate; simply add an equal volume of reagent to sample volume.
- **Use Less Enzyme:** The low background luminescence results in excellent signal-to-noise ratios and superior sensitivity not achieved by other caspase formats, allowing assays to be performed in 96- or 384-well formats.
- **Decrease Assay Time:** No sample preparation or manipulation required, and no extended incubation times are necessary as with fluorescent-based assays. Maximum sensitivity is achieved in as little as 0.5–1 hour.
- **Rely on a Performance-Tested Assay:** The assay delivers excellent Z′ factors in cell and purified enzyme models.
- **Get More Information:** Multiplex with other cell-based assays from Promega.
- **Experience Improved Caspase-9 Selectivity:** The Caspase-Glo® 9 Assay uses a luminogenic substrate containing the LEHD sequence, which has been shown to be selective for caspase-9. The assay includes an optional proteasome inhibitor (MG-132), which when added to the Caspase-Glo® 9 Reagent significantly reduces nonspecific background in cell-based assays.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB333

Storage Conditions: Store at –20°C protected from light.

Apo-ONE® Homogeneous Caspase-3/7 Assay

Product	Size	Cat.#
Apo-ONE® Homogeneous Caspase-3/7 Assay	1 ml	G7792
	10 ml	G7790
	100 ml	G7791
Available Separately	**Size**	**Cat.#**
Apo-ONE® Homogeneous Caspase-3/7 Buffer	100 ml	G7781

Using 100µl of Apo-ONE® Reagent per assay in a 96-well format, Cat.# G7792 provides sufficient reagents to perform 10 assays; Cat.# G7790, 100 assays; Cat.# G7791, 1,000 assays. Using 25µl of Apo-ONE® Reagent per assay in a 384-well format, Cat.# G7792 provides sufficient reagents to perform 40 assays; Cat.# G7790, 400 assays; Cat.# G7791, 4,000 assays.

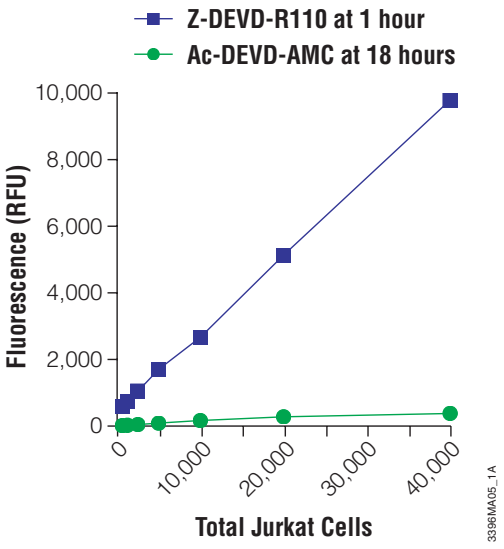
Description: The Apo-ONE® Homogeneous Caspase-3/7 Assay provides the necessary reagents for fast and sensitive measurement of active caspase-3 and -7 in a homogeneous format. The assay includes a profluorescent caspase-3/7 consensus substrate, rhodamine 110 bis-(N-CBZ-L-aspartyl-L-glutamyl-L-valyl-aspartic acid amide) (Z-DEVD-R110), and an optimized bifunctional cell lysis/activity buffer. The buffer efficiently lyses cultured mammalian cells and supports optimal caspase-3/7 enzymatic activity. The substrate and buffer are combined to make the Apo-ONE® Caspase-3/7 Reagent that is added directly to samples. Upon cleavage on the C-terminal side of the aspartate residue in the DEVD peptide substrate sequence by caspase-3/7 enzymes, the rhodamine 110 becomes fluorescent when excited at a wavelength of 498nm. The emission maximum is 521nm. The amount of fluorescent product generated is representative of the amount of active caspase-3/7 present in the sample.

Features:

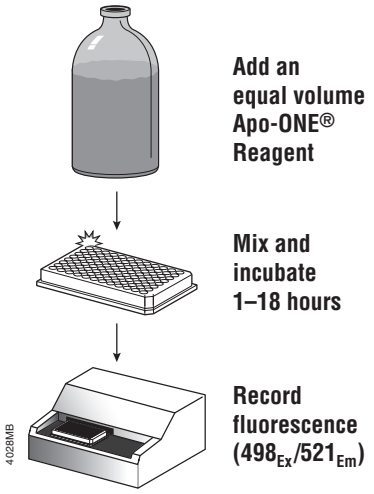
- **Get Results Faster:** The simple "add-mix-measure" format combined with the high sensitivity of the assay dramatically decreases the "time to first result" by eliminating cumbersome sample preparation and lengthy incubation steps.
- **Use Less Enzyme or Fewer Cells:** Optimized caspase-3/7 activity buffer, in conjunction with the R110-labeled substrate, allows for increased sensitivity over existing fluorescent caspase assay methods.
- **Adapt to Your Format and Throughput Needs:** The assay can be flexibly configured (from cuvette to 384-well plate) for use in high-throughput systems by maintaining a 1:1 ratio of sample to assay reagent and may be used with purified enzyme preparations, cell extracts or cultures of adherent, suspension or primary cells.
- **Get More Information:** Perform more than one assay on the same sample. This assay can be multiplexed with other assay methods such as the CellTiter-Blue® Assay (Cat.# G8080) or the Caspase-Glo® 8 or 9 Assays (Cat.# G8200 or G8210).
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB295

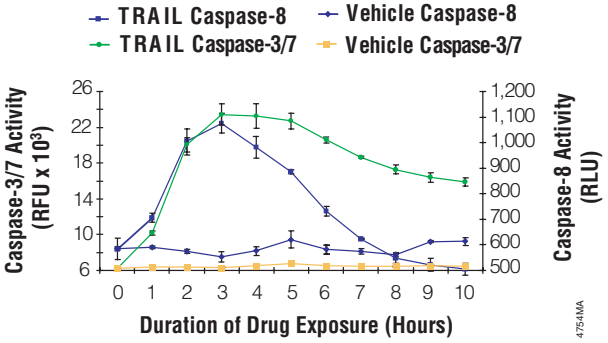
Storage Conditions: Store at –20°C protected from light and moisture.



Superior sensitivity of the Apo-ONE® Homogeneous Caspase-3/7 Assay compared to the AMC substrate-based assay.



Schematic overview of the Apo-ONE® Homogeneous Caspase-3/7 Assay protocol.



Multiplexing luminescent Caspase-Glo® 8 and Apo-ONE® Caspase-3/7 Assay. The time dependence of caspase-8 and caspase-3/7 activity is demonstrated.

CaspACE™ Assay System, Colorimetric

Product	Size	Cat.#
CaspACE™ Assay System, Colorimetric	50 assays	G7351
	100 assays	G7220

For Laboratory Use.

Description: The CaspACE™ Assay System, Colorimetric, provides reagents for measuring the activity of caspase-3. The system includes a colorimetric substrate and a cell-permeant inhibitor that allow quantitative measurement of caspase-3 (DEVDase) protease activity. The colorimetric substrate (Ac-DEVD-pNA) provided is labeled with the chromophore *p*-nitroaniline (pNA). pNA is released from the substrate upon cleavage by DEVDase. Free pNA produces a yellow color that is monitored by a spectrophotometer at 405nm. The amount of yellow color produced upon cleavage is proportional to the amount of DEVDase activity present in the sample.

The potent, irreversible and cell-permeant pan-caspase inhibitor Z-VAD-FMK is provided in the CaspACE™ Assay System, Colorimetric. The addition of the Z-VAD-FMK Inhibitor prior to the induction of apoptosis in cell culture inhibits the activation of the caspase cascade, including caspase-3.

Features:

- **Timely:** Measures an early indicator of apoptosis.
- **Quantitative or Qualitative:** Determine total caspase-3 activity or screen for inducers or inhibitors of caspase activity.
- **Versatile:** May be used with purified enzyme preparations, cell extracts or tissue lysates.

Protocol	Part#
Technical Bulletin	TB270

Storage Conditions: Store at –20°C. Store substrates and inhibitors in aliquots at –20°C away from light and moisture.

CaspACE™ FITC-VAD-FMK In Situ Marker

Product	Size	Cat.#
CaspACE™ FITC-VAD-FMK In Situ Marker	50 µl	G7461
	125 µl	G7462

Description: CaspACE™ FITC-VAD-FMK In Situ Marker is a fluorescent analog of the pan caspase inhibitor Z-VAD-FMK (carbobenzoxy-valyl-alanyl-aspartyl-[O-methyl]-fluoromethylketone). The fluorescein isothiocyanate (FITC) group has been substituted for the carbobenzoxy (Z) N-terminal blocking group to create the fluorescent apoptosis marker. This structure allows delivery of the inhibitor into the cell where it irreversibly binds to activated caspases. The FITC label allows for a single-reagent addition to assay for caspase activity in situ. The FITC-VAD-FMK is supplied as a 5mM solution in DMSO and is intended for in situ monitoring of caspase activity by fluorescence detection. The suggested concentration for use in anti-Fas-treated Jurkat cell culture is 10µM.

Features:

- **Simplify Your Protocol:** Add FITC-VAD-FMK, incubate, wash and view fluorescence.
- **Use a Variety of Detection Methods:** Detect apoptotic cells by fluorescence microscopy or flow cytometry; combine with other immunomarkers to assess cell populations or determine apoptotic frequency within a population; adaptable to high-throughput applications.
- **Get Results Faster:** Quick, single-reagent addition to cell culture; no preparation of cell extracts or long incubation steps. Use as a preliminary screen for apoptosis.
- **Get Reliable Results:** Synthesized peptide provides consistent results from every batch.
- **Use With Live Cells:** Easily moves in and out of cells and remains anchored inside cultured apoptotic cells.

Protocol	Part#
Promega Product Information	9PIG746

Storage Conditions: Store at –20°C protected from light and moisture.

DeadEnd™ Colorimetric TUNEL System

Product	Size	Cat.#
DeadEnd™ Colorimetric TUNEL System	20 reactions	G7360
	40 reactions	G7130

Description: The DeadEnd™ Colorimetric TUNEL System is a modified TUNEL Assay designed to provide simple, accurate and rapid detection of apoptotic cells in situ at the single-cell level. The DeadEnd™ Colorimetric TUNEL System measures nuclear DNA fragmentation, an important biochemical indicator of apoptosis. The system can be used to assay apoptotic cell death in both tissue sections and cultured cells. The DeadEnd™ Colorimetric TUNEL System end-labels the fragmented DNA of apoptotic cells using a modified TUNEL (TdT-mediated dUTP Nick-End Labeling) assay. Biotinylated nucleotide is incorporated at the 3′-OH DNA ends using Terminal Deoxynucleotidyl Transferase. Horseradish-peroxidase-labeled streptavidin (Streptavidin HRP) is then bound to these biotinylated nucleotides, which are detected using the peroxidase substrate, hydrogen peroxide, and the stable chromogen, diaminobenzidine. Using this procedure, apoptotic nuclei are stained dark brown.

Features:

- **Assay Cells or Tissue:** Detect apoptosis in thick tissue sections or assess cell morphology.
- **Simplify:** Includes DAB substrate and H_2O_2 for color detection and plastic coverslips that simplify sample handling.
- **Proven Applications:** Vibratome® sections of neuronal tissue, Jurkat cells, HL-60 cells.

Protocol	Part#
Technical Bulletin	TB199

Storage Conditions: Store the Equilibration Buffer, TdT Enzyme, Biotinylated Nucleotide Mix and Proteinase K at –20°C. Store the Streptavidin HRP, DAB 20X Chromogen, DAB Substrate 20X Buffer and Hydrogen Peroxide 20X at 4°C. Store the SSC 20X and Plastic Coverslips at room temperature.

DeadEnd™ Fluorometric TUNEL System

Product	Size	Cat.#
DeadEnd™ Fluorometric TUNEL System	60 reactions	G3250

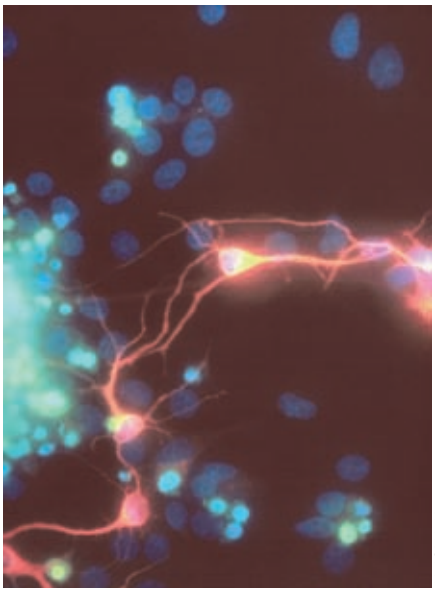
Description: The DeadEnd™ Fluorometric TUNEL System is a classic TUNEL Assay designed for the specific detection and quantitation of apoptotic cells within a cell population. The DeadEnd™ Fluorometric TUNEL System measures nuclear DNA fragmentation, an important biochemical hallmark of apoptosis in many cell types. The system is non-radioactive and provides simple, accurate and rapid detection of apoptotic cells in situ at the single-cell level or in cell suspensions. The DeadEnd™ Fluorometric TUNEL System measures the fragmented DNA of apoptotic cells by catalytically incorporating fluorescein-12-dUTP at 3´-OH DNA ends using the enzyme Terminal Deoxynucleotidyl Transferase (TdT), which forms a polymeric tail using the principle of the TUNEL (TdT-mediated dUTP Nick-End Labeling) assay. The fluorescein-12-dUTP-labeled DNA can then be visualized directly by fluorescence microscopy or quantitated by flow cytometry.

Features:

- **Save Money:** System provides sufficient reagents for 60 assays of 50µl each.
- **Save Time:** Direct incorporation of fluorescent nucleotide reduces number of incubation steps.
- **Choose Sample Type:** Use to detect apoptosis in cultured cells and formalin-fixed, paraffin-embedded tissue sections.
- **Convenient:** Plastic coverslips provided simplify sample handling.

Protocol	Part#
Technical Bulletin	TB235

Storage Conditions: Store at –20°C. Store the Nucleotide Mix protected from light at –20°C.



2272CA06_8A

Neural progenitor cells migrating away from a spherical cluster of apoptotic cells. The condensed nuclei (green) contain fragmented DNA, as indicated by fluorescent labeling with the DeadEnd™ Fluorometric TUNEL System, in contrast with larger intact nuclei stained with DAPI (blue). The cells were also processed for immunocytochemical staining using a primary antibody to βIII Tubulin (Cat.# G7121) and a Cy®3-conjugated secondary antibody where immature process-bearing neurons (red) are distinctly labeled.

Caspase Inhibitor Z-VAD-FMK

Product	Size	Cat.#
Caspase Inhibitor Z-VAD-FMK, 20mM	50 µl	G7231
	125 µl	G7232

Description: Z-VAD-FMK (carbobenzoxy-valyl-alanyl-aspartyl-[O-methyl]-fluoromethylketone) is a cell-permeant pan caspase inhibitor that irreversibly binds to the catalytic site of caspase proteases and can inhibit induction of apoptosis. For inhibition of apoptosis, Z-VAD-FMK should be added at the same time that apoptosis is induced. Z-VAD-FMK is provided at 20mM in DMSO for convenient addition to cell culture or extracts. The peptide is O-methylated in the P1 position on aspartic acid, providing enhanced stability and increased cell permeability. The suggested concentration for use in the anti-Fas mAb-treated Jurkat cell culture model system is 20µM.

Storage Conditions: Store at –20°C protected from light and moisture.

Caspase Inhibitor Ac-DEVD-CHO

Product	Size	Conc.	Cat.#
Caspase Inhibitor Ac-DEVD-CHO	100 µl	10 mM	G5961

Description: Ac-DEVD-CHO is an inhibitor of caspase-3/7 (DEVDase) activity. The concentration of inhibitor required to inhibit caspase activity must be determined empirically for each system. Ten micromolar inhibitor is sufficient to inhibit caspase activity in extracts of apoptotic THP-1 cells. Ac-DEVD-CHO is supplied as a 10mM solution in DMSO.

Storage Conditions: Store at –20°C protected from light and moisture.

Protease-Glo™ Assay

Product	Size	Cat.#
Protease-Glo™ Assay	1 each	G9451
Available Separately	**Size**	**Cat.#**
pGloSensor™-10F Linear Vector	1 µg	G9461

Description: The Protease-Glo™ Assay is a novel method to detect and measure protease activities using a genetically engineered firefly (*Photinus pyralis*) luciferase and represents one example of the GloSensor™ platform technology. The assay uses a circularly permuted firefly luciferase, the GloSensor™-10F protein, with a protease recognition site as the protease substrate. This assay system allows rapid generation of protease substrates through molecular cloning and coupled transcription/translation cell-free expression, thus enabling the facile evaluation of protease function. Oligonucleotides encoding a protease recognition sequence are designed and cloned into the GloSensor™-10F gene located on a linearized vector. The GloSensor™ protein containing the protease site of interest is then synthesized in a cell-free protein expression system and subsequently used as a protease substrate. Cleavage of the protease recognition sequence leads to activation of the GloSensor™ protein and light emission. The level of luminescence correlates to protease activity. The Protease-Glo™ Assay has the advantage of a bioluminescent readout, which provides easy quantitation, high sensitivity and wide dynamic range.

Visit the web application, Protease-Glo™ Assay Oligonucleotide Designer at: **www.promega.com/techserv/tools/proteaseglo/**, to see how to generate your protease recognition site of interest in the pGloSensor™-10F Linear Vector and express the protein using cell-free translation.

Features:

- **Flexible:** Use with P' requiring proteases.
- **Avoid Fluorescent Background Problems:** Physical and chemical features of luminescence overcome problems due to fluorescence interference.
- **Greater Sensitivity:** Ease and dynamic range of luminescence.
- **Open Platform System:** Create your own recognition substrates.
- **Interrogate Sequences:** Excellent tool to determine optimal protease recognition sequences or effects of amino acid substitutions.
- **Web Application:** Makes proper oligo design fast and easy; simply enter your amino acid sequence of interest. See: **www.promega.com/techserv/tools/proteaseglo/**.

Protocol	Part#
Technical Manual	TM303

Storage Conditions: Store all components at –20°C, except the TnT® SP6 High-Yield Wheat Germ Master Mix, which must be stored at –70°C.

Luminometer Plates

Product	Size	Cat.#
Luminometer Plates	50 plates	Z3291

Description: These plates are White 96-Well Cliniplate, Universal Binding, Flat Bottom, and are multiwell plates recommended for use with the Protease-Glo™ Assay. The plates offer excellent optical, binding precision and are compatible with all common instruments (manufactured by Thermo Fisher Scientific).

Features:

- **Compatible with All Common Instruments:** Excellent optical and binding properties.

Protocol	Part#
Technical Manual	TM303

Storage Conditions: Store at room temperature in a cool and dry location.

DUB-Glo™ Protease Assay

Product	Size	Cat.#
DUB-Glo™ Protease Assay (DUB/SENP/NEDP)	10 ml	G6260
	50 ml	G6261

Cat.# G6260 is sufficient for 100 assays at 100µl/assay or 200 assays at 50µl/assay in 96-well plates, or 400 assays at 25µl/assay in 384-well plates. Cat.# G6261 is sufficient for 500 assays at 100µl/assay or 1,000 assays at 50µl/assay in 96-well plates, or 2,000 assays at 25µl/assay in 384-well plates.

Description: The DUB-Glo™ Protease Assay (DUB/SENP/NEDP) is a homogeneous, bioluminescent assay that measures the activity of numerous deconjugating enzymes including deubiquitinating (DUB), deSUMOylating (SENP) and deneddylating (NEDP) proteases. These proteases reverse the protein modification by ubiquitin and ubiquitin-like proteins (Ubl proteins) and thus are integral components in the complex mechanisms of posttranslational protein regulation in eukaryotes.

Features:

- **Greater Sensitivity:** The luminescent format provides enough sensitivity to enable use of a simple peptide-based substrate, Z-RLRGG-aminoluciferin, for assaying deconjugating proteases. Fluorescence generally requires the use of full-length substrates.
- **Broad Dynamic Range:** The assays are linear over 2–3 logs of deconjugating protease concentrations.
- **Signal Stability:** The coupled-enzyme format results in very stable signal with a half-life >3 hours. Substrate depletion is not a concern as it is when using the full-length substrates, Ub-AMC, SUMO-AMC or Nedd8-AMC.
- **Fast:** Maximum sensitivity is reached in 10–30 minutes after reagent addition because the signal is not dependent on accumulation of cleaved product for sensitivity in the coupled-enzyme format.
- **Accurate and Robust:** The broad linear range and excellent sensitivity readily translate to accurate kinetic analysis of inhibitors. Assays can be scaled to 384-well with suitable Z´ factors.
- **Greater Flexibility:** The K_m values for the peptide substrates are much higher than they are for full-length substrates, yet the sensitivity of the luminescent assay allows the assay to be run significantly below K_m while still achieving good signal-to-background ratios for extended time periods. A single luminescent substrate concentration can be used for a wide variety of DUB/SENP/NEDP proteases without worrying about substrate depletion or substrate inhibition.
- **Batch-Processing Capability:** The homogeneous coupled-enzyme format results in a continuous signal, providing excellent stability and allowing plates to be read over an extended period of time.

Protocol	Part#
Technical Manual	TM319

Storage Conditions: Store components at –20°C protected from light.

DPPIV-Glo™ Protease Assay

Product	Size	Cat.#
DPPIV-Glo™ Protease Assay	10 ml	G8350
	50 ml	G8351

For Laboratory Use. Cat.# G8350 provides sufficient reagents for 100 assays at 100µl/assay or 200 assays at 50µl/assay in 96-well plates or 400 assays at 25µl/assay in 384-well plates. Cat.# G8351 provides sufficient reagents for 500 assays at 100µl/assay or 1,000 assays at 50µl/assay in 96-well plates or 2,000 assays at 25µl/assay in 384-well plates.

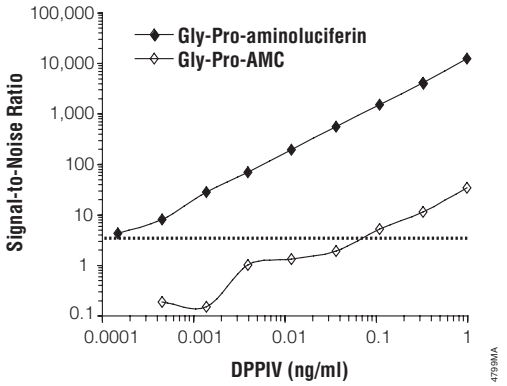
Description: The DPPIV-Glo™ Protease Assay is a homogeneous, luminescent assay that measures dipeptidyl peptidase IV (DPPIV) activity. DPPIV is a serine protease that cleaves N-terminal dipeptides from polypeptides with L-proline or L-alanine at the penultimate position. The DPPIV-Glo™ Assay provides a proluminescent DPPIV substrate, Gly-Pro-aminoluciferin, in a buffer system optimized for DPPIV and luciferase activities. The addition of a single DPPIV-Glo™ Reagent in an "add-mix-measure" format results in DPPIV cleavage of the substrate and generation of a "glow-type" luminescent signal produced by the luciferase reaction. In this homogeneous, coupled-enzyme format, the signal is proportional to the amount of DPPIV activity present. The assay is designed for use with purified enzyme preparations.

Features:

- **Simplified Method:** The homogeneous "add-mix-measure" protocol makes the assay highly amenable to automation.
- **Greater Sensitivity:** The assay is more sensitive than fluorescent-based DPPIV assays. In contrast to fluorescent assays, the luminescent assay avoids inherent fluorescent background signals and thus provides excellent signal-to-background readings. The assay is linear over more than 3 logs of DPPIV concentration and can detect less than 1pg/ml enzyme.
- **Faster Results:** The maximum signal (and maximum sensitivity) of the assay is reached in as little as 30 minutes after reagent addition and, unlike fluorescent assays, is not dependent on accumulation of cleaved product.
- **Amenable to Batch Processing:** The stability of the signal allows plates to be read over an extended period of time.

Protocol	Part#
Technical Bulletin	TB339

Storage Conditions: Store components at –20°C protected from light.



Sensitivity of the DPPIV-Glo™ Protease Assay compared to a fluorescent assay.

Proteasome-Glo™ Assays

Product	Size	Cat.#
Proteasome-Glo™ Chymotrypsin-Like Assay	10 ml	G8621
	50 ml	G8622
Proteasome-Glo™ Trypsin-Like Assay	10 ml	G8631
	50 ml	G8632
Proteasome-Glo™ Caspase-Like Assay	10 ml	G8641
	50 ml	G8642
Proteasome-Glo™ 3-Substrate System	10 ml	G8531
	50 ml	G8532

Cat.# G8632, G8641, G8642, G8531 For Laboratory Use. Cat.# G8621, G8631, G8641 and G8531 (for each of the 3 systems) are each sufficient for 100 assays at 100µl/assay in 96-well plates or 400 assays at 25µl/assay in 384-well plates. Cat.# G8622, G8632, G8642 and G8532 (for each of the 3 systems) are each sufficient for 500 assays at 100µl/assay in 96-well plates or 2,000 assays at 25µl/assay in 384-well plates.

Description: The **Proteasome-Glo™ Assays** are homogeneous, luminescent assays that individually measure the chymotrypsin-like, trypsin-like and caspase-like protease activities associated with the proteasome in an enzyme-based format. The 26S proteasome is a 2.5MDa multiprotein complex found in all eukaryotic cells. Proteasome-Glo™ Cell-Based Assays provide luminogenic proteasome substrates in buffers optimized for cell permeabilization, proteasome activity and luciferase activity. Addition of the Proteasome-Glo™ Cell-Based Reagent in an "add-mix-measure" format results in proteasome cleavage of the substrate and rapid generation of a luminescent signal produced by the luciferase reaction.

The three luminogenic substrates used to monitor specific protease activities include: Suc-LLVY-aminoluciferin for chymotrypsin-like, Z-LRR-aminoluciferin for trypsin-like, and Z-nLPnLD-aminoluciferin for caspase-like activity. Each luminogenic substrate is added to a buffer system optimized for its specific proteasome activity and luciferase activity. The reagents are added to test samples containing proteasome enzyme that cleaves the substrates, releasing luciferin, which is consumed by luciferase, producing "glow-type" luminescence correlating to enzyme activity or inhibition.

The **Proteasome-Glo™ 3-Substrate System** consists of three homogeneous bioluminescent assays in an enzyme-based format (each of these three assays also is available separately).

The **Proteasome-Glo™ Cell-Based 3-Substrate System** consists of three homogeneous bioluminescent assays that measure the three proteolytic activities associated with the proteasome in a cell-based format (each of these three assays also is available separately).

Features:

- **Simplified Method:** The "add-mix-measure" protocol minimizes handling steps and makes the assays amenable to automation.
- **Faster Results:** Maximum sensitivity is reached 10–30 minutes after reagent addition.
- **Greater Sensitivity:** The luminescent assay format avoids inherent fluorescent background signals, providing excellent signal-to-background readings. The assays are miniaturizable to 384-well format.

Protocol	Part#
Technical Bulletin	TB349

Storage Conditions: Store the Proteasome-Glo™ Assay components at –20°C.

Cell-Based Proteasome-Glo™ Assays

Product	Size	Cat.#
Proteasome-Glo™ Chymotrypsin-Like Cell-Based Assay	10 ml	G8660
	5 × 10 ml	G8661
	2 × 50 ml	G8662
Proteasome-Glo™ Trypsin-Like Cell-Based Assay	10 ml	G8760
	5 × 10 ml	G8761
Proteasome-Glo™ Caspase-Like Cell-Based Assay	10 ml	G8860
	5 × 10 ml	G8861
Proteasome-Glo™ 3-Substrate Cell-Based Assay System	10 ml	G1180
	50 ml	G1200

For Laboratory Use. Cat.# G8660, G8760, G8860 and G1180 (for each of the 3 systems) are each sufficient for 100 assays at 100µl/assay in 96-well plates or 400 assays at 25µl/assay in 384-well plates. Cat.# G1200 (for each of the 3 systems) is sufficient for 500 assays at 100µl/assay in 96-well plates or 2,000 assays at 25µl/assay in 384-well plates.

Description: The **Proteasome-Glo™ Cell-Based Assays** are homogeneous, luminescent assays that individually measure the chymotrypsin-like, trypsin-like and caspase-like protease activities associated with the proteasome complex in cultured cells. The 26S proteasome is a 2.5MDa multiprotein complex found in all eukaryotic cells. Proteasome-Glo™ Cell-Based Assays provide luminogenic proteasome substrates in buffers optimized for cell permeabilization, proteasome activity and luciferase activity. Addition of the Proteasome-Glo™ Cell-Based Reagent in an "add-mix-measure" format results in proteasome cleavage of the substrate and rapid generation of a luminescent signal produced by the luciferase reaction.

The three luminogenic substrates used to monitor specific protease activities include: Suc-LLVY-aminoluciferin for chymotrypsin-like, Z-LRR-aminoluciferin for trypsin-like, and Z-nLPnLD-aminoluciferin for caspase-like activity. Each luminogenic substrate is added to a buffer system optimized for its specific proteasome activity and luciferase activity. The reagents are added to cells in culture, and the proteasome cleaves the substrates, releasing luciferin, which is consumed by luciferase, producing "glow-type" luminescence correlating to enzyme activity or inhibition.

The **Proteasome-Glo™ Cell-Based 3-Substrate System** consists of three homogeneous bioluminescent assays that measure the three proteolytic activities associated with the proteasome in a cell-based format (each of these three assays also is available separately).

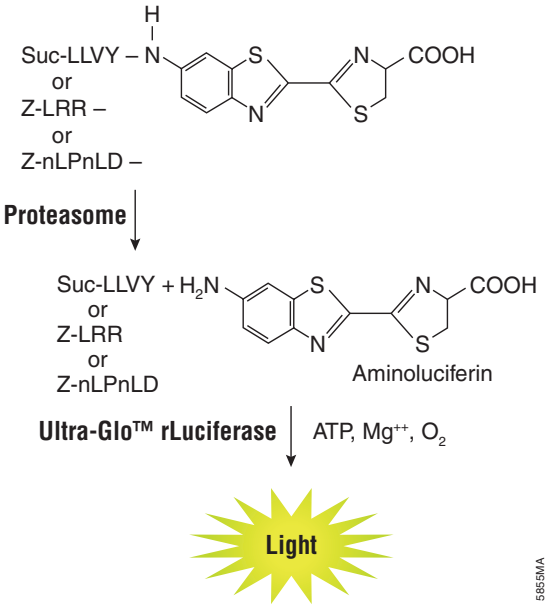
The **Proteasome-Glo™ 3-Substrate System** consists of three homogeneous bioluminescent assays in an enzyme-based format (each of these three assays also is available separately).

Features:

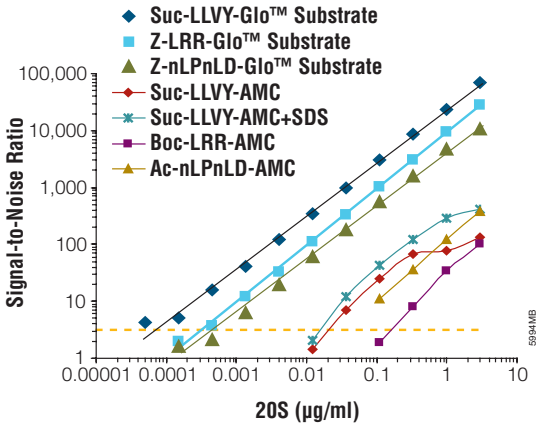
- **More Biologically Relevant Results:** Obtain activity data directly from a cellular environment with the Proteasome-Glo™ Cell-Based Assay.
- **Simplified Method:** The "add-mix-measure" protocol minimizes handling steps and makes the assays amenable to automation.
- **Faster Results:** Maximum sensitivity is reached 10–30 minutes after reagent addition.
- **Greater Sensitivity:** The luminescent assay format avoids inherent fluorescent background signals, providing excellent signal-to-background readings. The assays are miniaturizable to 384-well format.

Protocol	Part#
Technical Bulletin	TB346

Storage Conditions: Store the Proteasome-Glo™ Assay components at –20°C.



The luminogenic substrates containing the Suc-LLVY, Z-LRR, or ZnLPnLD sequence are recognized by the 20S proteasome. Following cleavage by the 20S proteasome, the substrate for luciferase (aminoluciferin) is released, allowing the luciferase reaction to produce light.



Luminescent proteasome assays are more sensitive than fluorescent proteasome assays.

Calpain-Glo™ Protease Assay

Product	Size	Cat.#
Calpain-Glo™ Protease Assay	10 ml	G8501
	50 ml	G8502

Description: The Calpain-Glo™ Protease Assay is a homogeneous, luminescent assay that measures calpain 1 (μ) and 2 (m) activities. Calpains are a family of calcium-activated cysteine proteases involved in cleaving a wide variety of proteins. Calpains modulate the biological activities of their substrates via limited proteolysis.

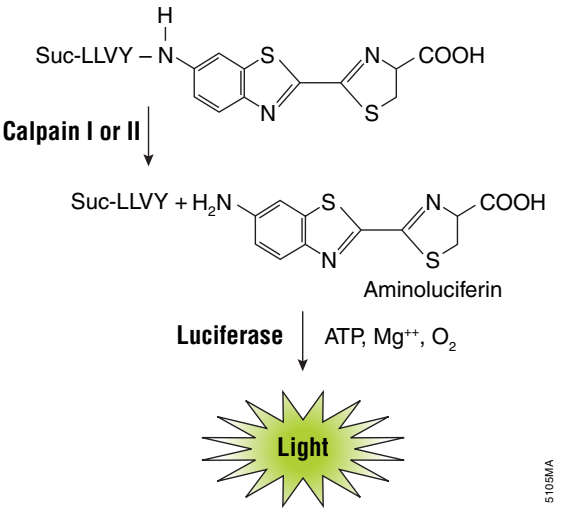
The Calpain-Glo™ Protease Assay provides a succinyl, proluminescent calpain substrate, Suc-LLVY-aminoluciferin, in a buffer system optimized for calpain and luciferase activities. The addition of the calpain reagent in an "add-mix-measure" format results in calpain cleavage of the substrate and rapid development of a "glow-type" luminescent signal produced by the luciferase reaction. The signal is proportional to the amount of calpain activity present. The assay is designed for use with purified enzyme preparations.

Features:

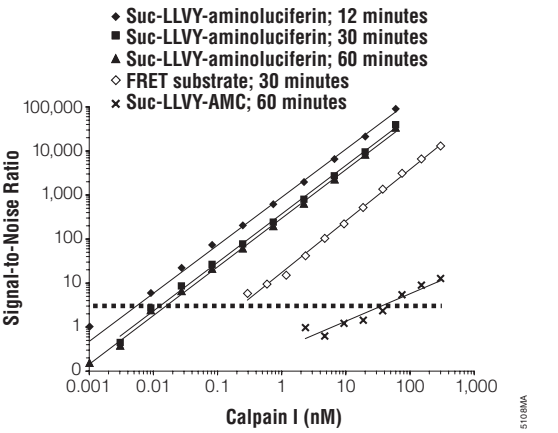
- **Faster Results:** The homogeneous, enzyme-coupled format is especially well suited for rapidly autolysed enzymes like calpain; maximum sensitivity is reached in as little as 10 minutes, while the enzyme is fully active.
- **Simple Protocol:** The homogeneous "add-mix-measure" protocol makes the assay easy to automate.
- **Greater Sensitivity:** The assay is up to 1,000 times more sensitive than competitive fluorometric assays. The luminescent assay avoids inherent fluorescent background signals, providing excellent signal-to-background readings. The assay is linear over 4 logs of calpain concentration.

Protocol	Part#
Technical Bulletin	TB344

Storage Conditions: Store components at –20°C protected from light.



The proluminescent substrate containing the Suc-LLVY sequence recognized by calpain. Following calpain cleavage, the substrate for luciferase (aminoluciferin) is released, allowing the luciferase reaction to occur and producing light.



Sensitivity of the Calpain-Glo™ Protease Assay compared to fluorescent assays.

Tryptase, Human, Recombinant, β

Product	Size	Cat.#
rhSkin β Tryptase	100 µg	G7061
rhLung β Tryptase	100 µg	G5631

Description: Tryptase is the predominant protein in mast cell granules and cleaves proteins at arginine and lysine residues. Tryptase is stored and released from mast cell granules upon activation. Skin β_I Tryptase, Human, Recombinant (rhSkin β Tryptase) and Lung β_{II} Tryptase, Human, Recombinant (rhLung β Tryptase) are neutral serine proteases. The human β tryptase enzymes have been cloned and stably expressed in *Pichia pastoris* as fully active tetrameric enzymes and purified by affinity chromatography. The two enzymes differ in buffer formulation, enzyme concentration and glycosylation pattern. rhLung Tryptase is provided at a much higher concentration (2mg/ml) in minimal buffer without heparin for chromatographic studies and with glycosylation more closely resembling cadaveric enzyme as demonstrated by glycosidase digestion followed by Western analysis of the two recombinant enzymes and native lung tryptase.

Specific Activity: Measured as the rate of hydrolysis of 0.4mM Nα CBZ-L-Lysine Thiobenzyl Ester as substrate coupled with Ellman's Reagent (5,5′-Dithio-bis(5-Nitrobenzoic Acid)) in a final volume of 1ml, incubating for 1 minute at 25°C, and monitoring the absorbance change at 410nm. One unit is defined as 1.0 absorbance unit change per minute.

rhSkin β Tryptase: >1,000 units/mg protein.
rhLung β Tryptase: >1,200 units/mg protein.

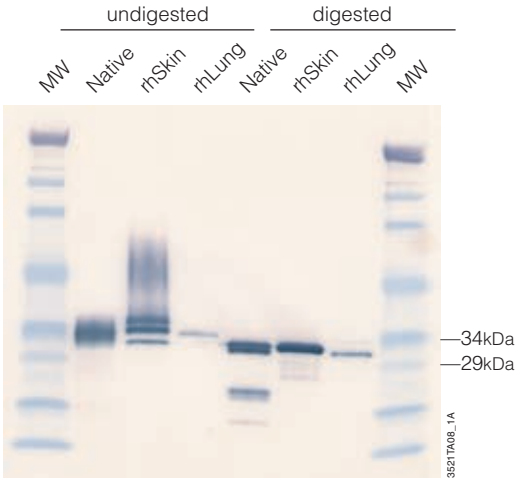
Concentration:

rhSkin β Tryptase: 200µg/ml.
rhLung β Tryptase: 2mg/ml.

Features:

- **High Specific Activity:** Specific activity is consistently 130–150% higher than native lung tryptase.
- **Consistent:** Recombinant protein expression results in uniform enzyme from batch to batch.
- **Safe:** Void of human pathogens associated with native cadaveric tryptase.
- **Pure:** Skin β and Lung β Tryptase are free of other contaminating proteases, providing more active enzyme per milligram of protein and eliminating extraneous protein interactions observed with native tryptase.

Storage Conditions: Store at –20°C.



Glycosidase digestion of human β tryptase with PNGase F yields single tryptase core protein. Western blot with Anti-Human Tryptase mAb (clone AA5, Cat.# G3361).

CYP450 Assay Systems

Product	Size	Cat.#
P450-Glo™ CYP3A4 Assay with Luciferin-IPA	10 ml	V9001
	50 ml	V9002
P450-Glo™ CYP3A4 Assay (Luciferin-PPXE)**DMSO-Tolerant Assay**	10 ml	V8911
	50 ml	V8912
P450-Glo™ CYP3A4 Assay (Luciferin-PFBE)**Cell-Based/Biochemical Assay**	10 ml	V8901
	50 ml	V8902
P450-Glo™ CYP1A1 Assay	10 ml	V8751
	50 ml	V8752
P450-Glo™ CYP1B1 Assay	10 ml	V8761
	50 ml	V8762
P450-Glo™ CYP1A2 Assay	10 ml	V8771
	50 ml	V8772
P450-Glo™ CYP2C8 Assay	10 ml	V8781
	50 ml	V8782
P450-Glo™ CYP2C9 Assay	10 ml	V8791
	50 ml	V8792
P450-Glo™ CYP3A4 Assay	10 ml	V8801
	50 ml	V8802
P450-Glo™ CYP3A7 Assay	10 ml	V8811
	50 ml	V8812
P450-Glo™ CYP2C19 Assay	10 ml	V8881
	50 ml	V8882
P450-Glo™ CYP2D6 Assay	10 ml	V8891
	50 ml	V8892
P450-Glo™ CYP3A4 Screening System with Luciferin-IPA	1,000 assays	V9920
P450-Glo™ CYP3A4 Screening System (Luciferin-PPXE)**DMSO-Tolerant Assay**	1,000 assays	V9910
P450-Glo™ CYP1A2 Screening System	1,000 assays	V9770
P450-Glo™ CYP2C9 Screening System	1,000 assays	V9790
P450-Glo™ CYP3A4 Screening System	1,000 assays	V9800
P450-Glo™ CYP2C19 Screening System	1,000 assays	V9880
P450-Glo™ CYP2D6 Screening System	1,000 assays	V9890
Available Separately	**Size**	**Cat.#**
NADPH Regeneration System	1,000 assays	V9510
Luciferin Detection Reagent	50 ml	V8921
	10 ml	V8920
Luciferin Detection Reagent with esterase	50 ml	V8931
	10 ml	V8930
Luciferin-NAT2	3 mg	P1721
Luciferin-3A7	3 mg	P1741
Luciferin-4A	3 mg	P1621
Luciferin-4F2/3	3 mg	P1651
Luciferin-4F12	3 mg	P1661
Luciferin-2J2/4F12 (ester)	3 mg	P1671
Luciferin-MultiCYP (ester)	3 mg	P1731

Luciferin Detection Reagent must be used in combination with the above pro-luciferin substrates. Use V8921 with P1721, P1741, P1621, P1651 and P1661. **V8931 must be used with P1671 and P1731**, as these substrates are ester derivatives.

Description: The **P450-Glo™ CYP450 Assays** provide a homogeneous, luminescent method for measuring cytochrome P450 activity. The assays are designed to measure the activities of P450s from recombinant and native sources and for testing the effects of analytes such as drugs and new chemical entities on P450 activities. These luminescent assays exhibit exquisite sensitivity, low background signals and broad dynamic range.

P450-Glo™ Assays employ luminogenic P450 substrates that are derivatives of beetle luciferin, a substrate for luciferase enzymes. The derivatives are not substrates for luciferase but are converted by P450s to luciferin, which in turn reacts with luciferase to produce light that is directly proportional to the activity of the P450.

The P450-Glo™ Assays generate a "glow-type" luminescent signal, produced using derivatized luciferins as P450 substrates and a recombinant stabilized luciferase (Ultra-Glo™ Luciferase) coupled with a proprietary buffer system. The half-life of the luminescent output is greater than two hours, eliminating the need for luminometers with injectors and allowing for batch plate processing. The formulation also minimizes the incidence of false positives due to inhibition of luciferase by analytes when screening for cytochrome P450 inhibitors.

The **P450-Glo™ CYP3A4 Assay with Luciferin-IPA** contains a substrate for cytochrome 3A4 that is well suited for all applications involving human CYP3A4 and is the best substrate available for cell-based applications. Luciferin-IPA is readily taken up by cells and rapidly converted into luciferin inside the cell, which reduces the incubation time required (typically 30–60 minutes). The low background and high signal-to-noise ratio produced using Luciferin-IPA means less starting material is required. DMSO is a common vehicle used in drug screening activities, the **P450-Glo™ CYP3A4 Assay with Luciferin-PPXE** is a DMSO-tolerant assay.

The **P450-Glo™ Screening Systems** provide a complete set of reagents for performing luminescent cytochrome P450 assays. The systems include a membrane preparation containing recombinant human cytochrome P450 enzyme, a luminogenic cytochrome P450 substrate appropriate for the enzyme, an NADPH Regeneration System, reaction buffer, Luciferin Detection Reagent and Luciferin-Free Water. The membranes are prepared from baculovirus-infected insect cells and contain human cytochrome P450 and P450 reductase (and cytochrome b5 for CYP2C9 and CYP3A4). The P450-Glo™ Screening Systems also contain a membrane fraction devoid of cytochrome P450 activity as a negative control. The assays are ideal for testing the effects of drugs and new chemical entities on cytochrome P450 enzyme activities.

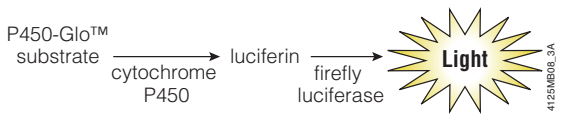
The separate pro-luciferin substrates can be used to monitor the activity of specific isoforms of cytochrome P450 or NAT2 as indicated in the name of the substrate. The Luciferin-MultiCYP is a promiscuous substrate that reacts with at least 21 P450 isoforms and is useful for measuring net CYP activity in a mixed population of P450s. The Luciferin-NAT2 is an excellent substrate for N-acetyltransferase 2 (NAT2), a phase II biotransformation enzyme that acetylates aromatic amine groups on xenobiotic compounds. This substrate shows little to no cross-reactivity with NAT1. For detailed information about the use of these reagents, please visit: **www.promega.com/enotes/applications.htm#adme**

Features:

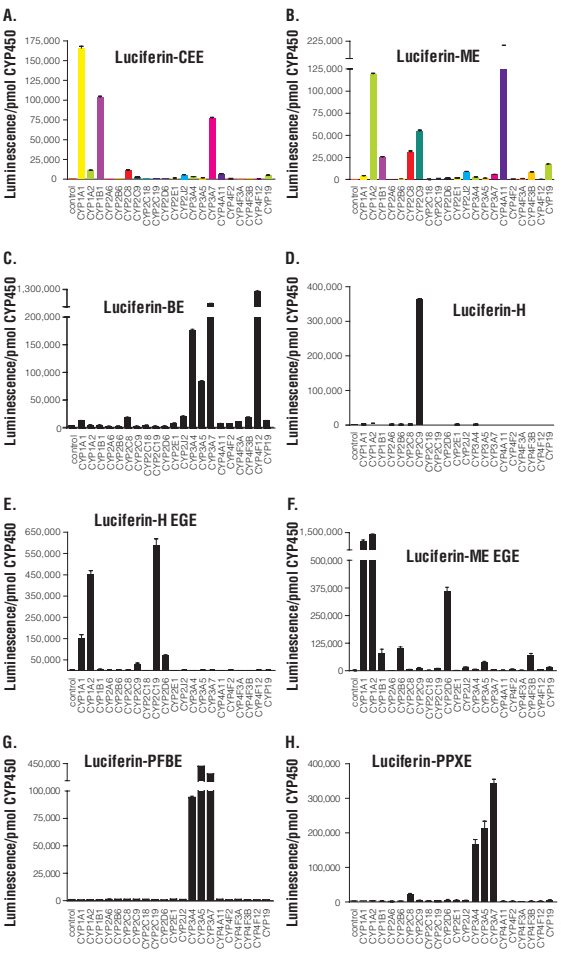
- **Complete Systems:** The screening systems include a membrane preparation containing recombinant human cytochrome P450 enzyme, a luminogenic cytochrome P450 substrate appropriate for the enzyme, an NADPH regeneration system, reaction buffer, Luciferin Detection Reagentand Luciferin-Free Water.
- **Obtain Reliable Results:** The broad dynamic range, low background and better sensitivity result in less ambiguous data.
- **Avoid Fluorescence Interference:** Luminescent output eliminates interference from fluorescent test compounds.
- **Save Time:** Homogeneous assay with simple "add-and-read" format.
- **Avoid False Hits:** Special formulation results in low false-hit rate.
- **Save Money:** Scalable to 384-well format, reducing cost per well.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
P450-Glo™ CYP450 Assay Systems Technical Bulletin	TB325
P450-Glo™ Screening Systems Technical Bulletin	TB340

Storage Conditions: Store the CYP1A2, CYP2C9 and CYP3A4 membranes at –70°C. Cytochrome P450 may lose activity with repeated freeze-thaw cycles. Avoid multiple freeze-thaw cycles by dispensing the CYP1A2, CYP2C9 and CYP3A4 membranes into single-use aliquots (e.g., 50µl for 96 reactions). Store aliquots at –70°C. All other components can be stored at –20°C or –70°C and protected from light.



Conversion of the P450-Glo™ substrate by cytochrome P450. Cytochrome P450 enzymes act on the P450-Glo™ luminogenic substrates to produce luciferin, a substrate for luciferase. Luciferase uses the luciferin to produce light.



Selectivity of the P450-Glo™ substrates for human CYP450 enzymes.

Pgp-Glo™ Assay Systems

Product	Size	Cat.#
Pgp-Glo™ Assay System	10 ml	V3591
Pgp-Glo™ Assay System with P-glycoprotein	10 ml	V3601

Description: The Pgp-Glo™ Assay Systems provide the necessary reagents for performing luminescent P-glycoprotein (Pgp) ATPase assays. Pgp, also known as MDR1 and ABCB1, is a 170kDa integral plasma membrane protein that functions as an ATP-dependent drug efflux pump and plays an important role in multidrug resistance and certain adverse drug-drug interactions. Compounds that interact with Pgp can be identified as stimulators or inhibitors of its ATPase activity. Compounds that are substrates for transport by Pgp typically stimulate its ATPase activity.

The Pgp-Glo™ Assay detects the effects of compounds on recombinant human Pgp in a cell membrane fraction. The assay relies on the ATP dependence of the light-generating reaction of firefly luciferase. ATP is first incubated with Pgp; then the Pgp ATPase reaction is stopped, and the remaining unmetabolized ATP is detected as a luciferase-generated luminescent signal. Pgp-dependent decreases in luminescence reflect ATP consumption by Pgp; thus the greater the decrease in signal, the higher the Pgp activity. Accordingly, samples containing compounds that stimulate the Pgp ATPase will have significantly lower signals than untreated samples.

Features:

- **Complete System:** Cat.# V3591 includes all the reagents required to run the assay except the P-glycoprotein: A Pgp reaction buffer, MgATP, Verapamil, Na_3VO_4, and a lyophilized ATP detection reagent and its reconstitution buffer. Cat.# V3601 includes all the reagents provided in the Pgp-Glo™ System with the addition of Recombinant Human Pgp Membranes to provide a completely optimized kit.
- **Stable Activities:** "Glow-type" signal allows processing of multiple samples without concern of variability over time.
- **Low False-Positive Rate:** Use of a proprietary stabilized firefly luciferase and a proprietary luciferase assay formulation minimizes the incidence of false positives due to inhibition of luciferase by analytes when screening for compounds that affect Pgp activity.
- **Simple:** The simple protocol makes the assay amenable to high-throughput screening in multiwell plates.

Protocol	Part#
Technical Bulletin	TB341

Storage Conditions: Store Recombinant Human Pgp Membranes at –70°C. All other components can be stored at –70°C or –20°C, protected from light.

MAO-Glo™ Assay Systems

Product	Size	Cat.#
MAO-Glo™ Assay	200 assays	V1401
	1,000 assays	V1402
MAO-Glo™ Assay with MAO-A	1,000 assays	V1560
Available Separately	**Size**	**Cat.#**
MAO-A	500 µl	V1452

Description: The MAO-Glo™ Assay provides a homogeneous luminescent method for measuring monoamine oxidase (MAO) activity from recombinant and native sources and for testing the effects of test compounds on MAO activity. The MAO-Glo™ Assay is performed by incubating the MAO enzyme source with a luminogenic MAO substrate. The substrate of the MAO-Glo™ Assay is a derivative of beetle luciferin. Upon reaction with MAO, the derivative is converted into luciferin, which in turn reacts with luciferase to produce light. The amount of light produced is directly proportional to the activity of MAO.

After the MAO reaction has been performed, the reconstituted Luciferin Detection Reagent is added. The reagent simultaneously stops the MAO reaction and initiates a stable glow-type luminescent signal with a half-life greater than 5 hours. This eliminates the need for strictly timed luminescent detection.

The **MAO-Glo™ Assay with MAO-A** contains human recombinant MAO-A enzyme expressed in yeast. The kit is very well suited for the rapid assessment of potential inhibition of MAO-A by new chemical entities and can be used for higher throughput applications such as primary screening. The MAO-A enzyme is also available separately.

The MAO-Glo™ Assay includes a luminogenic MAO substrate, two MAO Reaction Buffers (one that can be used with either MAO A or MAO B enzyme and one that is designed specifically for MAO B), a lyophilized Luciferin Detection Reagent and the Luciferin Detection Buffer. The user supplies the sample material containing MAO. Protocols are configured for multiwell plate formats but easily can be adapted for single-tube applications.

Features:

- **Complete Solution:** The MAO-Glo™ Assay with MAO-A contains monoamine oxidase A enzyme for convenient assessment of the effects of new chemical entities on MAO-A activity.
- **Speed:** The luminescence format eliminates the need for time-consuming analyses such as HPLC.
- **Simplified Method:** The simple "add and read" protocol makes the assay amenable to high-throughput screening in multiwell plates.
- **Greater Sensitivity:** Less MAO enzyme is required in these assays than in typical HPLC or fluorometric methods because of the enhanced sensitivity.
- **No Fluorescence Interference:** Luminescent output eliminates interference from fluorescent test compounds.
- **Stable Signal:** "Glow-type" luminescence provides a stable signal with a half-life of greater than 5 hours. This eliminates the need for strictly timed luminescent detection.

Protocol	Part#
Technical Bulletin	TB345

Storage Conditions: Store MAO-A enzyme (Cat.# V1452) at –70°C. Store all other components at –20°C protected from light.

UGT Activity Assays

Product	Size	Cat.#
UGT-Glo™ Assay	200 assays	V2081
	1,000 assays	V2082
UGT-Glo™ UGT1A1 Screening System	200 assays	V2120
	1,000 assays	V2121
UGT-Glo™ UGT2B7 Screening System	200 assays	V2130
	1,000 assays	V2131

Description: The UGT-Glo™ Assay provides a luminescent method for measuring UDP glucuronosyltransferase (UGT) activity. The UGT-Glo™ Assay is designed to measure UGT activity from a variety of sources, such as microsomes containing recombinantly expressed enzymes or microsomal preparations derived from mammalian tissues, and to test the effects of various chemicals on UGT activity.

The assay involves incubating UGT with a proluciferin substrate; a portion of the substrate gets conjugated with UDP, while the remainder is unmodified. Upon the addition of D-Cysteine, the unconjugated proluciferin is converted into luciferin and, in a coupled reaction with luciferase/luciferin, is converted into light. Conjugated proluciferin remains intact and does not contribute to the luminescence. Thus, the signal generated is inversely correlated with UGT activity present in the sample.

The UGT-Glo™ Assay contains two proluciferin substrates: the UGT Multi-enzyme Substrate, which is compatible with a wide range of UGTs, and the UGT1A4 Substrate, which reacts specifically with UGT1A4. The kit also contains Luciferin Detection Reagent and Reconstitution Buffer, UGT Buffer, D-Cysteine and UDPGA. The UGT-Glo™ Screening Systems contain the above reagents as well as the respective UGT isoforms and control membranes.

Features:

- **Speed:** The luminescent format eliminates the need for time-consuming analyses such as HPLC and LC/MS.
- **Simplified Method:** The simple "add and read" protocol makes the assay amenable to higher throughput screening in multiwell plates.
- **Sensitive:** Allows researchers to use less enzyme and scale down reaction volumes, which saves on reagent costs.

Protocol	Part#
Technical Bulletin	TB551

Storage Conditions: Store UGT enzymes and Control Membranes at –70°C. Store remaining components at –20°C.

DNA and RNA Purification

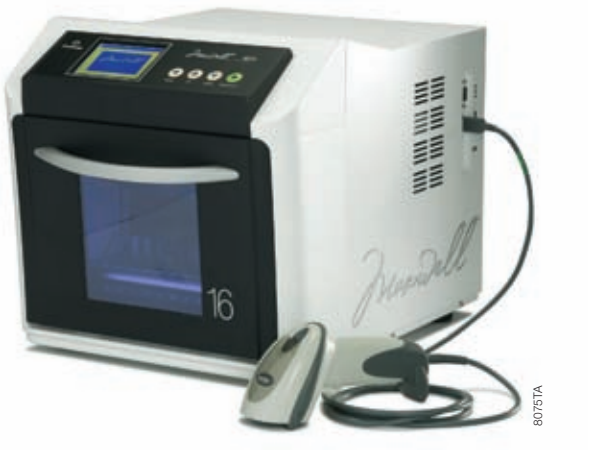
Automated Purification: Maxwell® 16 System	84
Plasmid Purification	89
Plasmid Purification: High-Throughput	94
Fragment Purification	97
Sequencing Reaction Purification	100
Genomic DNA Purification	101
Genomic DNA Purification from Plants and Food	106
DNA Purification Accessories	107
MagneSil® Accessories	108
Total RNA Purification	110
mRNA Purification	114
MagneSphere® Technology Accessories	115

Automated DNA, RNA, Viral Total Nucleic Acid and Polyhistidine-Tagged Protein Purification —Maxwell® 16 System

Product	Size	Cat.#
Maxwell® 16 Instrument	1 each	AS2000
Maxwell® 16 MDx Instrument	1 each	AS3000
Maxwell® 16 Forensic Instrument	1 each	AS3060
Available Separately	**Size**	**Cat.#**
Maxwell® 16 Cartridge Rack	1 each	AS1201
Maxwell® 16 Magnetic Elution Rack	1 each	AS1202
Maxwell® 16 LEV Cartridge Rack	1 each	AS1251
Maxwell® 16 SEV Hardware Kit	1 each	AS1200
Maxwell® 16 LEV Hardware Kit	1 each	AS1250
Thermal Serial Printer and Universal Power Cable	1 each	E2821
Maxwell® 16 LEV Magnet	1 each	AS1261

Cat.# AS2000, AS3000 For Laboratory Use. Cat.# AS3060 Not For Medical Diagnostic Use.

Maxwell® 16 Instrument (Cat. #AS2000).



Maxwell® 16 Instrument (Cat. #AS3000) with optional bar code reader.

Description: The Maxwell® 16 Instruments provide consistent hands-on, labor-saving automated purification of high-quality DNA, RNA, viral total nucleic acid or recombinant proteins for a broad range of downstream applications. The Maxwell® 16 Instrument can be purchased configured as an SEV Instrument (Standard Elution Volume 200–400µl) for maximum yield or LEV Instrument (Low Elution Volume 30–100µl) for maximum concentration. In addition, SEV and LEV instruments can be configured with the Flexi Method Firmware, allowing the user to program the Maxwell® 16 Instrument to further optimize performance. Your personal automation instrument configuration will be built to order. The Maxwell® 16 Instrument is preprogrammed with purification protocols, which when combined with kits containing prefilled reagent cartridges maximize simplicity and convenience. The instrument processes 1 to 16 samples in approximately 18–50 minutes (depending on sample type).

The Maxwell® 16 Instrument extracts DNA, RNA, viral total nucleic acid or recombinant proteins using paramagnetic particles, allowing optimal capture, washing and elution of the target material. Add samples or lysate directly to the prefilled reagent cartridges, and press start. Optimized reagent systems and automated methods are provided to purify from specified sample types to deliver maximum quality for downstream applications.

The Maxwell® 16 Instrument includes a 1-year basic warranty. Service programs are offered to extend coverage. If during the extended warranty period the instrument needs repair under normal use, Promega will be responsible for the repair. Service programs offer similar terms with the addition of the use of a temporary replacement instrument during the instrument repair period. Please contact Promega for complete warranty and service terms and limits.

The Maxwell® 16 Instrument is a General Purpose Medical Device (GPLE) in the USA. Visit: **www.promega.com/maxwell16/** for up-to-date information.

Features:

- **Recover Lost Time and Labor:** Automation gives you back your time and labor to complete your work.
- **Gain Confidence in Your Results:** Instrument design, optimized reagents and automated methods provide consistent yield and purity.
- **Improve Your Productivity:** Process up to 16 samples per instrument run in approximately 30–45 minutes.
- **Choose Your Sample Type:** Flexibility to purify from tissue, cells, blood and other samples.

Protocol	Part#
Maxwell® 16 Instrument Operating Manual	TM295
Maxwell® 16 MDx Instrument Operating Manual	TM320
Maxwell® 16 Forensic Instrument Technical Manual	TM321
Maxwell® Sample Track Software Technical Manual	TM314

Storage Conditions: Store at 22–25°C.

Automated DNA Purification Kits—Maxwell® 16 System

Product	Size	Cat.#
Maxwell® 16 LEV Blood DNA Kit	48 preps	AS1290
Maxwell® 16 Blood DNA Purification Kit	48 preps	AS1010
Maxwell® 16 Cell DNA Purification Kit	48 preps	AS1020
Maxwell® 16 Tissue DNA Purification Kit	48 preps	AS1030
Maxwell® 16 Mouse Tail DNA Purification Kit	48 preps	AS1120
Maxwell® 16 FFPE Tissue LEV DNA Purification Kit	48 preps	AS1130
Maxwell® 16 Cell LEV DNA Purification Kit	48 preps	AS1140
Available Separately	**Size**	**Cat.#**
Maxwell® 16 MDx Instrument	1 each	AS3000
Maxwell® 16 Instrument	1 each	AS2000
Maxwell® 16 LEV Magnet	1 each	AS1261

Cat.# AS1290, AS1010, AS1020, AS1030, AS1130, AS1140, AS3000, AS2000 For Laboratory Use.

Description: The Maxwell® 16 Genomic DNA Purification Kits are designed for use with the Maxwell® 16 Instrument. Seven kits are provided for DNA purification with corresponding optimized automated methods. You get consistent yield and purity from easy-to-use automation.

For genomic DNA purification, the Maxwell® 16 System is the only system that makes purification from tissue as easy as purification from blood or cells. The action of the plunger grinds solid tissue samples directly in the lysis buffer in the prefilled reagent cartridges. Integrated grinding replaces time- and labor-intensive use of lytic enzymes such as proteinase K or manual tissue grinding prior to purification.

Seven kits are provided for optimized DNA purification from eukaryotic tissue, blood, cells, mouse tail and FFPE tissue sections. Protocols for a variety of new samples are being developed. Visit: **www.promega.com/maxwell16/** for up-to-date information.

Features:

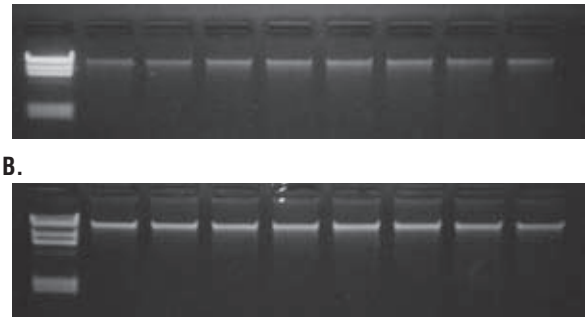
- **Achieve High Yield:** Efficient processing and higher sample capacity than comparable systems.
- **Enjoy Amazing Speed:** Hands-free purification of genomic DNA in 18–30 minutes.
- **Get More Time:** Easily process tissues and cells.

Protocol	Part#
Maxwell® 16 LEV Blood DNA Kit Technical Manual	TM333
Maxwell® 16 DNA Purification Kits Technical Manual	TM284
Maxwell® 16 Mouse Tail DNA Purification Kit Technical Manual	TM309
Maxwell® 16 FFPE Tissue LEV DNA Purification Kit Technical Bulletin	TB382
Maxwell® 16 Cell LEV DNA Purification Kit Technical Bulletin	TB383

Storage Conditions: Store at 5–40°C.

A.

B.



Genomic DNA purified from 8 samples of 200µl of whole human blood (Panel A) and 8 samples of 1cm of mouse tail (Panel B) was visualized on a 1% agarose gel stained with ethidium bromide.
Lane L: Lambda DNA/HindIII Markers (Cat.# G1711). Lanes 1–8: 5µl of purified genomic DNA.

DNA Yields from Various Starting Materials.

Sample Type	Sample Size	Yield
Whole blood	200µl	4–9µg (>3pg/white blood cell)
Whole blood	400µl	8–15µg (>3pg/white blood cell)
Mouse tail	1.2cm	20µg
Animal tissue	20–25mg	60–100µg (mouse liver)
Tissue culture cells	5×10^6	10µg (HeLa)
Gram– bacteria	2×10^9	10µg (BL21)
Gram+ bacteria	2×10^9	1µg (*B. cereus*)
Plant leaf (tomato)	25mg	10µg

9482LA

Automated RNA Purification Kits—Maxwell® 16 System

Product	Size	Cat.#
Maxwell® 16 Total RNA Purification Kit	48 preps	AS1050
Maxwell® 16 Tissue LEV Total RNA Purification Kit	48 preps	AS1220
Maxwell® 16 Cell LEV Total RNA Purification Kit	48 preps	AS1225
Available Separately	**Size**	**Cat.#**
Maxwell® 16 Instrument	1 each	AS2000

Cat.# AS1050, AS1220, AS1225, AS2000 For Laboratory Use.

Description: The Maxwell® 16 Total RNA Purification Kit, Maxwell® 16 Tissue LEV Total RNA Purification Kit and Maxwell® 16 Cell LEV Total RNA Purification Kit are designed for use with the Maxwell® 16 Instrument in either the standard or low elution volume (LEV) configuration. The kits provide high-quality, essentially DNA-free total RNA using novel approaches to selectively remove genomic DNA prior to automated RNA purification. You get enhanced sensitivity and improved confidence in your results for quantitative RT-PCR (qRT-PCR), RT-PCR, cDNA synthesis and other applications.

The Maxwell® 16 Total RNA Purification Kit extracts total RNA from white blood cell fraction of whole blood, tissue culture cell lines, PAXgene®-stabilized whole blood and plant leaf tissue and can be used in RNA cleanup applications from TRIzol® extractions or in vitro transcription reactions. Beta-Mercaptoethanol is included for use with certain sample types.

The Maxwell® 16 LEV Tissue and Cell Total RNA Purification Kits provide high-concentration (100ng/µl), essentially DNA-free total RNA from cultured cells, mammalian tissue and PAXgene®-stabilized white blood cells. It's personal automation working for you, so you can get more consistent gene expression analysis results with less hands-on labor.

The simple protocols require adding a cleared lysate to the reagent cartridge. Place the reagent cartridge into the Maxwell® 16 Instrument, and press start. Purified RNA is obtained in less than 45 minutes of hands-free instrument operation. No post-purification treatment with nuclease, cleanup or concentration is required to achieve superior performance in downstream applications.

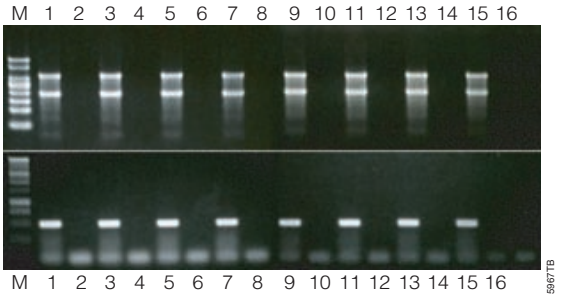
The Maxwell® 16 Total RNA Purification Kits are General Purpose Medical Devices (GPR) in the USA. Visit: **www.promega.com/maxwell16/** for up-to-date information.

Features:

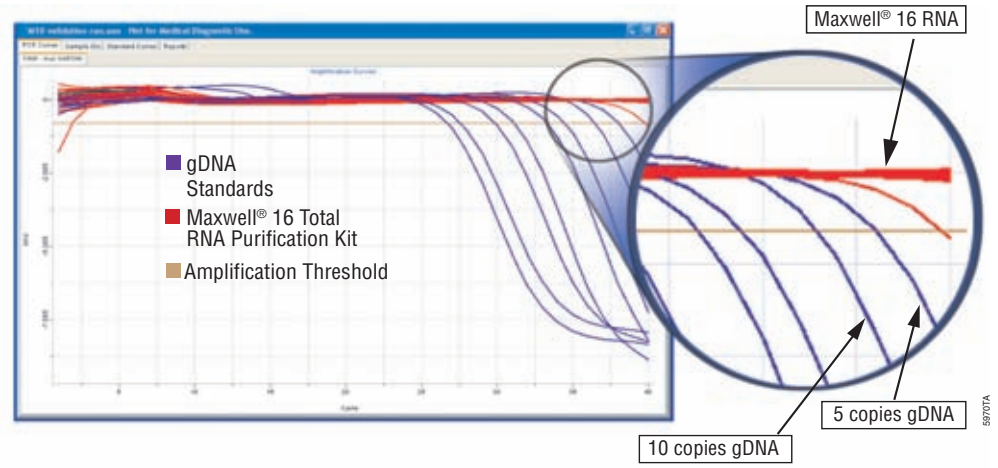
- **Enjoy Confidence in Your Application Results:** Essentially undetectable contaminating genomic DNA means fewer repeated experiments and unexplained or variable results.
- **Choose Your Sample Type:** Flexibility to purify from tissue, cells, blood and other samples.
- **Achieve High Yield and High Concentration:** High yields and high-concentration total RNA result in better performance in gene expression analysis applications.

Protocol	Part#
Maxwell® 16 Total RNA Purification Kit Technical Bulletin	TB351
Maxwell® 16 Tissue LEV Total RNA Purification Kit Technical Bulletin	TB367
Maxwell® 16 Cell LEV Total RNA Purification Kit Technical Bulletin	TB368

Storage Conditions: Store the kit components at room temperature (15–30°C). For Cat.# AS1225, upon receipt, remove the RNasin® Plus RNase Inhibitor and store at –20°C.



No detectable cross-contamination. Sixteen purification reactions were performed using an input of 25mg of mouse liver lysate (odd lanes) or SV RNA Lysis Buffer alone (even lanes). **Panel A.** Four-microliter aliquots of each purified sample were resolved by 1.2% agarose gel electrophoresis under denaturing conditions. Lane M, RNA Markers (Cat.# G3191). **Panel B.** Equivalent volumes (1µl) of each sample were amplified by endpoint RT-PCR using a primer pair specific for a portion of beta actin RNA. A total of five microliters of each amplification reaction was analyzed by 1.2% agarose gel electrophoresis and visualized by ethidium bromide staining. Lane M, 1kb DNA Ladder (Cat.# G5711).



Undetectable genomic DNA contamination. RNA was isolated from 24 replicate samples of 25mg of mouse liver and analyzed using the Plexor® qPCR System and a primer pair specific for a portion of the mouse glyceraldehyde-3-phosphate dehydrogenase (GAPDH) gene. An average of less than 0.1 copies of genomic DNA (gDNA) was observed in purified RNA.

Maxwell® 16 Flexi Method Firmware

Product	Size	Cat.#
Maxwell® 16 Flexi Method Firmware	1 each	AS6411

Description: Certain sample types present unique challenges for DNA, RNA or recombinant protein extraction. The Maxwell® 16 Flexi Method Firmware provides the flexibility and control to modify or create automated methods for the Maxwell® 16 Instrument. You have the ability to optimize multiple instrument parameters to tailor instrument operation to your unique needs. It's Personal Automation™ just the way you want it. The Maxwell® 16 Flexi Method Firmware allows users to change 5 key instrument operating parameters:

- Lysis time
- Binding
- Drying
- Elution
- Paramagnetic particle capture

You program the Maxwell® 16 Instrument by following on-screen prompts and entering changes through the instrument keypad; no external PC or programming knowledge is required. User-defined optimized methods are as easy to use as pushing the Start button. The Flexi Method Firmware also allows you to save and password-protect your unique methods. Make and save changes as you define the key instrument operating parameters that impact your successful results.

The Flexi Method Firmware can be installed on existing AS1000 and AS2000 Maxwell® 16 Instruments by purchasing the AS6411 CD-ROM, which contains the Firmware, installation software and Technical Manual. Flexi Method Firmware ordered with the purchase of a new AS2000 Instrument will be installed at the factory.

Features:

- **Achieve Confidence in your Results:** You control operation of key instrument operating parameters.
- **Address Key Unanswered Questions:** Flexibilty gives you the ability to optimize Maxwell® 16 operation to your sample and scientific needs.
- **Spend More Time Generating Data:** Follow simple on-screen prompts to program instrument from the keypad. Press Run to start.

Protocol	Part#
Technical Bulletin	TB381

Storage Conditions: Store at 22–25°C.

Automated Clinical Nucleic Acid Purification—Maxwell® 16 MDx System

Product	Size	Cat.#
Maxwell® 16 MDx Instrument	1 each	AS3000
Available Separately	**Size**	**Cat.#**
Maxwell® 16 Viral Total Nucleic Acid Purification Kit	48 preps	AS1150
Maxwell® 16 LEV Blood DNA Kit	48 preps	AS1290
Maxwell® 16 Blood DNA Purification Kit	48 preps	AS1010
Maxwell® 16 Cell DNA Purification Kit	48 preps	AS1020
Maxwell® 16 Tissue DNA Purification Kit	48 preps	AS1030
Maxwell® 16 FFPE Tissue LEV DNA Purification Kit	48 preps	AS1130
Maxwell® 16 Cell LEV DNA Purification Kit	48 preps	AS1140
Maxwell® 16 Total RNA Purification Kit	48 preps	AS1050
Maxwell® 16 Tissue LEV Total RNA Purification Kit	48 preps	AS1220
Maxwell® 16 Cell LEV Total RNA Purification Kit	48 preps	AS1225
Maxwell® 16 LEV Hardware Kit	1 each	AS1250
Maxwell® 16 LEV Cartridge Rack	1 each	AS1251
Maxwell® 16 SEV Hardware Kit	1 each	AS1200
Maxwell® 16 Cartridge Rack	1 each	AS1201
Thermal Serial Printer and Universal Power Cable	1 each	E2821
Maxwell® 16 LEV Magnet	1 each	AS1261

Cat.# AS3000, AS1150, AS1290, AS1010, AS1020, AS1030, AS1130, AS1140, AS1050, AS1220, AS1225 For Laboratory Use.

Description: The Maxwell® 16 MDx Instrument provides easy-to-use, consistent and reliable automated nucleic acid extraction of one to 16 samples, bar-code sample tracking, a touch-screen interface and UV decontamination. Both AS3000 instrument packages include the bar-code reader, UV light and Maxwell® Sample Track Software. You choose either low elution volume (50–100µl, LEV) or standard elution volume (300–400µl, SEV) format. Run report data can be transferred from the Maxwell® 16 MDx Instrument to a PC or to an external printer. Data transferred to a PC is can be uploaded to a laboratory information management system (LIMS). The Maxwell® 16 MDx Instrument is labeled as General Purpose Laboratory Equipment (GPLE) in the USA. For the rest of the world, it is intended for research use only.

Features:

- **Fast, Hands-Free Purification:** Improves workflow, and allows staff to perform other value-added tasks.
- **Consistent, Reliable Performance:** Less rework; confidence in results.
- **Easy-to-Use:** Immediate productivity gains; minimal operator training required.
- **Small Size:** Takes up less room on the lab bench. Fits inside biosafety cabinet or hood.
- **Bar-Code Sample Tracking Capability:** Eliminates sample mixup, and data can be integrated into LIMS.
- **UV Light:** Helps decontamination.

Protocol	Part#
Maxwell® 16 MDx Instrument Technical Manual	TM320
Maxwell® Sample Track Software Technical Manual	TM314

Storage Conditions: Store at 15–40°C.

Maxwell® 16 Service and Support

Product	Size	Cat.#
Maxwell® 16 Premier Warranty	1 each	SA2000
Maxwell® 16 Standard Service Agreement	1 each	SA2010
Maxwell® 16 Premier Service Agreement	1 each	SA2015
Maxwell® 16 Preventative Maintenance	1 each	SA2020
Maxwell® 16 Installation Qualification	1 each	SA1001
Maxwell® 16 Operational Qualification	1 each	SA1011
Maxwell® 16 Installation and Operational Qualification	1 each	SA1021

Cat.# SA2000, SA2010, SA2015 Not available in all markets. Please contact your local representative for details.

Description: The **Standard Warranty**, included in the system price, covers all parts, labor and shipping to and from our depot repair location as well as a loaner instrument upon request. The loaner will be shipped via standard ground shipment and will arrive in 5 to 7 working days. We will repair your instrument and return it to you performing to original factory specifications.

The **Premier Warranty** (SA2000) covers all parts, labor and shipping to and from our depot repair location as well as your choice of a loaner instrument within 1 working day or on-site repair by a factory-trained service technician. We will repair your instrument and return it to you performing to original factory specifications. It also includes one preventive maintenance visit.

The **Standard Service Agreement** (SA2010) covers all parts, labor and shipping to and from our depot repair location as well as a loaner instrument upon request. The loaner will be shipped via standard ground shipment and will arrive in 5 to 7 working days. If your Maxwell® 16 Instrument needs repair, we will provide a box for shipment of the instrument back to our service facility. We will repair it and return it to you performing to original factory specifications.

The **Premier Service Agreement** (SA2015) includes all parts, labor and shipping to and from our depot repair location as well as your choice of a loaner instrument within 1 working day or on-site repair by a factory-trained service technician. You can utilize our depot repair and receive a loaner instrument in one working day or you can elect to have one of our service technicians service it in your lab. Additionally, it includes one annual preventive maintenance visit per year.

In order to keep the system operating at peak performance, Promega recommends that Maxwell® 16 Instruments receive a **Preventive Maintenance** (SA2020) check after 12 months of use. During this procedure, our qualified service personnel test the instrument, check parts for wear and replace them as needed. Additionally, the system is aligned and performance is verified. Documentation for your files is provided. The preventive maintenance service is performed by returning the instrument to the factory.

The **Installation Qualification** (SA1001) provides a series of formal on-site instrument checks, delivers written documentation of instrument functionality, and demonstrates that everything ordered with your instrument is supplied and installed in your laboratory. Upon delivery to the lab, the instrument and its components will be visually inspected and reviewed for completeness. Following the inspection, the instrument will be powered on to confirm that the system is properly functional.

The **Operational Qualification** (SA1011) demonstrates that the Maxwell® 16 will function according to its operational specifications. An instrument specialist will check the instrument's alignment and then perform an operational test run to ensure that all of the hardware modules function correctly. Following the documentation of these tests, familiarization training with the instrument's operators will occur. The specialist will also explain all of the sections of the instrument log book.

The **Installation and Operational Qualification** package (SA1021) includes all of the components from both SA1001 and SA1011 in one service product.

Features:

- **Multiple Options to Meet Your Needs:** Allows you to select the warranty coverage or service agreement that best meets the needs of your lab.
- **Factory-Trained Specialists:** Ensures your instrument is repaired quickly and effectively.
- **Expert Technical Service:** Promega experts can help you solve problems quickly.
- **Fixed-Cost Service Products:** Predictable support expenditures.
- **Ongoing System Documentation:** Allows audit tracing and compliance.
- **Comprehensive Service and Support:** Makes certain there is minimal instrument downtime.

8291TA

Wizard® *Plus* SV Minipreps DNA Purification Systems

Product	Size	Cat.#
Wizard® *Plus* SV Minipreps DNA Purification System	50 preps	A1330
	250 preps	A1460
	1,000 preps	A1465
Wizard® *Plus* SV Minipreps DNA Purification System + Vacuum Adapters	50 preps	A1340
	250 preps	A1470
Available Separately	**Size**	**Cat.#**
Alkaline Protease Solution	3 ml	A1441

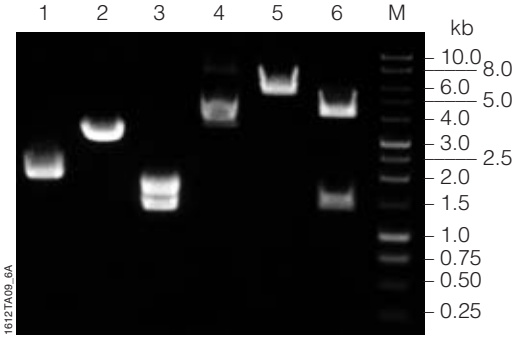
Description: The Wizard® *Plus* SV Minipreps DNA Purification System, a silica membrane-based system, provides a simple and reliable method for rapid isolation of plasmid DNA. The entire miniprep procedure can be completed in 45 minutes or less, depending on the number of samples processed. Using the system, plasmid DNA can be purified from 1–10ml of overnight *E. coli* culture. The purified plasmid DNA can be used directly for automated fluorescent BigDye® terminator DNA sequencing as well as for other standard molecular biology techniques without further manipulation. It also can be used for in vitro transcription reactions when supplemented with a ribonuclease inhibitor such as Recombinant RNasin® Ribonuclease Inhibitor (Cat.# N2511).

Features:

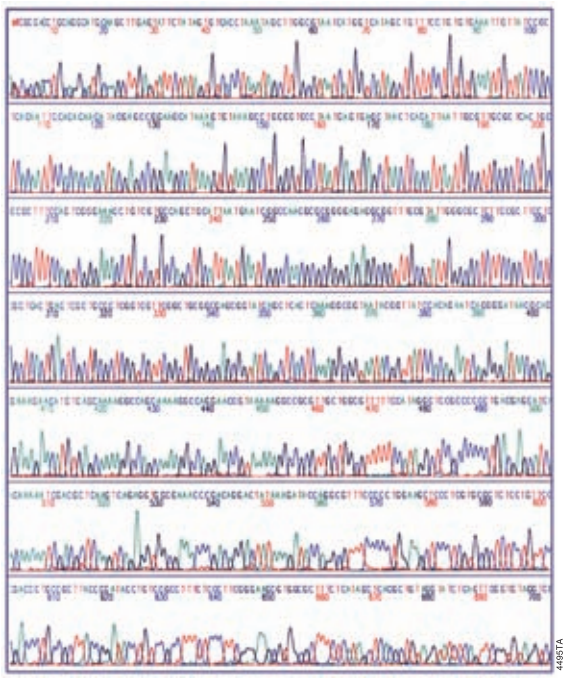
- **Improved Productivity:** 20 minipreps processed in less than 45 minutes.
- **High Performance:** 1–20µg of high-quality plasmid DNA, enough for multiple applications.
- **Safety and Convenience:** No phenol extractions or precipitations required.
- **Flexibility:** Choice of spin (microcentrifuge) or vacuum purification formats.
- **Consistent Quality:** Alkaline protease step improves plasmid quality.
- **Confidence in Results:** Purified DNA meets a target of >98% accuracy over 500 bases using pGEM®-3Zf(+) Vector in BigDye® terminator sequencing.

Protocol	Part#
Technical Bulletin	TB225

Storage Conditions: Store at 22–25°C.



High-quality restriction digests using plasmid purified with the Wizard® *Plus* SV Minipreps DNA Purification System. High-copy pGEM®-3Zf(+) Vector (lanes 1–3) and low-copy pALTER®-1 Vector (lanes 4–6) were each digested in two separate Promega restriction enzyme reactions. All lanes show reproducible and efficient cutting of the plasmid DNA. Lane M is the 1kb DNA Ladder (Cat.# G5711).



pGEM®-3Zf(+) plasmid sequenced with the T7 Promoter Primer (Cat.# Q5021) using DNA purified with the Wizard® *Plus* SV Minipreps DNA Purification System.

PureYield™ Plasmid Miniprep System

Product	Size	Cat.#
PureYield™ Plasmid Miniprep System	100 preps	A1223
	250 preps	A1222

Description: The PureYield™ Plasmid Miniprep System is designed to rapidly isolate highly pure plasmid DNA. The system provides a rapid method for purification of up to 15µg of plasmid DNA from 600µl to 3ml of bacteria culture. Plasmid DNA can be purified in as little as 10 minutes. The PureYield™ Plasmid Miniprep System incorporates a unique Endotoxin Removal Wash designed to remove substantial amounts of protein, RNA and endotoxin contaminants from purified plasmid DNA. Removal of contaminants improves the robustness of sensitive applications such as eukaryotic transfection, in vitro transcription and coupled in vitro transcription/translation (e.g., TnT® Quick Coupled Transcription/Translation System). Purification is achieved without isopropanol precipitation of purified plasmid DNA or extensive centrifugation, providing rapid purification from a single method.

The system has been designed for use with centrifugation or vacuum (e.g., the Vac-Man® Laboratory Vacuum Manifold).

Features:

- **Improved Productivity:** Rapid protocol purifies plasmid DNA in 10 minutes.
- **Robust Performance:** High purity and concentration of plasmid DNA gives proven performance in transfection, cell-free expression and other molecular biology applications.
- **Confidence in Results:** Lysis/neutralization indicator dye ensures success every time.
- **Flexible:** Centrifugation and vacuum protocols are available.

Protocol	Part#
Technical Bulletin	TB374

Storage Conditions: Store all system components at 22–25°C.

PureYield™ Plasmid Midiprep System

Product	Size	Cat.#
PureYield™ Plasmid Midiprep System	25 preps	A2492
	100 preps	A2495
	300 preps	A2496
Available Separately	**Size**	**Cat.#**
Cell Resuspension Solution (CRA)	315 ml	A7115
Cell Lysis Solution (CLA)	315 ml	A7125
Neutralization Solution (NSB)	500 ml	A1485
Eluator™ Vacuum Elution Device	4 each	A1071
Cat.# A1071 For Laboratory Use.		

Description: The PureYield™ Plasmid Midiprep System is designed to isolate transfection-quality plasmid DNA. The system provides a rapid method for purification of 100–200µg of plasmid DNA from 50ml bacteria culture. Plasmid DNA can be purified in as little as 30 minutes with the vacuum protocol, greatly reducing the time spent on purification compared to silica resin or other membrane-column methods. An alternative protocol allows purification of over 400µg of high-copy-number plasmid from 250ml of *E. coli* culture.

The PureYield™ Plasmid Midiprep System incorporates a unique Endotoxin Removal Wash designed to remove substantial amounts of protein, RNA and endotoxin contaminants from purified plasmid DNA. Removal of contaminants improves the robustness of sensitive applications such as eukaryotic transfection, in vitro transcription and coupled in vitro transcription/translation (e.g., TnT® Quick Coupled Transcription/Translation System). Purification is achieved without isopropanol precipitation of purified plasmid DNA or extensive centrifugation, providing rapid purification from a single method.

The system has been designed for use with centrifugation or vacuum (e.g., the Vac-Man® Laboratory Vacuum Manifold).

The Eluator™ Vacuum Elution Device is used to elute nucleic acids from PureYield™ Midiprep or Maxiprep columns. It consists of two pieces, a blue base and a clear column assembly. The base interfaces with a Vacuum Manifold that contains Luer-Lok® fittings, such as the Vac-Man® Laboratory Vacuum Manifold (Cat.# A7231), and holds a 1.5ml tube to capture the eluted nucleic acids. The column assembly accepts PureYield™ Midiprep or Maxiprep columns.

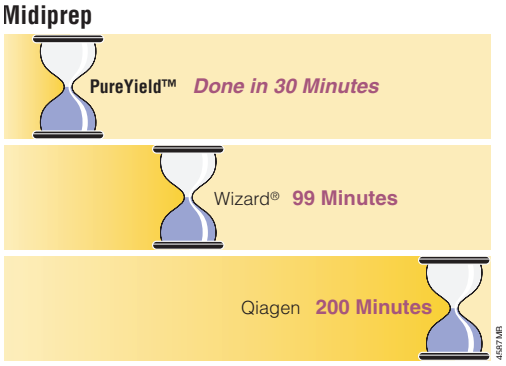
The Eluator™ Device eliminates the requirement for a centrifuge with a swinging bucket rotor for nucleic acid purification, simplifying and speeding purification protocols.

Features:

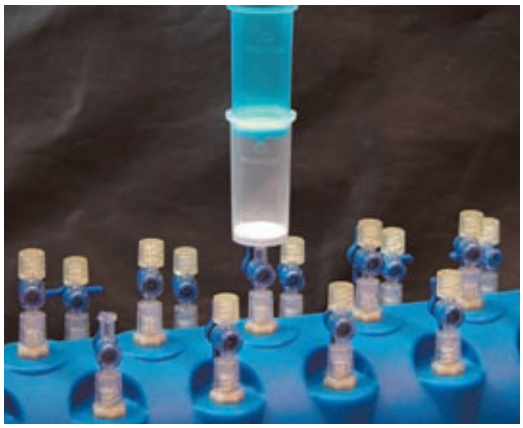
- **Improved Productivity:** Vacuum protocol allows plasmid DNA purification in as little as 30 minutes.
- **Confidence in Results:** High purity and concentration of plasmid DNA gives proven performance in transfection, in vitro expression and other molecular biology applications.
- **Ease of Use:** Simple protocol eliminates tedious high-speed centrifugation, gravity-drip columns, and post-elution alcohol precipitation.
- **Flexibility:** PureYield™ membrane column allows purification of large amounts of plasmid DNA, exceeding the capabilities of other midiprep systems.

Protocol	Part#
Technical Manual	TM253

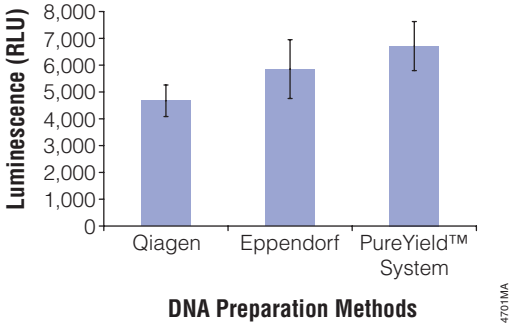
Storage Conditions: Store all system components at 22–25°C.



Comparison of time required per midiprep for different systems. Each system protocol was performed according to the manufacturer's instructions using 50ml of an overnight culture of JM109 bacteria transformed with high-copy-number plasmid [pGEM®-3Zf(+) Vector, Cat.# P2271]. Total time to perform midiprep is noted.



PureYield™ Plasmid Midiprep System. Proper assembly of Lysate Clearing Column (blue) and DNA Binding Column (white) for use with the DNA Purification by Vacuum protocol.



Comparison of transfection of plasmid DNA purified with the PureYield™ System and other midiprep systems. psiCHECK™-2 Vector (Cat.# C8021), which carries a firefly luciferase gene, was isolated from *E. coli* using the PureYield™ System, the Qiagen HiSpeed® Plasmid Midi Kit or the Eppendorf Perfectprep® Plasmid Midi Kit.HeLa cells were transfected using 0.07µg of DNA in a total of 25µl.The firefly luciferase signal was monitored with the Dual-Glo® Luciferase Assay System (Cat.# E2920).

PureYield™ Plasmid Maxiprep System

Product	Size	Cat.#
PureYield™ Plasmid Maxiprep System	10 preps	A2392
	25 preps	A2393
Available Separately	**Size**	**Cat.#**
Eluator™ Vacuum Elution Device	4 each	A1071

Cat.# A1071 For Laboratory Use. This product requires the use of a vacuum pump and vacuum manifold.

Description: The PureYield™ Plasmid Maxiprep System isolates transfection-quality plasmid DNA. The system provides a rapid method for purification of up to 1mg of plasmid DNA from a 250ml bacterial culture. Plasmid DNA can be purified rapidly with the vacuum protocol, greatly reducing the time spent on purification compared to silica resin or other membrane-column methods.

The PureYield™ Plasmid Maxiprep System incorporates a unique Endotoxin Removal Wash designed to remove substantial amounts of protein, RNA and endotoxin contaminants from purified plasmid DNA. Removal of contaminants improves the robustness of sensitive applications such as eukaryotic transfection, in vitro transcription and coupled in vitro transcription/translation (e.g., TnT® Quick Coupled Transcription/Translation System). Purification is achieved without isopropanol precipitation of purified plasmid DNA.

The system has been designed for use with a vacuum source and vacuum manifold (e.g., the Vac-Man® Laboratory Vacuum Manifold).

The Eluator™ Vacuum Elution Device is used to elute nucleic acids from PureYield™ Midiprep or Maxiprep columns. It consists of two pieces, a blue base and a clear column assembly. The base interfaces with a Vacuum Manifold that contains Luer-Lok® fittings, such as the Vac-Man® Laboratory Vacuum Manifold (Cat.# A7231), and holds a 1.5ml tube to capture the eluted nucleic acids. The column assembly accepts PureYield™ Midiprep or Maxiprep columns.

The Eluator™ Device eliminates the requirement for a centrifuge with a swinging bucket rotor for nucleic acid purification, simplifying and speeding purification protocols.

Features:

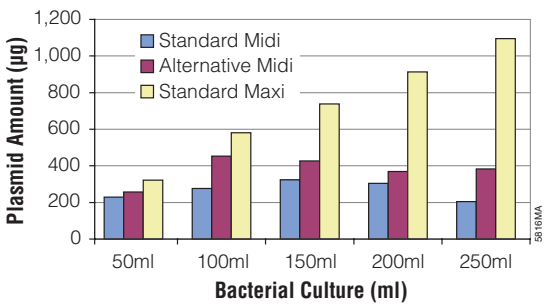
- **Improved Productivity:** Vacuum protocol simplifies purification of multiple samples at one time.
- **Confidence in Results:** High purity and concentration of plasmid DNA gives proven performance in transfection, in vitro expression and other molecular biology applications.
- **Ease of Use:** Simple protocol eliminates tedious, gravity-drip columns and post-elution alcohol precipitation.
- **Flexibility:** PureYield™ membrane column allows purification of large amounts of plasmid DNA, exceeding the capabilities of other maxiprep systems.

Protocol	Part#
Technical Manual	TM280

Storage Conditions: Store at 22–25°C.

Maxiprep

Comparison of time required per prep using different systems. PureYield™ system protocols were performed according to the instructions using 250ml overnight cultures of JM109 bacteria transformed with a high-copy-number plasmid. Total times to perform the PureYield™ preps are noted. Other times are estimated based on protocols.



Plasmid yield from various culture volumes using the PureYield™ Maxiprep and Midiprep Systems. Increasing amounts of JM109 containing the phMGFP plasmid were grown and processed using the PureYield™ Plasmid Systems. Lysate was prepared using the midiprep standard vacuum protocol, the midiprep alternative lysate clearing protocol and the standard maxiprep protocol. The midiprep standard protocol is recommended only for 50ml cultures.

Wizard® *Plus* Minipreps DNA Purification Systems

Product	Size	Cat.#
Wizard® *Plus* Minipreps DNA Purification System	50 preps	A7100
	100 preps	A7500
	250 preps	A7510
Available Separately	**Size**	**Cat.#**
Cell Resuspension Solution (CRA)	150 ml	A7112
Cell Lysis Solution (CLA)	150 ml	A7122
Neutralization Solution (NSA)	150 ml	A7131
Column Wash Solution (CWB)	125 ml	A8102
Wizard® Minipreps DNA Purification Resin	250 ml	A7141
Wizard® Minicolumns	250 each	A7211

This product requires plungers (not provided) for use with the purification protocol without a vacuum manifold.

Description: The resin-based Wizard® *Plus* Minipreps DNA Purification System provides a simple and reliable method for rapid isolation of plasmid DNA. When using the standard protocol, the entire miniprep process can be completed in 15 minutes or less, with no organic extractions or ethanol precipitations. Minipreps may be processed individually or in multiples with the Vac-Man® (20-sample capacity, Cat.# A7231) or Vac-Man® Jr. (2-sample capacity, Cat.# A7660) Laboratory Vacuum Manifold. DNA is eluted from the Wizard® Minicolumn in Nuclease-Free Water (Cat.# P1193). The purified plasmid can be used directly for automated fluorescent DNA sequencing and restriction digestion without further manipulation and also can be used for in vitro transcription reactions supplemented with a ribonuclease inhibitor, such as Recombinant RNasin® Ribonuclease Inhibitor (Cat.# N2511).

The Wizard® Minipreps DNA Purification Resin is used in the isolation and preparation of plasmid DNA in conjunction with the Wizard® *Plus* Minipreps DNA Purification Systems. The resin is available with the systems and as a standalone product.

Features:

- **High Performance:** DNA is suitable for most molecular biology applications, including fluorescent sequencing.
- **Confidence in Results:** Purified DNA meets a target of >98% accuracy over 500 bases using pGEM®-3Zf(+) Vector in BigDye® terminator sequencing.
- **Fast:** Entire procedure may be completed in 15 minutes or less.
- **Convenient:** No phenol extractions or ethanol precipitations required.

Protocol	Part#
Technical Bulletin	TB117

Storage Conditions: Store at 22–25°C.

Wizard® *Plus* Midipreps DNA Purification System

Product	Size	Cat.#
Wizard® *Plus* Midipreps DNA Purification System	25 preps	A7640
Available Separately	**Size**	**Cat.#**
Cell Resuspension Solution (CRA)	150 ml	A7112
Cell Lysis Solution (CLA)	150 ml	A7122
Neutralization Solution (NSA)	150 ml	A7131
Column Wash Solution (CWB)	125 ml	A8102
Wizard® Midipreps DNA Purification Resin	1,000 ml	A7701
Wizard® Midicolumns	100 each	A7651

Description: The resin-based Wizard® *Plus* Midipreps DNA Purification System provides a simple and reliable method for rapid isolation of plasmid DNA. When using the standard protocol, the entire midiprep process can be completed in 90 minutes or less, yielding up to 200µg of high-quality DNA with no organic extractions or ethanol precipitations. Multiple midipreps can be easily processed at one time with the Vac-Man® (20-sample capacity, Cat.# A7231) or Vac-Man® Jr. (2-sample capacity, Cat.# A7660) Laboratory Vacuum Manifold. DNA is eluted from the Wizard® Midicolumn in Nuclease-Free Water (Cat.# P1193). The purified plasmid can be used directly for automated fluorescent DNA sequencing or restriction digestion without further manipulation and also can be used for in vitro transcription reactions supplemented with a ribonuclease inhibitor such as Recombinant RNasin® Ribonuclease Inhibitor (Cat.# N2511). The system includes sufficient reagents for 25 DNA isolations from 10–100ml of liquid culture.

Features:

- **Fast:** Rapid batch column method used for DNA isolation.
- **Safe:** Eliminates the need for cesium chloride:ethidium bromide gradient centrifugation and does not require organic extractions.
- **Reliable:** Yields plasmid DNA of comparable quantity and quality to cesium chloride:ethidium bromide gradient techniques that are much more time- and labor-intensive.
- **High Performance:** DNA is suitable for restriction enzyme digestions, automated fluorescent DNA sequencing, transformation and subcloning.
- **Confidence in Results:** Purified DNA meets a target of >98% accuracy over 500 bases using pGEM®-3Zf(+) Vector in BigDye® terminator sequencing.

Protocol	Part#
Technical Bulletin	TB173

Storage Conditions: Store at 22–25°C.

Wizard® *Plus* Maxipreps DNA Purification System

Product	Size	Cat.#
Wizard® *Plus* Maxipreps DNA Purification System	10 preps	A7270
Available Separately	**Size**	**Cat.#**
Cell Resuspension Solution (CRA)	150 ml	A7112
Cell Lysis Solution (CLA)	150 ml	A7122
Neutralization Solution (NSA)	150 ml	A7131
Column Wash Solution (CWB)	125 ml	A8102
Wizard® Maxipreps DNA Purification Resin	500 ml	A7401
Wizard® Maxi/Megapreps Filtering System	50 each	A7421

Description: The Wizard® *Plus* Maxipreps DNA Purification System provides a simple and rapid resin-based batch column method for purification of plasmid DNA that eliminates the need for cesium chloride:ethidium bromide gradient centrifugation. Use of this system requires only a centrifuge, a vacuum source and a vacuum manifold. This system typically yields 300µg–1mg of high-copy-number plasmid DNA (200–20,000bp) from a 100–500ml culture in less than three hours. The purified DNA is eluted in Nuclease-Free Water (Cat.# P1193) and can be used directly for DNA sequencing and restriction digestion without further manipulation. The DNA also can be used for in vitro transcription reactions supplemented with a ribonuclease inhibitor such as Recombinant RNasin® Ribonuclease Inhibitor (Cat.# N2511).

Features:

- **Flexible:** DNA is suitable for restriction enzyme digestions, automated fluorescent DNA sequencing, transformation and subcloning.
- **High Quality:** Yields plasmid DNA of comparable quantity and quality to cesium chloride:ethidium bromide gradient techniques that are much more time- and labor-intensive.
- **Fast:** Rapid batch binding and column washing method used for DNA isolation.
- **Safe:** Eliminates the need for cesium chloride:ethidium bromide gradient centrifugation and does not require organic extractions.

Protocol	Part#
Technical Bulletin	TB139

Storage Conditions: Store at 22–25°C.

Wizard® *Plus* Megapreps DNA Purification System

Product	Size	Cat.#
Wizard® *Plus* Megapreps DNA Purification System	5 preps	A7300
Available Separately	**Size**	**Cat.#**
Cell Resuspension Solution (CRA)	150 ml	A7112
Cell Lysis Solution (CLA)	150 ml	A7122
Neutralization Solution (NSA)	150 ml	A7131
Column Wash Solution (CWB)	125 ml	A8102
Wizard® Megapreps DNA Purification Resin	1,000 ml	A7361
Wizard® Maxi/Megapreps Filtering System	50 each	A7421

Description: Wizard® *Plus* Megapreps DNA Purification System provides a simple and rapid method for large-scale purifications of plasmid DNA that eliminates the need for cesium chloride:ethidium bromide gradient centrifugation. Use of this system requires only a centrifuge, a vacuum source and a vacuum manifold. The system yields greater than one milligram of high-copy-number plasmid DNA (200–20,000bp) from a 1,000ml culture in less than three hours. The purified DNA is eluted in Nuclease-Free Water (Cat.# P1193) or TE buffer and can be used directly for DNA sequencing and restriction digestion without further manipulation. The DNA also can be used for in vitro transcription reactions supplemented with a ribonuclease inhibitor such as Recombinant RNasin® Ribonuclease Inhibitor (Cat.# N2511).

Features:

- **Fast:** Rapid batch binding and column washing method used for DNA isolation
- **Safe:** Eliminates the need for cesium chloride:ethidium bromide gradient centrifugation and does not require organic extractions.
- **Reliable:** Yields plasmid DNA of comparable quantity and quality to cesium chloride:ethidium bromide gradient techniques that are much more time- and labor-intensive.
- **Yield:** Each megaprep produces >1mg of DNA from 1,000ml of bacterial culture when using a high-copy-number plasmid.
- **Quality:** DNA is suitable for restriction enzyme digestions, automated fluorescent DNA sequencing, transformation and subcloning.

Protocol	Part#
Technical Bulletin	TB140

Storage Conditions: Store at 22–25°C.

Wizard MagneSil Tfx™ System

Product	Size	Cat.#
Wizard MagneSil Tfx™ System	4 × 96 preps	A2380
Available Separately	**Size**	**Cat.#**
Endotoxin Removal Resin	100 ml	A2191
4/40 Wash Solution	115 ml	A2221

Description: The Wizard MagneSil Tfx™ System provides a simple and reliable method for the rapid isolation of transfection-quality plasmid DNA in a 96-well, high-throughput format. The use of MagneSil® Paramagnetic Particles for lysate clearing as well as DNA capture circumvents the need for centrifugation or vacuum manifolds, allowing DNA purification with the Wizard MagneSil Tfx™ System to be completely automated.

An automated method has been developed for use of this product with a Beckman Coulter Biomek® FX robotic workstation. This procedure requires approximately 45 minutes to process a single 96-well plate. The method can be adapted to other robotic workstations, such as the Beckman Coulter Biomek® 2000 or the Tecan Genesis® instrument.

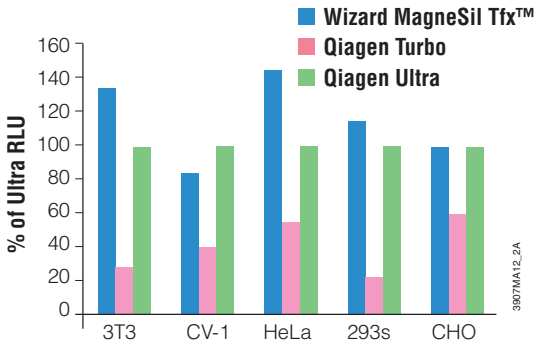
Features:

- **Improve Transfection Results:** Use of Endotoxin Removal Resin cuts endotoxin carryover as much as 95% over standard sequencing-grade automated plasmid systems.
- **Enhance Mammalian Protein Expression:** Three- to fivefold increase in protein expression compared to plasmid isolated from an automated sequencing-grade plasmid purification system.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB314

Storage Conditions: Store at 22–25°C.

Transfection Results



A comparison of transfection efficiencies for different DNA purification systems with various cell lines. A variety of eukaryotic cell lines were transfected with pGL3-Control Vector purified using the Wizard MagneSil Tfx™ System or Qiagen Turbo or Ultra systems. Transfection efficiency was determined by measuring firefly luciferase luminescence, and the results were normalized to those for the Qiagen Ultra system.

Wizard® SV 96 and SV 9600 Plasmid DNA Purification Systems

Product	Size	Cat.#
Wizard® SV 96 Plasmid DNA Purification System	1 × 96 preps	A2250
	5 × 96 preps	A2255
Wizard® SV 9600 Plasmid DNA Purification System	100 × 96 preps	A2258
Available Separately	**Size**	**Cat.#**
Wizard® SV Wash Solution	185 ml	A1311
Wizard® SV 96 Wash Solution	370 ml	A1318
Wizard® SV 96 Neutralization Solution	500 ml	A1481
	950 ml	A1488
Wizard® SV 96 Cell Resuspension Solution	500 ml	A7113
	800 ml	A7118
Wizard® SV 96 Cell Lysis Solution	500 ml	A7123
	800 ml	A7128
Nuclease-Free Water	150 ml	P1195
Alkaline Protease Solution	3 ml	A1441
Wizard® SV 96 Binding Plates	10 pack	A2271
	100 pack	A2278
Wizard® SV 96 Lysate Clearing Plates	10 pack	A2241
	100 pack	A2248

Cat.# A2258 is supplied with the components listed for Cat.# A2250 except for Alkaline Protease Solution, Nuclease-Free Water, 96-Well Deep Well Plates, Elution Plates and Plate Sealers. This product requires the use of a vacuum pump and 96-well vacuum manifold (Vac-Man® 96 Vacuum Manifold, Cat.# A2291).

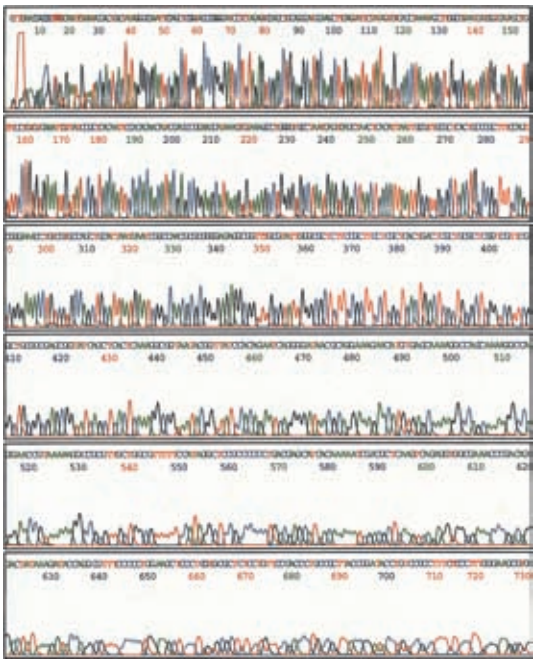
Description: The Wizard® SV 96 and SV 9600 Plasmid DNA Purification Systems provide a simple and reliable method for the rapid isolation of plasmid DNA using a silica-membrane, 96-well, high-throughput format. A single plate can be processed in 60 minutes or less. The purified plasmid can be used directly for automated fluorescent DNA sequencing as well as for other standard molecular biology techniques, including restriction enzyme digestion. The Wizard® SV 96 and SV 9600 Systems are designed for use either in a manual format or with Beckman Coulter or PerkinElmer automated instruments.

Features:

- **Performance by Design:** Vac-Man® 96 Vacuum Manifold eliminates waste handling and allows simultaneous lysate clearing and DNA binding. The novel plate design prevents cross-contamination during sample processing.
- **Flexibility:** Designed for use in both manual and automated formats.
- **Confidence in Results:** Purified DNA meets a target of >98% accuracy over 600 bases using pGEM®-3Zf(+) Vector DNA in BigDye® terminator sequencing.
- **Automation:** Validated automated methods available at: **www.promega.com/automethods/**
- **Your Choice of Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Wizard® SV 96 System Technical Bulletin	TB272
Wizard® SV 9600 System Technical Bulletin	TB292

Storage Conditions: Store at 22–25°C.



Electropherogram of plasmid DNA sequence following isolation by the Wizard® SV 96 System and cycle sequencing using BigDye® terminator reactions. Results demonstrate greater than 600 consecutive bases analyzed with greater than 98% accuracy of base identity.

Wizard® MagneSil® Plasmid Purification System

Product	Size	Cat.#
Wizard® MagneSil® Plasmid Purification System	4 × 96 preps	A1630
	8 × 96 preps	A1631
Wizard® MagneSil® Plasmid Purification System, HTP1	100 × 96 preps	A1635
Available Separately	**Size**	**Cat.#**
MagneSil® RED	100 ml	A1641
MagneSil® BLUE	100 ml	A2201
Cell Resuspension Solution	500 ml	A7114
Cell Lysis Solution	500 ml	A7124
Neutralization Solution	500 ml	A7132
Elution Buffer	500 ml	A1655
Collection Plates (4-pack)	1 each	A9161

Description: The Wizard® MagneSil® Plasmid DNA Purification System provides a simple and reliable method for the rapid isolation of plasmid DNA in a 96-well, high-throughput format. The purified plasmid can be used directly for automated fluorescent sequencing, such as with BigDye® terminator sequencing chemistry, as well as for other standard molecular biology techniques including restriction enzyme digestion.

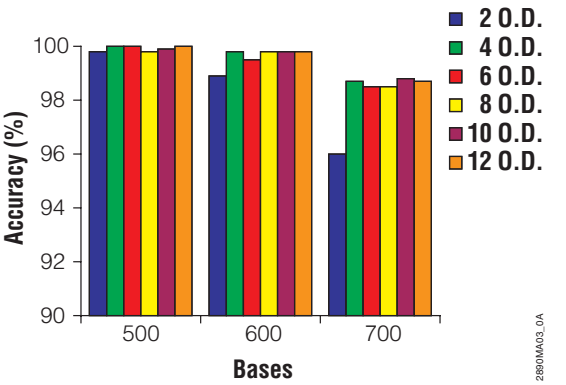
The use of the MagneSil® Paramagnetic Particles for lysate clearing (BLUE) as well as DNA capture (RED) circumvents the need for centrifugation or vacuum manifolds, making the system ideal for full automation on a Beckman Coulter or Tecan instrument.

Features:

- **Improve Productivity:** Process multiple plates without user intervention.
- **Gain Confidence:** Consistent performance in fluorescent sequencing reactions.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB286

Storage Conditions: Store at 22–25°C.



Accuracy by read length for plasmid DNA sequenced following isolation using Wizard® MagneSil® Plasmid Purification System. Results demonstrate >700 consecutive bases analyzed with >98% accuracy of base identity.

Wizard® SV Gel and PCR Clean-Up System

Product	Size	Cat.#
Wizard® SV Gel and PCR Clean-Up System	50 preps	A9281
	250 preps	A9282
	1,000 preps	A9285
Available Separately	**Size**	**Cat.#**
Membrane Binding Solution	20 ml	A9301
Vacuum Adapters	20 each	A1331

Vacuum Adapters (Cat.# A1331) are required for use of the vacuum format with the Wizard® SV Gel and PCR Clean-Up System and must be purchased separately.

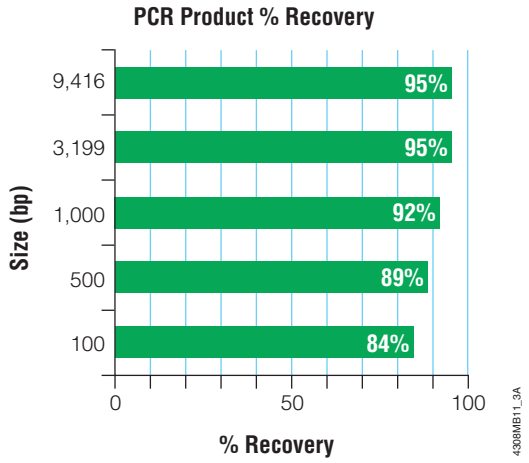
Description: The Wizard® SV Gel and PCR Clean-Up System is designed to extract and purify DNA fragments of 100bp to 10kb from standard or low-melting agarose gels or to purify products directly from PCR and other common reactions such as restriction digests. Up to 95% recovery is achieved depending upon the DNA fragment size. PCR products are commonly purified to remove excess nucleotides and primers. This membrane-based system, which can bind up to 40µg of DNA, allows recovery of isolated DNA fragments or PCR products in as little as 15 minutes, depending on the number of samples processed. The purified DNA can be used for automated fluorescent DNA sequencing, cloning, labeling, restriction enzyme digestion or in vitro transcription/translation without further manipulation.

Features:

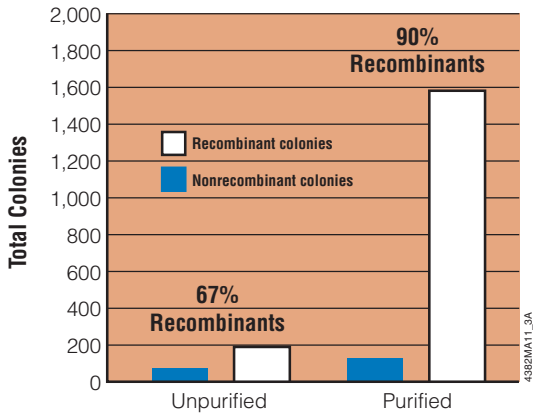
- **Improved Productivity:** Purify DNA fragments or PCR products in as little as 15 minutes.
- **Enhanced Cloning Results:** Up to 95% recovery eluted in as little as 15µl.
- **Confidence in Results:** Purified DNA routinely achieves 700 bases with >98% accuracy in automated fluorescent sequencing.
- **Applications Tested:** DNA is suitable for automated fluorescent sequencing, cloning, labeling, restriction enzyme digestion or in vitro transcription/translation without further manipulation.
- **One System to Do It All:** One system can replace up to four kits from other suppliers.

Protocol	Part#
Technical Bulletin	TB308

Storage Conditions: Store at 22–25°C.



Percent recovery of PCR products (100bp–9kb) from agarose gel slices using the Wizard® SV Gel and PCR Clean-Up System.



Purification of PCR products enhances cloning success. A 500bp PCR product was purified with the Wizard® SV Gel and PCR Clean-Up System and cloned into the pGEM®-T Easy Vector. Both the percent recombinants and total number of colonies increase with a pure PCR product. White bars represent recombinant colonies. Blue bars represent nonrecombinant colonies.

Wizard® PCR Preps DNA Purification System

Product	Size	Cat.#
Wizard® PCR Preps DNA Purification System	50 preps	A7170
	250 preps	A2180
Available Separately	**Size**	**Cat.#**
Wizard® PCR Preps DNA Purification Resin	250 ml	A7181
Direct Purification Buffer	25 ml	A7241
Wizard® Minicolumns	250 each	A7211

This product requires plungers (not provided) for use with the purification protocol without a vacuum manifold.

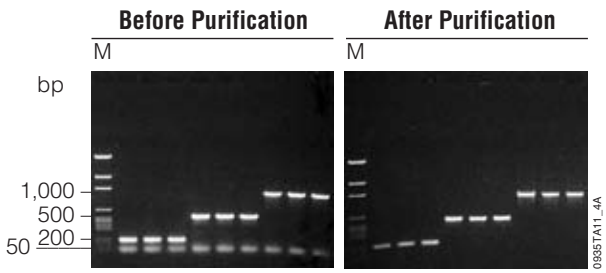
Description: The Wizard® PCR Preps DNA Purification System provides a simple, reliable way to purify double-stranded PCR-amplified DNA. Using the 15-minute batch column purification method, PCR products are effectively separated from contaminants, including primer-dimers and amplification primers. This system also can be used to purify DNA fragments from agarose gels. The DNA can be eluted in water or TE buffer, free of salts or macromolecular contaminants. Multiple PCR Preps may be processed easily at one time with the Vac-Man® Laboratory Vacuum Manifold (Cat.# A7231).

Features:

- **Improved Productivity:** Purify PCR products directly from reactions in 15 minutes.
- **Flexibility:** Separate PCR products from other reaction components such as primers and primer-dimers or from gel slices.
- **Labor Saving Format:** Process multiple purifications simultaneously using the Vac-Man® Laboratory Vacuum Manifold.

Protocol	Part#
Technical Bulletin	TB118

Storage Conditions: Store at 22–25°C.



Recovery of PCR products using Wizard® PCR Preps Resin. A representative sample from the simultaneous purification of 96 PCR products was chosen to determine the effectiveness of the procedure by gel electrophoresis. Equivalent amounts from before and after purification were separated on a 1% agarose gel and stained with ethidium bromide.

Wizard® DNA Clean-Up System

Product	Size	Cat.#
Wizard® DNA Clean-Up System	100 preps	A7280

This product requires plungers (not provided) for use with the purification protocol without a vacuum manifold.

Description: The Wizard® DNA Clean-Up System provides a simple and effective way to purify linear and circular DNA (200–50,000bp) from many molecular biology reactions. Using a quick batch-column procedure, the entire process can be completed in 15 minutes or less with no organic extractions or ethanol precipitations. DNA is eluted in water or TE buffer, ready for use.

Features:

- **Improved Productivity:** Results in 15 minutes or less.
- **Convenience:** No phenol extractions or ethanol precipitations.
- **Flexibility:** Works with a wide range of DNA sizes from 200–50,000bp in length.

Protocol	Part#
Technical Bulletin	TB141

Storage Conditions: Store at 22–25°C.

Wizard® SV 96 PCR Clean-Up System

Product	Size	Cat.#
Wizard® SV 96 PCR Clean-Up System	1 × 96 preps	A9340
	4 × 96 preps	A9341
	8 × 96 preps	A9342
	100 × 96 preps	A9345
Available Separately	**Size**	**Cat.#**
Membrane Binding Solution	20 ml	A9301

This product requires the use of a vacuum pump and 96-well vacuum manifold (Vac-Man® 96 Vacuum Manifold, Cat.# A2291).

Description: The Wizard® SV 96 PCR Clean-Up System is designed for high-throughput purification of 100bp to 10kb PCR products from excess nucleotides, primers and primer dimers. This membrane-based system allows recovery of >90% in as little as 20 minutes. The purified DNA can be used for automated fluorescent sequencing, cloning, labeling, restriction digestion or microarray analysis without further manipulation. The Wizard® SV 96 PCR Clean-Up System uses 96-well filtration without the need to disassemble the manifold. Filtrate waste is delivered directly to a vacuum trap, eliminating the need to dispose of collected waste within the manifold assembly. Protocols are available for automated instruments from Beckman Coulter and PerkinElmer.

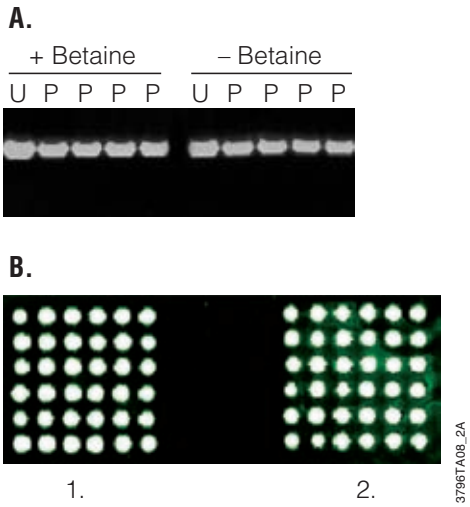
Features:

- **High Performance:** Optimized methods deliver purified PCR products suitable for demanding applications such as microarray analysis.
- **Confidence:** Average recovery for 100–500bp fragments of >90%. Automated fluorescent sequencing Phred* 20 scores >600.
- **Automation:** Validated automated methods available at: **www.promega.com/automethods/**
- **Your Choice of Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

*A Phred score is a widely recognized method to measure the quality of DNA sequences. Phred is a base-calling program for DNA sequence traces available from Codoncode Corporation.

Protocol	Part#
Technical Bulletin	TB311

Storage Conditions: Store at 22–25°C.



Microarray of purified PCR products. PCR products (300bp) were amplified in the presence or absence of 1M betaine, then purified using the Wizard® SV 96 PCR Clean-Up System. **Panel A.** Agarose gel analysis. Purified (P) and unpurified (U) PCR products amplified with (+) or without (–) betaine were separated on an ethidium bromide-stained, 2% agarose gel. **Panel B.** Representative microarray blocks of purified PCR product hybridized to complementary Cy®-labeled cDNA. Betaine interferes with microarray analysis, so the fact that the microarray data for PCR with and without betaine are equivalent clearly demonstrates removal of betaine using the Wizard® SV 96 PCR Clean-Up System. **1.** PCR product amplified under standard amplification conditions (–betaine). **2.** 1M betaine added to the PCR mix.

Wizard® MagneSil® Sequencing Reaction Clean-Up System

Product	Size	Cat.#
Wizard® MagneSil® Sequencing Reaction Clean-Up System	4 × 96 preps	A1831
	8 × 96 preps	A1832
Wizard® MagneSil® Sequencing Reaction Clean-Up System, HTP1	100 × 96 preps	A1835
Available Separately	**Size**	**Cat.#**
MagneSil® GREEN	100 ml	A8231

Cat.# A1831 For Laboratory Use.

Description: The Wizard® MagneSil® Sequencing Reaction Clean-Up System was developed for high-throughput purification of sequencing reactions, including BigDye® Terminator reactions. Cleanup is performed using the proprietary MagneSil® GREEN Paramagnetic Particles with standard, nonskirted 96-well amplification plates. No user intervention is required from the time the plates are placed on the instrument until the samples are ready for loading onto the fluorescent DNA sequencer. Protocols are available for automated instruments from Beckman Coulter and Tecan.

The system relies upon the MagnaBot® II for magnetic separation. The Plate Clamp 96 and Plate Stand are recommended for automated use because they ensure PCR plates are uniformly flat for liquid transfer on a robotic instrument.

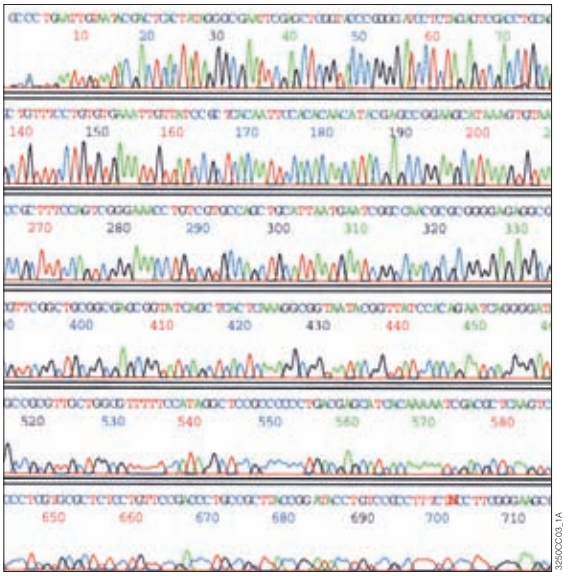
Features:

- **Get Immediate Results:** Validated, walkaway method.
- **Gain Confidence in Results:** Purified products are approved for fluorescent sequencing reactions. Phred* 20 quality scores ≥650 bases.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**

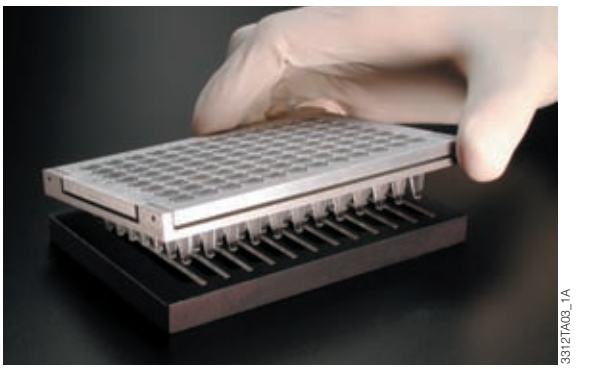
*A Phred score is a widely recognized method to measure the quality of DNA sequences. Phred is a base-calling program for DNA sequence traces available from Codoncode Corporation.

Protocol	Part#
Technical Bulletin	TB287

Storage Conditions: Store at 22–25°C.



Electropherogram of purified sequencing reactions analyzed on an ABI PRISM® 3700 DNA Sequencer. BigDye® terminator reactions purified with the Wizard® MagneSil® Sequencing Reaction Clean-Up System.



Cleanup is performed using the MagnaBot® II Magnetic Separation Device (Cat.# V8351) accompanied by the Plate Clamp 96 (Cat.# V8251). These devices are designed to work with most robotic platforms.

ReliaPrep™ Large Volume HT gDNA Isolation System

Product	Size	Cat.#
ReliaPrep™ Large Volume HT gDNA Isolation System	96 × 10ml preps	A1751
Available Separately	**Size**	**Cat.#**
ReliaPrep™ LV 32 HSM Instrument	1 each	A1715
ReliaPrep™ 32 LV HSM Standard Service Agreement	1 each	SA3070
ReliaPrep™ LV 32 HSM Instrument Cover	1 each	A1712
ReliaPrep™ Tube Rack	1 each	A1713
ReliaPrep™ Tube Rack Stand	1 each	A1714

Cat.# A1751 For Laboratory Use. Cat.# A1715 includes the instrument, cover, tube rack and tube rack stand.

Products may not be available in all countries.

Description: The ReliaPrep™ Large Volume HT gDNA Isolation System isolates genomic DNA (gDNA) from blood samples ranging from 3ml to 10ml of blood in a scalable format. The chemistry eliminates tedious centrifugation steps as well as the use of hazardous chemicals, which are inherent in precipitation-based chemistries. The system has been automated on the Hamilton Robotics MICROLAB® STARplus liquid-handling workstation, allowing walkaway purification of genomic DNA from 3–10ml of whole blood, regardless of sample storage or shipping conditions. For low-throughput isolation of gDNA from up to 32 samples at one time, the ReliaPrep™ 32 LV HSM can be used in a manual mode, where the user performs the pipetting functions. The HSM has an LCD screen that directs the user through the protocol.

Features:

- **Decrease Hands-On Time:** Automation reduces operator time spent on instrument setup and takedown by allowing walkaway operation for 96 samples at a time.
- **Remove Protocol Bottlenecks:** Heater Shaker Magnet eliminates the need to move samples on the robot deck, reducing instrument failures; precipitation-free chemistry dramatically reduces purification failures.
- **Achieve Peace of Mind:** Automated liquid level sensing for all samples and solutions with operator notification allows recovery of samples in case of error.
- **Isolate Pure DNA from All Samples:** Purification chemistry is equally effective at recovering DNA from pristine as well as challenged (hemolysed or frozen) samples.
- **Save a Day or Two of Processing:** Samples are eluted in buffer, ready for use in downstream assays or archiving, eliminating resuspension of pelleted DNA, which can take 24–48 hours.
- **Reduce Waste:** Chemistry is automatically scaled for each sample, using only the reagent required for optimal purification. Plastic use is also conserved, reducing liquid and solid waste during sample runs.
- **Increase Flexibility:** Scheduling software allows 96 sample runs or batching of 32 samples at a time to fit different laboratory workflows. Ability to process frozen samples allows samples to be stored prior to processing when sample backlogs occur.

Protocol	Part#
Technical Manual	TM341

Storage Conditions: Store at 15–30°C.

ReliaPrep™ Blood gDNA Miniprep System

Product	Size	Cat.#
ReliaPrep™ Blood gDNA Miniprep System	100 preps	A5081
	250 preps	A5082

For Laboratory Use.

Description: The ReliaPrep™ Blood gDNA Miniprep System provides a complete, ready-to-use method for purification of gDNA from up to 200µl of blood or body fluid, consistently isolating pure, intact gDNA without the use of alcohol washes or precipitations. Genomic DNA can be prepared from fresh or frozen blood in less than 40 minutes with expected DNA yields of 4–10µg, depending on the white blood cell count of the blood sample.

Features:

- **Easy to Use:** Reagents are supplied "ready to go"; no additions required.
- **Save Time:** Eluted DNA obtained in 30 minutes or less.
- **No Ethanol:** Eliminates alcohol inhibition and carryover.
- **Pure gDNA:** Improved A_{260}/A_{230} ratios vs. the leading competitor.
- **Peace of Mind:** Consistent results from run to run and between users even with hemolyzed samples.
- **Concentrated DNA:** Good recovery and purity in as little as 50µl elution.

Protocol	Part#
Technical Manual	TM330

Storage Conditions: Store at 15–30°C.

Wizard® Genomic DNA Purification Kit

Product	Size	Cat.#
Wizard® Genomic DNA Purification Kit	100 isolations × 300 µl	A1120
	500 isolations × 300 µl	A1125
	100 isolations × 10 ml	A1620
Available Separately	**Size**	**Cat.#**
Cell Lysis Solution (Genomic Purification)	1 liter	A7933
Nuclei Lysis Solution	50 ml	A7941
	1 liter	A7943
Protein Precipitation Solution	25 ml	A7951
	350 ml	A7953
DNA Rehydration Solution	50 ml	A7963
RNase A Solution, 4mg/ml	1 ml	A7973
Proteinase K	100 mg	V3021

Note: Cat.# A1620 is not provided with RNase A Solution.

Description: The Wizard® Genomic DNA Purification Kit provides a simple, solution-based method for isolation of DNA from white blood cells, tissue culture cells, animal tissue, plant tissue, yeast and Gram-positive and Gram-negative bacteria. DNA purified with this system is suitable for a variety of applications, including amplification, digestion with restriction endonucleases and membrane hybridizations (e.g., Southern and dot/slot blots).

Features:

- **Improved Productivity:** Rapidly isolate genomic DNA from blood, tissue culture, animal and plant cells, bacteria and yeast in approximately 60 minutes.
- **Scalability:** Reagent volumes can be adjusted to correspond to the amount of material to be processed.
- **Flexibility:** Genomic DNA purified from a variety of sample types is suitable for a variety of applications.
- **Your Choice of Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Manual	TM050

Storage Conditions: Store at 22–25°C.

DNA Yields from Various Starting Materials Using the Wizard® Genomic DNA Purification Kit.

Source	Amount of Starting Material	Typical DNA Yield
Whole Blood	300µl	5–15µg
	1ml	25–50µg
	10ml	250–500µg
	96-well plate, 50µl/well	0.2–0.7µg
Tissue Culture Cells	10^6–10^7 cells	5–30µg
Animal Tissue		
Mouse Liver	11mg	15–20µg
Mouse Tail	0.5–1cm of tail	10–30µg
Insect Cells	5×10^6 cells	16µg
Plant Leaf Tissue	40mg	7–12µg
Bacterial Culture*	10^8–10^{10} cells	5–20µg
Yeast*	1.9×10^8 cells	4.5–6.5µg

*Overnight culture.

9483LA

Wizard® SV Genomic DNA Purification System

Product	Size	Cat.#
Wizard® SV Genomic DNA Purification System	50 preps	A2360
	250 preps	A2361
Available Separately	**Size**	**Cat.#**
Proteinase K	100 mg	V3021
Wizard® SV Lysis Buffer	50 ml	Z3052
Wizard® SV Wash Solution	185 ml	A1311
Nuclei Lysis Solution	50 ml	A7941
EDTA, 0.5M (pH 8.0), Molecular Biology Grade	100 ml	V4231
RNase A Solution, 4mg/ml	1 ml	A7973

Cat.# V3021, Z3052, A1311, A7941 For Laboratory Use.

Description: The Wizard® SV Genomic DNA Purification System provides a fast, simple, membrane-based technique for preparing genomic DNA from cultured cells and tissue, including mouse tails. Genomic DNA can be purified from cultured cells in about 20 minutes. Isolation from tissue or mouse tails requires an overnight digestion with Proteinase K (Cat.# V3021). Amplifiable genomic DNA can be isolated from up to 5×10^6 cells, 20mg of tissue or up to 1.2cm of a mouse tail tip without a centrifugation clearing step.

The Wizard® SV Genomic DNA Purification System can be used in either a microcentrifuge (spin) or vacuum protocol. Up to 20 samples can be processed at once in the vacuum format with the Vac-Man® Laboratory Vacuum Manifold (Cat.# A7231) and the Vacuum Adapters (Cat.# A1331).

Features:

- **Improved Productivity:** Obtain genomic DNA approximately 20 minutes after lysis.
- **High Yield:** Purify 20–30µg of DNA per prep from 1.2cm mouse tail.
- **Format Choice:** Perform purification by either spin or vacuum formats.

Protocol	Part#
Technical Bulletin	TB302

Storage Conditions: Store at 22–25°C.

Average Yield of Genomic DNA Purified From Various Tissues Using the Wizard® SV and SV 96 Genomic DNA Purification Systems.

Sample Type	Starting Amount	Average Yield
Mouse Tail Clipping	20mg	20µg
Mouse Liver	20mg	15µg
Mouse Heart	20mg	10µg
Mouse Brain	20mg	6µg
CHO Cells	1×10^6 cells	5µg
NIH/3T3 Cells	1×10^6 cells	9µg
293 Cells	1×10^6 cells	8µg

9484LA

Wizard® SV 96 Genomic DNA Purification System

Product	Size	Cat.#
Wizard® SV 96 Genomic DNA Purification System	1 × 96 preps	A2370
	4 × 96 preps	A2371
Available Separately	**Size**	**Cat.#**
Proteinase K	100 mg	V3021
Wizard® SV Lysis Buffer	50 ml	Z3052
Wizard® SV Wash Solution	185 ml	A1311
Nuclei Lysis Solution	50 ml	A7941
EDTA, 0.5M (pH 8.0), Molecular Biology Grade	100 ml	V4231
RNase A Solution, 4mg/ml	1 ml	A7973

Cat.# V3021, Z3052, A1311, A7941 For Laboratory Use. This product requires the use of a vacuum pump and 96-well vacuum manifold (Vac-Man® 96 Vacuum Manifold, Cat.# A2291).

Description: The Wizard® SV 96 Genomic DNA Purification System provides a high-throughput, membrane-based technique for consistent preparation of genomic DNA from cultured cells and tissue, including mouse tails. Amplifiable genomic DNA can be isolated from up to 5×10^6 cells, 20mg of tissue or up to 1.2cm of a mouse tail tip without a centrifugation clearing step.

With the Wizard® SV Genomic DNA purification system, genomic DNA is purified from cell lysates using 96-well vacuum filtration. Washing the bound DNA requires no disassembly of the manifold, and filtrate waste products are delivered directly to a vacuum trap, eliminating the need to empty waste collection trays.

The Wizard® SV Genomic DNA Purification System is designed for use either in a manual format or with Beckman Coulter or PerkinElmer automated instruments.

Features:

- **Improve Productivity:** Obtain genomic DNA from mouse tails in 45–60 minutes, genomic DNA from cultured cells in 30 minutes. No spins required.
- **Achieve High Yield:** Purify 20–30µg of DNA per prep from 1.2cm of mouse tail.
- **Gain Confidence in Applications:** Purified DNA ready for amplification.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB303

Storage Conditions: Store at 22–25°C.

MagneSil® ONE, Fixed Yield Blood Genomic System

Product	Size	Cat.#
MagneSil® ONE, Fixed Yield Blood Genomic System	1 × 96 preps	MD1370
Available Separately	**Size**	**Cat.#**
Lysis Buffer, Blood	160 ml	MD1392
Anti-Foam Reagent	300 µl	MD1431
MagneSil® PMPs—Fixed Yield	25 ml	MD1451
Alcohol Wash, Blood	120 ml	MD1412
Elution Buffer, Blood	45 ml	MD1421
Collection Plates (4-pack)	1 each	A9161

Description: The MagneSil® ONE, Fixed Yield Blood Genomic System purifies 1µg of DNA (+/– 50%) from 60µl of anti-coagulated whole blood. Purification of a "fixed yield" of DNA eliminates the need to quantitate and normalize concentrations postpurification. The highly pure DNA isolated is suitable for use in PCR, multiplex PCR and SNP genotyping applications.

Walkaway automation is available on the Beckman Coulter Biomek® FX in a 96-well format. Process 96 samples in about 1 hour with no hands-on time following robot setup.

Features:

- **Improve Productivity:** Use walkaway automation to extract genomic DNA and eliminate DNA quantitation prior to PCR.
- **Achieve Consistent Results:** Obtain 1µg (fixed yield) of highly pure DNA from 60µl of blood.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB313

Storage Conditions: Store at 20–25°C.

Fixed-Tissue Genomic DNA Purification

Product	Size	Cat.#
MagneSil® Genomic, Fixed Tissue System	100 samples	MD1490

For Laboratory Use.

Description: The MagneSil® Genomic, Fixed Tissue System provides a fast, simple technique to prepare genomic DNA from formalin-fixed, paraffin-embedded tissue. After an overnight Proteinase K digestion, genomic DNA can be manually purified from formalin-fixed, paraffin-embedded thin tissue sections in less than an hour. Amplifiable genomic DNA can be isolated from 10µm thin sections without centrifugation of the lysate prior to purification. Up to 12 samples can be processed in the manual format using the MagneSphere® Technology Magnetic Separation Stand (twelve-position) (Cat.# Z5342).

Features:

- **Purify High-Quality DNA:** The composition of the wash buffers and protocol have been optimized to yield genomic DNA that is largely free of small DNAs, a potent inhibitor of PCR amplification.
- **Rely on Performance-Tested Amplification Results:** Amplify targets in multiplex PCR and targets as large as 450–1,800bp.

Protocol	Part#
Technical Bulletin	TB319

Storage Conditions: MD1490 consists of two separate items shipped at different temperatures. MD1170 (part 1 of 2 for MD1490 - Processing Module) contains Proteinase K, DTT and Incubation Buffer, which are shipped on dry ice. Store MD1170 at –20°C.

MD1180 (part 2 of 2 for MD1490 - Purification Module) contains Lysis Buffer, 2X Wash Buffer, Resin and Elution Buffer, which are shipped at room temperature, 22–25°C. Store MD1180 at room temperature, 22–25°C.

MagneSil® Blood Genomic, Max Yield System

Product	Size	Cat.#
MagneSil® Blood Genomic, Max Yield System	1 × 96 preps	MD1360
Available Separately	**Size**	**Cat.#**
Anti-Foam Reagent	300 µl	MD1431
MagneSil® Paramagnetic Particles	25 ml	MD1441
Salt Wash, Blood	90 ml	MD1401
Alcohol Wash, Blood	70 ml	MD1411
Elution Buffer, Blood	45 ml	MD1421
Collection Plates (4-pack)	1 each	A9161

Cat.# MD1431, MD1441, MD1401, MD1411, MD1421 For Laboratory Use.

Description: The MagneSil® Blood Genomic, Max Yield System provides automated high-throughput DNA purification on the Beckman Coulter Biomek® FX using MagneSil® Paramagnetic Particle technology. DNA from 96 samples of anti-coagulated human whole blood is purified in about 1 1/2 hours with no hands-on time once the robot protocol is initiated. Studies on DNA recovery and purity and PCR results show no cross-contamination between samples in adjacent wells. Purified DNA is qualified for single-locus "simple PCR" as well as more demanding applications such as multiplex PCR (e.g., PowerPlex® 16 System [Cat.# DC6531], Y Chromosome Deletion Detection System [Cat.# MD1531]) and SNP genotyping.

Features:

- **Improve Productivity:** Walkaway automation of genomic DNA extraction.
- **Achieve Maximum Yield:** The average yield of 96 purified samples from normal healthy adults is ≥4µg.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB312

Storage Conditions: Store at 22–25°C.

MagneSil® Genomic, Large Volume System

Product	Size	Cat.#
MagneSil® Genomic, Large Volume System	8 preps	A4080
	48 preps	A4082
	96 preps	A4085
Available Separately	**Size**	**Cat.#**
eLysis Buffer, Large Volume System	1 L	A4091

For Laboratory Use.

Description: The MagneSil® Genomic, Large Volume System, is designed for scalable, automated genomic DNA isolation from large-volume samples, eliminating laborious centrifugation steps and the use of hazardous organic solvents. The system has been automated on the Tecan Freedom EVO® liquid handler, providing walkaway purification of genomic DNA from a variety of starting materials, including 1–10ml whole blood samples, regardless of sample storage or shipping conditions. The instrument uses only the amount of reagents required to process each sample, maximizing efficiency and value per prep.

The MagneSil® Genomic, Large Volume System, uses a robust noncentrifugation-based automated method to purify genomic DNA from fresh, frozen or mishandled blood and other samples with similar yields and quality. The system bypasses many of the challenges of traditional centrifugation-based methods by lysing the entire whole blood sample and then directly capturing total genomic DNA from the lysed sample using MagneSil® Paramagnetic Particles (PMPs). The genomic DNA bound to the MagneSil® PMPs is washed to remove contaminants such as heme and cellular proteins, then eluted into an aqueous solution ready for use in downstream applications. There is no need for tedious and lengthy DNA rehydration. The purified genomic DNA is suitable for a variety of downstream applications such as single and multiplex PCR, restriction digestion and real-time PCR.

Features:

- **Improve Productivity:** Walkaway automation from blood-collection tube to application-ready DNA.
- **Rely on an Integrated Solution:** One reagent system and automated method provide yield and purity from any sample type (fresh or frozen blood, samples of unknown quality and mixed sample populations).
- **Enjoy Smart Scalability:** Scale sample size from 1–10ml of blood, batch size from 1–96 samples and reagent usage from input sample volume.
- **Achieve Turnkey Automation:** Optimized protocol available for the Tecan Freedom EVO® instrument. This and other validated automated methods are available at: **www.promega.com/automethods/**

Protocol	Part#
Technical Bulletin	TB549

Storage Conditions: Store at 22–25°C.

ReadyAmp™ Genomic DNA Purification System

Product	Size	Cat.#
ReadyAmp™ Genomic DNA Purification System	100 reactions	A7710

Description: The ReadyAmp™ Genomic DNA Purification System yields single-stranded DNA (ssDNA) from whole blood or blood stains that may be used directly in amplification reactions without further manipulation. The process takes less than one hour and requires no organic extractions or ethanol precipitations.

Features:

- **Simple and Effective:** ReadyAmp™ resin removes PCR inhibitors.
- **Convenient:** Isolated DNA can be used directly in PCR amplifications.

Protocol	Part#
Technical Bulletin	TB190

Storage Conditions: Store at 22–25°C.

MagneSil® KF, Genomic System

Product	Size	Cat.#
MagneSil® KF, Genomic System	200 preps	MD1460
Available Separately	**Size**	**Cat.#**
MagneSil® KF, Paramagnetic Particles	40 ml	MD1471
Lysis Buffer, KF	160 ml	MD1521

For Laboratory Use.

Description: The MagneSil® KF, Genomic System is designed for easy, walkaway, low- to moderate-throughput automated genomic DNA purification from blood and other samples. For blood samples, lysis occurs concurrently with DNA binding to MagneSil® Paramagnetic Particles. After washes to remove heme and proteins, purified genomic DNA is ready for PCR and other downstream applications. The system is designed to purify 2–6µg of genomic DNA from 200µl of anti-coagulated liquid blood.

The MagneSil® KF, Genomic System is designed to run on the Thermo Electron KingFisher® mL instrument, which automates DNA purification in a flexible 1- to 15-sample batch, 25-minute walkaway format. The compact size of the KingFisher® mL allows it to be used on the benchtop or in a laminar flow hood. Please contact Thermo Electron for more information on the KingFisher® mL instrument.

Features:

- **Improve Productivity:** Use automated 25-minute optimized, walkaway protocol with no training. Eliminate laborious manual methods.
- **Rely On a Performance-Tested System:** Purified DNA is tested in PCR, multiplex PCR, fluorescent STR analysis and SNP genotyping applications.
- **Conserve Valuable Lab Space:** The small footprint (30 × 30 × 30cm) of the Thermo Electron KingFisher® mL instrument delivers automated throughput that makes sense for smaller labs. No external PC required.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**

Protocol	Part#
Technical Bulletin	TB322

Storage Conditions: Store at 22–25°C. Do not freeze the MagneSil® KF Paramagnetic Particles.

MagaZorb® DNA Mini-Prep Kit

Product	Size	Cat.#
MagaZorb® DNA Mini-Prep Kit	50 preps	MB1001
	200 preps	MB1004
	800 preps	MB1008
MagaZorb® DNA Mini-Prep 96-Well Kit	200 preps	MB1101

Available Separately	Size	Conc.	Cat.#
Proteinase K (PK) Solution	16 ml	20 mg/ml	MC5008

Description: The MagaZorb® DNA Kit provides an easy, fast and cost-effective technique for isolating PCR-quality DNA. Using one simple protocol, a high yield of purified DNA can be isolated from a wide variety of sources including whole blood (fresh or frozen, citrate-, heparin- or EDTA-treated), buffy coat, leukocytes, milk, seminal fluid, dried blood spots, cultured cells, tissue (fresh, frozen or formalin-fixed paraffin-embedded), saliva, urine, stool, hair, buccal swabs and vaginal swabs.

Features:

- **Convenient:** Contains all needed reagents so that no reagent preparation is required.
- **Efficient:** Eliminates the need for centrifugation, vacuum filtration or column separation, increasing sample throughput and improving reproducibility.
- **Safe:** Does not require organic solvents, eliminating the need for special storage or waste disposal.

Protocol	Part#
MagaZorb® DNA Kit Technical Bulletin	TB376
MagaZorb® DNA 96-Well Kit Technical Bulletin	TB379

Storage Conditions: Store at 22–25°C.

Wizard® Magnetic 96 DNA Plant System

Product	Size	Cat.#
Wizard® Magnetic 96 DNA Plant System	2 × 96 preps	FF3760
	4 × 96 preps	FF3761

Available Separately	Size	Cat.#
Wash Buffer, Plant	40 ml	A3811

Description: The Wizard® Magnetic 96 DNA Plant System is designed for manual or automated 96-well, high-throughput purification of DNA from plant leaf and seed tissue. The system has been validated with corn and tomato leaf, as well as with canola and sunflower seeds. The DNA purified from these samples can be used in PCR as well as more demanding applications such as RAPD analysis. Unlike column-based systems, the binding of nucleic acids to magnetic particles can occur in solution, enhancing contact with the wash buffer and increasing nucleic acid purity.

Protocols are available for Beckman Coulter instruments.

Features:

- **Improved Productivity:** Manual and automated 96-well protocols cut purification time compared to CTAB extraction.
- **Ease of Handling:** Eliminates organic extractions, multiple centrifugations and cumbersome filter plates.
- **Confidence in Applications Performance:** Validated for both leaf and seed tissue by PCR and RAPD analysis.
- **Automation:** Validated automated methods available at: **www.promega.com/automethods/**
- **Your Choice of Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB289

Storage Conditions: Store at 22–25°C.

Wizard® Magnetic DNA Purification System for Food

Product	Size	Cat.#
Wizard® Magnetic DNA Purification System for Food	200 preps	FF3750
	400 preps	FF3751

Available Separately	Size	Cat.#
Lysis Buffer A, Food	100 ml	A8191
Lysis Buffer B, Food	100 ml	Z3191
Precipitation Solution, Food	150 ml	Z3201

Description: The Wizard® Magnetic DNA Purification System for Food is designed for purification of DNA from a variety of food samples including corn seeds, cornmeal, soybeans, soy flour and soy milk. Processed food, such as corn chips, chocolate and chocolate-containing foods, lecithin and vegetable oils may also be used with the suggested protocol variations. The DNA purified from these samples can be used in PCR-based testing for genetically modified organism (GMO) DNA sequences.

Features:

- **Improved Productivity:** Obtain results in one-third the time of current methods.
- **Ease of Handling:** Requires minimal centrifugation and eliminates organic extractions.
- **Versatility and Robustness:** Validated with a broad variety of foodstuffs, including difficult samples such as lecithin and vegetable oils.

Protocol	Part#
Technical Bulletin	TB284

Storage Conditions: Store at 22–25°C.

Vac-Man® Laboratory Vacuum Manifold

Product	Size	Cat.#
Vac-Man® Laboratory Vacuum Manifold, 20-sample capacity	1 each	A7231
Available Separately	**Size**	**Cat.#**
One-Way Luer-Lok® Stopcocks	10 each	A7261
Vacuum Adapters	20 each	A1331

Description: The Vac-Man® Manifold is a sturdy, chemical-resistant manifold capable of processing 20 samples simultaneously. Each Vac-Man® Manifold is supplied with 20 individual Luer-Lok® Stopcocks, so each port is controlled individually. The Spinlock II Adapters can be attached to the upper row of stopcocks for easy manipulation of the stopcock valves. Promega PureYield™ Plasmid prep columns interface directly with the manifold, while the Wizard® SV minicolumns require the use of Vacuum Adapters for vacuum-based protocols. The Eluator™ Vacuum Elution Device (Cat.# A1071) can be used to elute plasmid DNA from PureYield™ Plasmid Midiprep and Maxiprep columns using vacuum.

Cat.# A7231 Includes:

- Vac-Man® Laboratory Vacuum Manifold, 20-sample capacity
- One-Way Luer-Lok® Stopcocks
- Neoprene Stopper
- Spinlock II Adapters

Features:

- **Improved Productivity:** Process up to 20 samples simultaneously.
- **Flexibility:** Control each vacuum port individually.
- **Safety:** Eliminate the need to handle waste during nucleic acid purification.

Protocol	Part#
Technical Bulletin	TB125

Storage Conditions: Store at 22–25°C.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Vac-Man® Jr. Laboratory Vacuum Manifold

Product	Size	Cat.#
Vac-Man® Jr. Laboratory Vacuum Manifold, 2-sample capacity	1 each	A7660

Description: The Vac-Man® Jr. Laboratory Vacuum Manifold is the ideal system for rapid, effective nucleic acid purification. Each manifold comes with a set of 2 individually controlled One-Way Luer-Lok® Stopcocks, enabling up to 2 miniprep samples to be processed at one time. The Vac-Man® Jr. Laboratory Vacuum Manifold can process one midiprep or maxiprep at a time.

Features:

- **Improved Productivity:** Simultaneously process up to 2 nucleic acid purifications.
- **Flexibility:** Use with standard 1- to 2-liter sidearm flasks.
- **Safety:** Eliminate the need to handle waste during nucleic acid purification.

Protocol	Part#
Technical Bulletin	TB244

Storage Conditions: Store at 22–25°C.

Vac-Man® 96 Vacuum Manifold

Product	Size	Cat.#
Vac-Man® 96 Vacuum Manifold	1 each	A2291
Available Separately	**Size**	**Cat.#**
Collar for Vac-Man® 96 Vacuum Manifold	1 each	A2311

Description: The Vac-Man® 96 Vacuum Manifold is designed for manual use with the Wizard® SV 96 Plasmid DNA Purification System, Wizard® SV 96 Genomic DNA Purification System, Wizard® SV 96 PCR Clean-Up System and the SV 96 Total RNA Isolation System for isolating up to 96 samples simultaneously. The unique manifold design allows the delivery of filtrate waste and wash solutions to an external waste trap (not included), eliminating the need to disassemble the manifold during processing to remove collected waste.

Cat.# A2291 Includes:

- Vacuum Manifold Base (with vacuum port and guide pins)
- Manifold Collar (with vacuum port)
- Manifold Bed (with guide pins)

Features:

- **Safe:** Eliminates the need to handle waste during nucleic acid purification.
- **Flexible:** Adapts for use alone or with robotic nucleic acid purification platforms.

Protocol	Part#
Technical Bulletin	TB272

Storage Conditions: Store at 22–25°C.

Wizard® SV 96 Lysate Clearing Plates

Product	Size	Cat.#
Wizard® SV 96 Lysate Clearing Plates	10 pack	A2241
	100 pack	A2248

Description: The Wizard® SV 96 Lysate Clearing Plates are used with the Wizard® SV 96 Binding Plates (Cat.# A2271, A2278) and the Vac-Man® 96 Vacuum Manifold (Cat.# A2291) for simultaneous lysate clearing and DNA binding in the Wizard® SV 96 (Cat.# A2250, A2255) and Wizard® SV 9600 (Cat.# A2258) Plasmid DNA Purification System protocols.

Wizard® SV 96 Binding Plates

Product	Size	Cat.#
Wizard® SV 96 Binding Plates	10 pack	A2271
	100 pack	A2278

Description: The Wizard® SV 96 Binding Plates are used with the Wizard® SV 96 Plasmid DNA Purification System (Cat.# A2250, A2255), Wizard® SV 96 Genomic DNA Purification System (Cat.# A2370, A2371) and Wizard® SV 96 PCR Clean-Up System (Cat.# A9340, A9341, A9342) to isolate DNA, or with the SV 96 Total RNA Isolation System (Cat.# Z3500, Z3505) to isolate RNA. The isolation procedures can be performed manually or on a robotic platform. The Binding Plates are designed for use with the Vac-Man® 96 Vacuum Manifold (Cat.# A2291) or a comparable manifold.

MagneSil® Reagents

Product	Size	Cat.#
MagneSil® BLUE	100 ml	A2201
MagneSil® RED	100 ml	A1641
MagneSil® GREEN	100 ml	A8231

Description: MagneSil® BLUE and **MagneSil® RED** are used with the Wizard® MagneSil® Plasmid DNA Purification System. The use of the MagneSil® Paramagnetic Particles for lysate clearing (BLUE) as well as DNA capture (RED) circumvents the need for centrifugation or vacuum manifolds.

MagneSil® GREEN is used in the Wizard® MagneSil® Sequencing Reaction Clean-Up System to purify sequencing reactions, including BigDye® terminator DNA sequencing reactions, in a high-throughput system.

MagneSil® Accessories

Product	Size	Cat.#
MagnaBot® 384 Magnetic Separation Device	1 each	V8241
MagnaBot® 96 Magnetic Separation Device	1 each	V8151
MagnaBot® II Magnetic Separation Device	1 each	V8351
MagnaBot® Large Volume Magnetic Separation Device	1 each	V3471
Plate Clamp 96	1 each	V8251
Plate Stand	1 each	V8261
MagnaBot® Adapter T1	1 each	V8481
MagnaBot® Spacer	1 each	V8381
MagnaBot® Spacer 1/8 inch	1 each	V8581
MagnaBot® Spacer 1/16 inch	1 each	V8681
1/4 inch Foam Spacer	1 each	Z3301
Heat Transfer Block	1 each	Z3271
Deep Well MagnaBot® 96 Magnetic Separation Device	1 each	V3031
384-Well Plate, Flat	10 /pk	V5291
384-Well Plate, Conical	10 /pk	V5311
Shaker Top Adapter	1 each	Z3671
Heat Block Insert	1 each	Z3651
Heat Block Adapter, 50ml Tubes	1 each	Z3661
Tube Holder, 50ml Tubes	1 each	Z3631

Cat.# V3031, V3471, V8241, Z3631, Z3651, Z3661, Z3671 For Laboratory Use.

Description: The MagnaBot® 384 Magnetic Separation Device (Cat.# V8241) is designed for high-throughput bioseparation using MagneSil® Paramagnetic Particles in 384-well plates (flat or conical) as an alternative to vacuum filtration and centrifugation.

The MagnaBot® 96 Magnetic Separation Device (Cat.# V8151) is designed for high-throughput bioseparation using magnetic particles such as MagneSil® Paramagnetic Particles, which use the principle of magnetic separation as an alternative to vacuum filtration and centrifugation separation formats. This device is compatible with Collection Plates (Cat.# A9161), 1.2ml, Round-Bottom Deep Well Plates (Cat.# V6771) and Greiner 96-well P.S. V-bottom plates (Greiner Cat.# 651101). A generic 96-well plate may not be compatible.

The MagnaBot® II Magnetic Separation Device (Cat.# V8351) is designed to work with a 96-well PCR plate. A 96-well PCR plate containing MagneSil® Paramagnetic Particles is placed on the unit to draw the particles to the side and away from the bottom of each well. This allows for the quantitative removal of liquids.

The MagnaBot® Large Volume Magnetic Separation Device (Cat.# V3471) is designed for high-throughput bioseparation of magnetic particles, such as MagneSil® Paramagnetic Particles, in large-volume samples (e.g., 2ml deep-well plates). The MagnaBot® Large Volume Magnetic Separation Device is made up of the **Tube Holder, 50ml Tubes (Cat.# Z3631)**, and the Magnetic Base. The Tube Holder has a footprint of a standard multiwell plate and is used to hold up to eight common 50ml conical tubes (Corning or Falcon) for manipulation manually or on the deck of a robotic workstation. The Magnetic Base has a footprint of a standard multiwell plate and is used to capture magnetic particles to the sides of tubes when used with the Tube Holder.

The Plate Clamp 96 (Cat.# V8251) is recommended for automated protocols and designed to ensure that nonskirted PCR plates are uniformly flat for liquid transfer on a robotic platform. Not designed for skirted PCR plates.

The Plate Stand (Cat.# V8261) positions the Plate Clamp 96 assembly on the deck of a robotic workstation.

The MagnaBot® Adapter T1 (Cat.# V8481) is an aluminum holder that centers the MagnaBot® 96 Magnetic Separation Device in a Tecan Genesis® RoMa microplate carrier.

The MagnaBot® Spacers (Cat.# V8381, V8581, V8681) are designed to optimize use of the MagnaBot® 96 Magnetic Separation Device (Cat.# V8151) and the Deep Well MagnaBot® 96 Magnetic Separation Device (Cat.# V3031). The spacers adjust the height of a 96-well plate placed onto the MagnaBot® 96 Magnetic Separation Devices to optimize magnetization. Refer to specific robotic protocols and applications for recommended uses.

The Deep Well MagnaBot® 96 Magnetic Separation Device (Cat.# V3031) is designed for high-throughput bioseparation using magnetic particles such as MagneSil® Paramagnetic Particles for magnetic separation in deep-well plates as an alternative to vacuum filtration and centrifugation separation formats. This MagneSil® technology allows the use of paramagnetic particles for automated nucleic acid purification applications such as plasmid purification, PCR clean-up and dye terminator clean-up prior to automated sequencing.

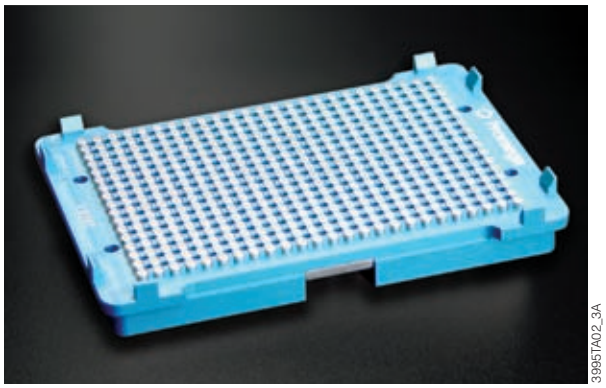
The Heat Block Insert (Cat.# Z3651) and Heat Block Adapter, 50ml Tubes (Cat.# Z3661), are designed to be used with the MagnaBot® Large Volume Magnetic Separation Device for efficient transfer of heat to common (Corning or Falcon) 50ml conical tubes. Both are designed for use with a Fisher Heat Block (Part# 11-718-2, 11-718-6 or 11-718-8) or VWR Heat Block (Part# 13259-032, 13259-036 or 13259-038).

The Shaker Top Adapter (Cat.# Z3671) holds the MagnaBot® Large Volume Magnetic Separation Device on the IKA KS130 Control Orbital Shaker (IKA Works Cat.# 2980100) and is required for use.

Features:

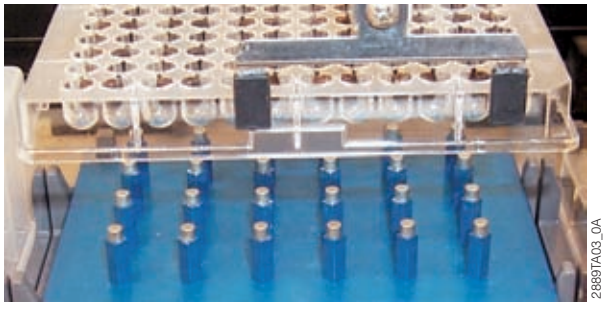
- **Use with Many Platforms:** The product is compatible with numerous instrument platforms including the Tecan Freedom EVO®.
- **Separate Particles Quickly and Easily:** Up to eight 50ml disposable conical tubes (Corning or Falcon) containing paramagnetic particles in suspension are placed on the MagnaBot® Large Volume Magnetic Separation Device with the magnetic plates resting at the interior edge of tubes, one magnetic plate for a pair of tubes. The 4 powerful magnetic plates rapidly attract the magnetic particles to the side and away from the bottom of each tube in seconds, allowing easy removal of the particle-free fluid.

Storage Conditions: Store at 22–25°C.



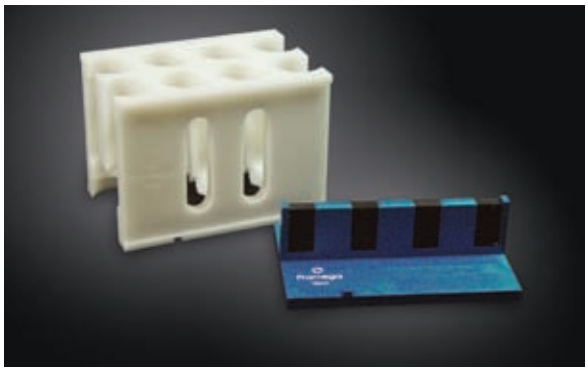
MagnaBot® 384 Magnetic Separation Device (Cat.# V8241).

MagnaBot® 96 Magnetic Separation Device (Cat.# V8151).



MagnaBot® 96 Magnetic Separation Device (Cat.# V8151) with a 96-well Collection Plate and robotic gripper arm.

MagnaBot® II Magnetic Separation Device (Cat.# V8351).



MagnaBot® Large Volume Magnetic Separation Device (Cat.# V3471) with Tube Holder, 50ml Tubes (Cat.# Z3631).

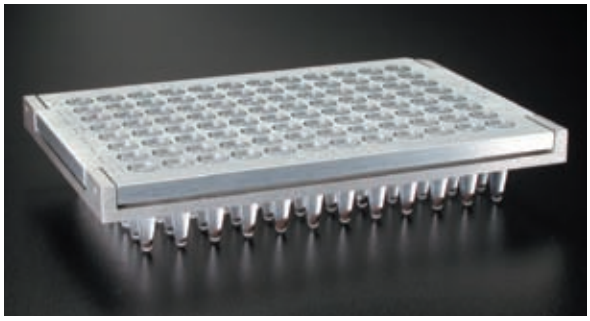


Plate Clamp 96 (Cat.# V8251) with a 96-well PCR plate.

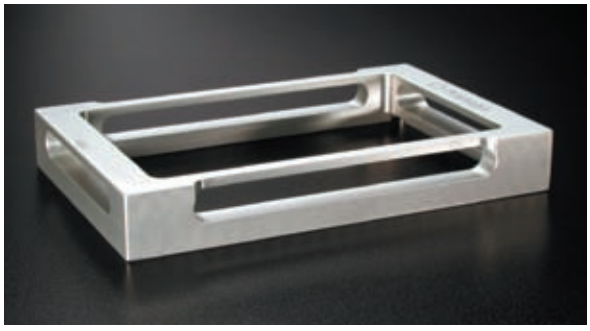


Plate Stand (Cat.# V8261).

Section Contents
Table of Contents

SV Total RNA Isolation System

Product	Size	Cat.#
SV Total RNA Isolation System	10 preps	Z3101
	50 preps	Z3100
	250 preps	Z3105
Available Separately	**Size**	**Cat.#**
Red Blood Cell Lysis Solution (CLB)	200 ml	Z3141
RNA Lysis Buffer (RLA)	50 ml	Z3051

For Laboratory Use.

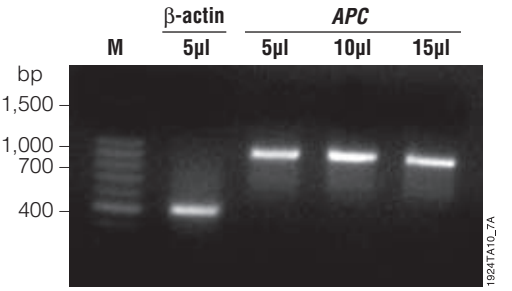
Description: The SV Total RNA Isolation System provides a fast and simple technique for preparation of intact total RNA from tissues, cultured cells and white blood cells in as little as one hour. Using this membrane-based purification system, up to 60mg of tissue can be processed per purification, depending on tissue type. The system incorporates a DNase treatment step directly on the minicolumn membrane. This step substantially reduces genomic DNA contamination, which can interfere with amplification-based methodologies. Purification is achieved without the use of phenol:chloroform extractions or ethanol precipitations, and there is no DNase carryover in the final RNA preparation.

Features:

- **Safety and Efficiency:** Rapid isolation of high yields of total RNA without the use of hazardous compounds like phenol.
- **Flexibility:** Single system for isolation directly from blood, cells or tissue. Two methods available for purification: microcentrifugation (spin) or vacuum.
- **Confidence:** Purified RNA suitable for all routine molecular biology applications, including RT-PCR and Northern blotting.

Protocol	Part#
Technical Manual	TM048

Storage Conditions: Store at 22–25°C.



RNA was 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 β-actin or human *adenomatous polyposis coli (APC)* gene with the Access RT-PCR System (Cat.# A1250). Lane M = 100bp DNA Ladder (Cat.# G2101).

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

Samples	Maximum Amt. to Process	Avg. Yield per Prep (µg)	Avg. Yield per mg Tissue (µg)	A_{260}/A_{280}
Mouse Tissues				
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
Rat Tissues				
Pancreas	30mg	100	3.5	1.9
Heart	60mg	16	0.27	2.1
Lung	60mg	36	0.6	2.1
Bacteria				
E. coli	1×10^9 cells	36	N/A	2.0
Yeast				
S. cerevisiae	4×10^7 cells	19	N/A	2.1
Plant				
Tomato Leaf	30mg	4.6	0.15	2.0
Cell Line				
RAW264.7	5×10^6 cells	51	N/A	2.1

N/A = Not applicable

9485LA

PureYield™ RNA Midiprep System

Product	Size	Cat.#
PureYield™ RNA Midiprep System	10 preps	Z3740
	50 preps	Z3741
Available Separately	**Size**	**Cat.#**
RNA Lysis Buffer (RLA)	50 ml	Z3051
RNA Wash Solution (RWA)	58.8 ml	Z3091
Red Blood Cell Lysis Solution (CLB)	200 ml	Z3141
Eluator™ Vacuum Elution Device	4 each	A1071

For Laboratory Use.

Description: The PureYield™ RNA Midiprep System isolates intact, pure total RNA from essentially any sample type for use in a wide range of applications. The use of a novel Clearing Agent enables the rapid purification of total RNA with undetectable levels of genomic DNA contamination without using DNase. A novel combination of reagents, membranes and protocol leads to yields of up to 1mg of total RNA without organic solvents, protease digestions or alcohol precipitations. One kit can be used to isolate pure total RNA from a wide variety of sample types, such as tissues, cultured cells, bacteria, yeast, plants and blood. The protocol also can be adapted for other sample types.

Commonly used methods provide total RNA that is contaminated with genomic DNA. This contamination can interfere with sensitive methods, such as real-time RT-PCR and microarray analysis. The PureYield™ RNA Midiprep System avoids this problem by selectively removing the genomic DNA prior to total RNA purification. The eluted total RNA is free of detectable DNA and ready for use in sensitive downstream applications.

The system has been designed for use with centrifugation or vacuum (e.g., the Vac-Man® Laboratory Vacuum Manifold) formats.

The Eluator™ Vacuum Elution Device is used to elute nucleic acids from PureYield™ Midiprep or Maxiprep columns. It consists of two pieces, a blue base and a clear column assembly. The base interfaces with a Vacuum Manifold that contains Luer-Lok® fittings, such as the Vac-Man® Laboratory Vacuum Manifold (Cat.# A7231), and holds a 1.5ml tube to capture the eluted nucleic acids. The column assembly accepts PureYield™ Midiprep or Maxiprep columns.

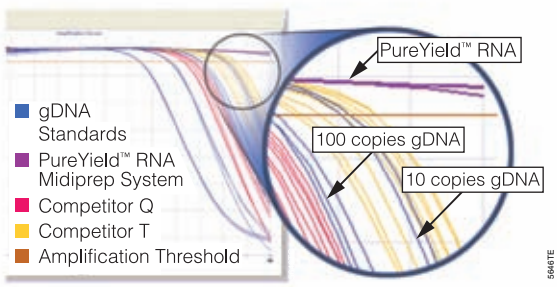
The Eluator™ Device eliminates the requirement for a centrifuge with a swinging bucket rotor for nucleic acid purification, simplifying and speeding purification protocols.

Features:

- **Enhanced Results:** Purified total RNA with undetectable genomic DNA contamination improves results in downstream applications.
- **Improved Productivity:** Purifying total RNA without the use of DNase treatment reduces steps during purification and in downstream applications.
- **Safety and Efficiency:** Rapid purification of high yields of total RNA without the use of hazardous organic solvents.
- **Flexibility:** Single system for purifying total RNA directly from cultured cells, bacteria, yeast, plants and other sample types.

Protocol	Part#
Technical Manual	TM279

Storage Conditions: Store the RNA Lysis Buffer (RLA) with added β-Mercaptoethanol (BME) at 4°C. Store all other components at 22–25°C.



RNA purified with the PureYield™ RNA Midiprep System has no detectable genomic DNA contamination. Total RNA was isolated from 1×10^8 HEK 293T cells using the PureYield™ RNA Midiprep System, a competitor's kit and a competitor's reagent. One hundred nanograms of each total RNA sample was assayed using the Plexor® qPCR System (Cat.# A4011) to detect genomic DNA contamination. Human Genomic DNA (Cat.# G3051) in quantities of 10^4, 10^3, 10^2 and 10^1 copies was used as a standard. The PureYield™ RNA Midiprep System samples showed no detectable genomic DNA. Competitor Q and Competitor T showed an average of 227 and 17 copies, respectively. The horizontal purple line in the upper right corner of this figure indicates no detectable genomic DNA in the PureYield™ RNA Midiprep System sample.

Average Yields of Total RNA Isolated from Tissues and Cells.

Sample Type	Maximum Amount to Process	Average Yield per Prep (µg)[1]	Average A_{260}/A_{230}	Average A_{260}/A_{280}
Rat Tissues				
Liver	300mg	1025.8	1.7	1.8
Lung	300mg	217.0	1.9	2.1
Bacteria				
E. coli	1×10^{10} cells	782.7	2.5	2.1
Cell Line				
HEK 293T	5×10^7 cells	453.3	2.1	1.9
HeLa	5×10^7 cells	329.2	1.8	2.0

[1] The average total RNA yield shown is from a 1ml elution. A second 1ml elution yielded an additional average of 366.4µg (rat liver), 47.0µg (rat lung), 196.8µg (*E. coli*), 45.7µg (HEK 293T cells) and 73.8µg (HeLa cells) of total RNA.

9486LA

RNAgents® Denaturing Solution

Product	Size	Cat.#
RNAgents® Denaturing Solution	120 ml	Z5651

For Laboratory Use.

Description: RNAgents® Denaturing Solution lyses cells or tissue under conditions that rapidly inhibit ribonucleases, using two potent inhibitors of RNase, guanidine thiocyanate and β-mercaptoethanol. The RNAgents® Denaturing Solution is designed to be used in concert with acidic phenol:chloroform and alcohol (isopropanol) for purification of total RNA.

Storage Conditions: Store at 4°C.

SV 96 Total RNA Isolation System

Product	Size	Cat.#
SV 96 Total RNA Isolation System	1 × 96 each	Z3500
	5 × 96 each	Z3505
Available Separately	**Size**	**Cat.#**
RNA Lysis Buffer (RLA)	50 ml	Z3051
RNA Wash Solution (RWA)	58.8 ml	Z3091
Nuclease-Free Water	150 ml	P1195

For Laboratory Use. This product requires the use of a vacuum pump and 96-well vacuum manifold (Vac-Man® 96 Vacuum Manifold, Cat.# A2291).

Description: The SV 96 Total RNA Isolation System provides a high-throughput technique to prepare intact RNA from tissue and cultured cells. Total RNA can be purified from 96 samples in less than an hour without centrifugation. The system also incorporates a DNase treatment step that is designed to substantially reduce genomic DNA contamination, which can interfere with amplification-based methodologies. Purification is achieved without phenol:chloroform extraction or ethanol precipitation, and there is no detectable DNase carryover in the final RNA preparation.

Protocols are available for Beckman Coulter and PerkinElmer instruments.

Features:

- **Confidence in Results:** The product is tested to ensure that purified RNA will perform optimally in RT-PCR.
- **Unique Design:** Novel vacuum manifold eliminates waste handling. Novel plate design prevents cross-contamination during sample processing.
- **Flexibility:** The system is designed for both manual and automated formats.
- **Automation:** Validated automated methods available at: **www.promega.com/automethods/**
- **Your Choice of Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB294

Storage Conditions: Store the SV RNA Lysis Buffer with β-Mercaptoethanol (BME) added at 4°C. Store all other components at 22–25°C.

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

Samples	Maximum Amt. to Process	Avg. Yield per Prep (µg)	Avg. Yield per mg Tissue (µg)	A_{260}/A_{280}
Mouse Tissues				
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
Rat Tissues				
Pancreas	30mg	100	3.5	1.9
Heart	60mg	16	0.27	2.1
Lung	60mg	36	0.6	2.1
Bacteria				
E. coli	1×10^9 cells	36	N/A	2.0
Yeast				
S. cerevisiae	4×10^7 cells	19	N/A	2.1
Plant				
Tomato Leaf	30mg	4.6	0.15	2.0
Cell Line				
RAW264.7	5×10^6 cells	51	N/A	2.1

N/A = Not applicable

9487LA

MagneSil® Total RNA mini-Isolation System

Product	Size	Cat.#
MagneSil® Total RNA mini-Isolation System	4 plate	Z3351

Description: The MagneSil® Total RNA mini-Isolation System provides a high-throughput 96-well format for fast, simple preparation of intact total RNA from small amounts of cell culture (≤1 × 10^5 tissue culture cells), tissue (≤2mg tissue lysate in 100µl) or freshly isolated whole blood (≤20µl). The protocol enables high-throughput automated purification on a variety of liquid-handling workstations. Isolation of total RNA in a 384-well format from cell culture (≤1 × 10^3 cells) and freshly isolated whole blood (≤5µl) also may be performed. Total RNA purification is achieved without vacuum filtration, centrifugation or precipitation. The 96-well total RNA isolation procedure takes about 30 minutes to complete using a liquid-handling workstation.

Total RNA purified using this system is suitable for a variety of molecular biology applications including endpoint RT-PCR amplification and real-time RT-PCR.

Features:

- **Improve Productivity:** Only 30 minutes are required to process one 96-well plate, or 50 minutes for one 384-well plate on a Beckman Coulter Biomek® FX liquid handler.
- **Improve Real-Time PCR Performance:** Elution volumes as low as 15µl provide concentrated RNA without the need for time-consuming vacuum concentration.
- **Gain Confidence in Results:** DNase I treatment is included to remove genomic DNA contamination.
- **Achieve Convenience:** Robotic protocols require no user intervention once you start the automated robotic method.
- **Automate This Assay:** Validated automated methods are available at: **www.promega.com/automethods/**

Protocol	Part#
Technical Bulletin	TB328

Storage Conditions: Store at 22–25°C.

MagaZorb® Total RNA Mini-Prep Kit

Product	Size	Cat.#
MagaZorb® Total RNA Mini-Prep Kit	50 preps	MB2001
	200 preps	MB2004

Description: The MagaZorb® RNA Kit provides an easy, fast and cost-effective technique for isolating PCR-quality total RNA. Using one simple protocol, a high yield of purified total RNA can be isolated from various sources including whole blood (fresh or citrate-, heparin- or EDTA-treated), buffy coat, leukocytes and tissue (fresh or frozen).

Features:

- **Convenient:** Contains all needed reagents so that no reagent preparation is required.
- **Efficient:** Eliminates the need for centrifugation, vacuum filtration or column separation, increasing sample throughput and improving reproducibility.
- **Safe:** Does not require organic solvents, eliminating the need for special storage or waste disposal.

Protocol	Part#
Technical Bulletin	TB378

Storage Conditions: Store at 22–25°C.

PolyATtract® System 1000

Product	Size	Cat.#
PolyATtract® System 1000 with Magnetic Stand	Scalable	Z5420
PolyATtract® System 1000 without Magnetic Stand	Scalable	Z5400
PolyATtract® System 1000 Magnetic Separation Stand	1 each	Z5410
Available Separately	**Size**	**Cat.#**
PolyATtract® GTC Extraction Buffer	120 ml	Z5531

Cat.# Z5420, Z5400, Z5531 For Laboratory Use.

Description: The PolyATtract® System 1000 isolates messenger RNA directly from crude cell or tissue lysates, eliminating the need for total RNA isolations. This system uses the MagneSphere® technology for the purification of poly(A)+ RNA, eliminating the need for oligo(dT) cellulose columns. The increased yield of mRNA using this system allows the detection of low-copy-number mRNAs in relatively small amounts of material using Northern blot analysis. The isolated mRNA is suitable for all molecular biology applications, including in vitro translation, cDNA synthesis, PCR analysis, ribonuclease (RNase) protection assays, primer extension and Northern blots.

The MagneSphere® Technology Magnetic Separation Stands can be used in conjunction with any of the PolyATtract® Systems and are ideal for applications requiring multiple paramagnetic isolations of biomolecules.

Features:

- **Improved Productivity:** mRNA purification directly from tissue or cells in 45 minutes or less. Allows quick collection of magnetic particles.
- **Flexibility:** Works with tissue amounts from 5mg–2g per isolation. Magnetic separation stand (Cat.# Z5410) accommodates 1.5ml, 2ml, 15ml and 50ml tube sizes.
- **Convenience:** No lengthy ethanol precipitation steps, phenol:chloroform extractions, or overnight ultracentrifugation through cesium chloride gradients and lithium chloride (LiCl) precipitations.

Protocol	Part#
Technical Manual	TM228

Storage Conditions: Store at 4°C. Do not freeze the MagneSphere® Paramagnetic Particles.

PolyATtract® mRNA Isolation Systems

Product	Size	Cat.#
PolyATtract® mRNA Isolation System I (Refill for Z5200)	3 isolations	Z5210
PolyATtract® mRNA Isolation System II with Magnetic Stand	3 isolations	Z5200
PolyATtract® mRNA Isolation System III with Magnetic Stand	15 isolations	Z5300
PolyATtract® mRNA Isolation System IV (Refill for Z5300)	15 isolations	Z5310
Available Separately	**Size**	**Cat.#**
Biotinylated Oligo(dT) Probe (50pmol/µl)	35 µl	Z5261
MagneSphere® Technology Magnetic Separation Stand (two-position)	1.5 ml	Z5332
	12 × 75 mm	Z5333

Cat.# Z5210, Z5200, Z5300, Z5310, Z5261 For Laboratory Use.

Description: Cat.# Z5200 contains sufficient reagents for 3 separate mRNA isolations, each from 1–5mg of total RNA. Cat.# Z5210 contains the same reagents as Cat.# Z5200, excluding the Magnetic Separation Stand. Cat.# Z5300 contains sufficient reagents for 15 separate mRNA isolations, each from 100–1,000µg of total RNA. Cat.# Z5310 contains the same reagents as Cat.# Z5300, excluding the Magnetic Separation Stand.

The PolyATtract® mRNA Isolation Systems use the MagneSphere® technology to isolate mRNA rapidly and effectively from total RNA. The systems use a biotinylated oligo(dT) primer to hybridize, at high efficiency in solution, to the 3´ poly(A)+ region present in most mature eukaryotic mRNAs. The hybrids are bound to streptavidin coupled to paramagnetic particles, captured using a magnetic separation stand and washed at high stringency. The mRNA is eluted from the solid phase by the simple addition of ribonuclease-free, deionized water. With total RNA as the starting material, poly(A)+ mRNA is isolated in approximately 45 minutes. The isolated mRNA is suitable for all molecular biology applications, including in vitro translation and cDNA synthesis.

Features:

- **Improved Productivity:** Entire mRNA purification process can be completed in approximately 45 minutes.
- **Highly Pure mRNA:** Due to the strength and selectivity of the interaction between streptavidin and biotin, mRNA bound to the biotinylated oligo(dT) is captured by streptavidin-coated magnetic particles.
- **Confidence in Your Applications:** Isolated mRNA is suitable for use with in vitro translation, RT-PCR and cDNA synthesis.
- **Flexibility:** Configurations for use with large or small amounts of cells and tissues.

Protocol	Part#
Technical Manual	TM021

Storage Conditions: Store at 4°C. Do not freeze the MagneSphere® Paramagnetic Particles.

Magnetic Separation Stands

Product	Size	Cat.#
MagnaBot® 96 Magnetic Separation Device	1 each	V8151
MagneSphere® Technology Magnetic Separation Stand (two-position)	0.5 ml	Z5331
	1.5 ml	Z5332
	12 × 75 mm	Z5333
MagneSphere® Technology Magnetic Separation Stand (twelve-position)	0.5 ml	Z5341
	1.5 ml	Z5342
	12 × 75 mm	Z5343
PolyATtract® System 1000 Magnetic Separation Stand	1 each	Z5410
20-Position Microcentrifuge Tube Magnetic Separator	1 each	CD4002

Description: The MagneSphere® Technology Magnetic Separation Stands can be used in conjunction with any of the PolyATtract® Systems and are ideal for applications requiring multiple paramagnetic isolations of biomolecules. These stands use the same strong rare earth magnet used in the PolyATtract® Systems Magnetic Separation Stands and come in a variety of sizes to accommodate 2–96 samples.

The 20-Position Microcentrifuge Tube Magnetic Separator (Cat.# CD4002) utilizes a microcentrifuge tube rack that can be removed from the high-strength magnets for wash steps or incubation in a water bath. The rack is designed to hold the microcentrifuge tubes so that they will not fall out even when turned upside down, and it can withstand temperatures of up to 80°C for convenient manipulation of sample tubes.

Features:

- **Flexible:** MagneSphere® Technology Stands can be used with both MagneSphere® and PolyATtract® System components.
- **Convenient:** Cat.# Z5331, Z5332, Z5333 enable simultaneous processing of the contents of two tubes of various sizes. Cat.# Z5341, Z5342 and Z5343 enable simultaneous processing of the contents of twelve tubes of various sizes.

Protocol	Part#
Technical Bulletin	TB246

Storage Conditions: Store at 22–25°C.

MagneSphere® Magnetic Separation Stands Compatible with the PolyATtract® Systems.

Stand Cat.#	Sample Size	Compatible Product
2-Position Stand		
Z5331	5–10mg	PolyATtract® System 1000
Z5332	5–35mg	PolyATtract® System 1000 PolyATtract® System III or IV
	1 × 10^6 cells	PolyATtract® System 1000
Z5333	35–100mg	PolyATtract® System 1000 PolyATtract® System I or II
Z5410	0.1–1g or 10^7–10^8 cells	PolyATtract® System 1000
12-Position Stand		
Z5341	5–10mg	PolyATtract® System 1000
Z5342	5–35mg or 1 × 10^6 cells	PolyATtract® System 1000 PolyATtract® System III or IV
Z5343	35–100mg	PolyATtract® System 1000

9488LA

3993TA02_3A

MagnaBot® 96 Magnetic Separation Device (Cat.# V8151).


2312TA07_8A

MagneSphere® Technology Magnetic Separation Stand (two-position) (Cat.# Z5331, Z5332, Z5333).


2309TA07_8A

MagneSphere® Technology Magnetic Separation Stand (twelve-position) (Cat.# Z5341, Z5342, Z5343).


2311TA07_8A

PolyATtract® System 1000 Magnetic Separation Stand (Cat.# Z5410).

Streptavidin MagneSphere® Paramagnetic Particles

Product	Size	Conc.	Cat.#
Streptavidin MagneSphere® Paramagnetic Particles	9ml (15 × 0.6 ml)	1 mg/ml	Z5481
	25 ml	1 mg/ml	Z5482

For Laboratory Use.

Description: The Streptavidin MagneSphere® Paramagnetic Particles (PMPs) may be used in the magnetic separation or purification of a wide variety of biotinylated nucleic acid or protein molecules. The particles are quality-tested and approved for isolation of both nucleic acids and proteins/antibodies.

Features:

- **Confidence:** The Streptavidin MagneSphere® Paramagnetic Particles feature strong, specific binding to biotinylated molecules.
- **Improved Purity:** Enable binding, washing and magnetic separation from undesired materials in a solution.
- **Flexibility:** Applications include purification of DNA, mRNA and proteins.

Protocol	Part#
Technical Bulletin	TB246

Storage Conditions: Store at 4°C. Do not freeze the paramagnetic particles.

Streptavidin

Product	Size	Cat.#
Streptavidin	1 mg	Z7041

For Laboratory Use.

Description: Promega Streptavidin is purified by affinity chromatography and is of the highest quality available.

Storage Conditions: Store at –20°C.

Genetic Identity

Sample Preparation	118
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DNA IQ™ Reference Sample Kit for Maxwell® 16

Product	Size	Cat.#
DNA IQ™ Reference Sample Kit for Maxwell® 16	48 preps	AS1040
Available Separately	**Size**	**Cat.#**
Maxwell® 16 Forensic Instrument	1 each	AS3060
Maxwell® 16 Instrument	1 each	AS2000
Maxwell® 16 SEV Hardware Kit	1 each	AS1200

Cat.# AS1040 For Research Use Only. Not for use in diagnostic procedures. Cat.# AS2000 For Laboratory Use. Cat.# AS3060 Not For Medical Diagnostic Use.

Description: The DNA IQ™ Reference Sample Kit for Maxwell® 16 is designed for optimal DNA extraction from buccal swabs, FTA® blood card punches, liquid blood or other high-concentration DNA reference samples. These samples are typically encountered in forensic, convicted-offender database and paternity testing. The kit contains the same trusted reagents used in the DNA IQ™ System in a convenient prepackaged format and is optimized to yield a final DNA concentration that minimizes the need for concentration or dilution prior to amplification. Liquefied samples are placed directly into the cartridges, and genomic DNA ready for amplification is obtained in approximately 20 minutes.

The Maxwell® 16 Instrument allows efficient, automated purification from a wide range of sample types. The instrument is preprogrammed with purification protocols, which combined with prefilled reagent cartridges, maximize simplicity and convenience. The instrument processes up to 16 samples per instrument run. The purified DNA is of high quality and at high yield and concentration, suitable for direct use in a variety of downstream applications. The Maxwell® 16 Instrument, a magnetic-particle-handling device, purifies DNA using paramagnetic particles, allowing optimal capture, washing and elution of the target material.

The Maxwell® 16 Instrument includes a one-year basic warranty. Several products are offered to extend the warranty. If during the extended warranty period the instrument needs repair under normal use, Promega will be responsible for the repair. Premium warranties offer similar terms and the use of a temporary replacement instrument during the instrument repair period. Please contact Promega for complete warranty terms and limits.

Find out more at: **www.promega.com/maxwell16/**

Features:

- **Maximize Your Time:** Automating DNA extraction reduces hands-on bench time spent manually extracting DNA.
- **Gain Confidence in Your Results:** Instrument design, optimized reagents and automated methods provide consistent yield and purity.
- **Use Trusted DNA IQ™ Chemistry:** The DNA IQ™ System is the recognized leader in automated DNA extraction chemistries and is included in the prefilled Maxwell® 16 reagent cartridges.

Protocol	Part#
Technical Bulletin	TB347

Storage Conditions: Store at 22–25°C.

DNA IQ™ Casework Pro Kit for Maxwell® 16

Product	Size	Cat.#
DNA IQ™ Casework Pro Kit for Maxwell® 16	48 preps	AS1240
Available Separately	**Size**	**Cat.#**
Maxwell® 16 Forensic Instrument	1 each	AS3060
Maxwell® 16 Instrument	1 each	AS2000
LEV Plungers	50 /pk	AS6151

Cat.# AS1240, AS1210, AS3060 Not For Medical Diagnostic Use. AS2000 For Laboratory Use.

Description: The DNA IQ™ Casework Pro Kit for Maxwell® 16 includes newly designed plungers and optimized preprocessing, which results in improved DNA yields.

The DNA IQ™ Casework Pro Kit is designed for optimal DNA extraction from forensic casework samples. These samples may include blood stains, semen stains, hairs, cigarette butts, tissue samples, and trace or "touch" DNA samples regularly encountered in forensic DNA analysis. The kits contain the same trusted reagents used in the DNA IQ™ System in a convenient, prefilled cartridge format and are optimized to provide a final DNA extract in a concentrated format.

The DNA IQ™ Casework Pro Kit for Maxwell® 16 uses a plastic cartridge and plunger that allow DNA elution in a final volume of no more than 50µl. DNA IQ™ Lysis Buffer, Resin and Wash Buffer are included in the prefilled cartridge, and DNA IQ™ Elution Buffer is included in the kit to ensure proper storage of the extract. The DNA IQ™ Casework Pro Kit is compatible with the Maxwell® 16 Forensic Instrument, which includes the hardware necessary to use this kit.

Find out more at: **www.promega.com/maxwell16/**

Features:

- **Reduced Elution Volumes:** Elute your sample in less than 50µl of DNA IQ™ Elution Buffer. No need for post-purification concentration steps.
- **Confidence in Your Chemistry:** The DNA IQ™ System is the recognized leader in automated DNA extraction chemistries and is included in the prefilled Maxwell® 16 reagent cartridges.
- **Preprogrammed Methods:** There is no need for programming or an external computer. The Maxwell® 16 Instrument comes preloaded with all of the necessary methods, which are optimized for maximum performance.

Protocol	Part#
Technical Manual	TM332

Storage Conditions: Store at 15–30°C.

DNA IQ™ System

Product	Size	Cat.#
DNA IQ™ System	100 reactions	DC6701
	400 reactions	DC6700
Tissue and Hair Extraction Kit (for use with DNA IQ™)	100 reactions	DC6740
Available Separately	**Size**	**Cat.#**
Lysis Buffer	150 ml	A8261
2X Wash Buffer	70 ml	A8271
Elution Buffer	50 ml	A8281
DNA IQ™ Resin	50 ml	A8251
MagnaBot® Flat Top Magnetic Separation Device	1 each	V6041
MagneSphere® Technology Magnetic Separation Stand (two-position)	1.5 ml	Z5332
MagneSphere® Technology Magnetic Separation Stand (twelve-position)	1.5 ml	Z5342
DNA IQ™ Spin Baskets	1,000 /bag	V1221
DTT, Molecular Grade (Dry Powder)	5 g	V3151
MagnaBot® 96 Magnetic Separation Device	1 each	V8151
Microtubes, 1.5ml	1,000 /bag	V1231
Proteinase K	100 mg	V3021

Cat.# V3021 For Laboratory Use. Cat.# A8261, A8271, A8281, A8251, V1221 Not For Medical Diagnostic Use.

Description: The DNA IQ™ System is a DNA isolation system designed specifically for forensic and paternity laboratories. This system employs novel paramagnetic particles to isolate clean DNA for use with short tandem repeat (STR) analysis. The DNA IQ™ System can be used to extract DNA from a variety of sample types, including stains and liquid samples.

The unique DNA IQ™ Resin removes PCR inhibitors and contaminants frequently encountered in casework samples. When working with larger sample volumes, such as those found in paternity and databasing, the DNA IQ™ System can deliver a consistent amount of total DNA. Samples including buccal swabs, liquid blood and stains on FTA® and other blood cards have been used with the DNA IQ™ System. More information about sample types that have been used with this product can be found at:
www.promega.com/dnaiqsamples/

Some samples, such as tissue and hair, require pretreatment with proteinase K. The Tissue and Hair Extraction Kit (for use with DNA IQ™) includes Proteinase K and other reagents that aid the break up of most tissue and hair samples and remove proteins and other components from the DNA. The DNA then can be purified using the DNA IQ™ System.

The DNA IQ™ System has been tested with the Plexor® HY System and PowerPlex® 16 and 16 HS Systems to ensure a streamlined process. This translates into reliable products that give optimal results from isolation to quantitation and STR analysis.

Genomic DNA isolation using the DNA IQ™ System has been automated on the Biomek® 2000 and 3000 laboratory automation workstations, as well as the Tecan Freedom EVO® liquid handler. Please contact Promega Technical Services for additional information.

Features:

- **Rapid:** Only a few quick steps to obtain clean DNA with fewer PCR inhibitors.
- **Flexible:** One simple system for use with casework, paternity and database samples.
- **Efficient:** Sensitive to minute sample sizes. In addition, no harmful organic solvents such as phenol and chloroform are used, so use of a hood is not required and disposal of hazardous chemicals is eliminated.

Protocol	Part#
DNA IQ™ System—Small Sample Casework Protocol	TB296
DNA IQ™ System—Database Protocol	TB297
Tissue and Hair Extraction Kit (for use with DNA IQ™) Technical Bulletin	TB307
Automated DNA IQ™ System Protocol for the Beckman Coulter Biomek® 3000	EP033

Storage Conditions: Store the DNA IQ™ System at 22–25°C. Store the Tissue and Hair Extraction Kit (for use with DNA IQ™) at –20°C.

Slicprep™ 96 Device

Product	Size	Cat.#
Slicprep™ 96 Device	10 pack	V1391

For Laboratory Use.

Description: The Slicprep™ 96 Device allows solid material to be incubated with a solution in a basket that is placed in a deep-well plate. Following incubation, the basket is raised with a collar for an additional 0.5ml of space below the basket. This allows removal of the incubation liquid and solubilized material from the solid support without having to transfer material to another tube or plate. One-millimeter holes in the bottom of the basket allow rapid flow of liquid in and out of the baskets. The device is manufactured with polypropylene to reduce adsorption of biological material onto the plastic and give it strength and stability over a wide temperature range. The components are manufactured and assembled in a HEPA-filtered clean room with gloved and gowned personnel to reduce the chance of DNA contamination.

The package contains 10 units of the Slicprep™ 96 Device. Each unit consists of three components: the 96 Spin Basket, 96 Deep Well Plate and U-Shaped Collar, which is used to raise the baskets during centrifugation.

Storage Conditions: Store at 22–25°C.

Differex™ System

Product	Size	Cat.#
Differex™ System	50 samples	DC6801
	200 samples	DC6800
Differex™ Digestion Buffer	150 ml	A8501
Differex™ Separation Solution	40 ml	A8511
Manual Differex™ Magnet	1 each	V1591
Available Separately	**Size**	**Cat.#**
Lysis Buffer	150 ml	A8261
DNA IQ™ Resin	50 ml	A8251
2X Wash Buffer	70 ml	A8271
Elution Buffer	50 ml	A8281
DTT, Molecular Grade (Dry Powder)	5 g	V3151
Proteinase K	100 mg	V3021
MagnaBot® Flat Top Magnetic Separation Device	1 each	V6041

Cat.# DC6801, DC6800, A8501, A8511, A8261, A8251, A8271, A8281 Not for Medical Diagnostic Use. Cat.# V3021 For Laboratory Use.

Description:

The Differex™ System extracts sexual assault samples easily and quickly. It provides a simple and fast method for separating male and female fractions of a sample, making it possible to analyze samples more quickly and efficiently.

The Differex™ System offers recovery similar to that of the standard method commonly used for differential extraction. The Differex™ System is used in combination with the DNA IQ™ System and Slicprep™ 96 Device on robotic platforms to extract up to 48 differential extractions in less than 5 hours, including incubation time, and less than 1 hour of hands-on time.

Automated Differex™ System methods are available for the Biomek® 2000 and 3000 laboratory automation workstations, as well as the Tecan Freedom EVO® liquid handler. Please contact Promega Technical Services for additional information. A manual protocol for the Differex™ System is included for laboratories not yet using robotic platforms for DNA extraction.

Features:

- **Automated Differential Extractions:** The Differex™ System is the first and only system that allows a forensic laboratory to automate every step of differential extraction.
- **Direct Compatibility with the DNA IQ™ System and Downstream STR Applications:** Clean DNA extracts mean you can be confident in your ability to obtain results regardless of your choice of STR systems.
- **Robust Results With Even Tough Samples:** The Differex™ System works with challenging new and old samples typical of those from sexual assaults.
- **More Information About Automated Differex™ System: www.promega.com/applications/hmnid/automation/automation_sp.htm**

Protocol	Part#
Automated Differex™ System Protocol for the Tecan Freedom EVO® System	EP030
Automated Differex™ System Protocol for the Beckman Coulter Biomek® 2000	EP031
Automated Differex™ System Protocol for the Beckman Coulter Biomek® 3000	EP032
Differex™ System Technical Bulletin	TBD020
Differex™ System—For Use With the Differex™ Magnet Technical Manual	TM331

Storage Conditions: Store at room temperature.

Manual Differex™ Magnet.

Genetic Identity Automation Hardware

Product	Size	Cat.#
Shaker Integration Plate	1 each	V3691
Deep Well Heat Transfer Block	1 each	V6741
VARIOMAG® Teleshake (110V, for North America use only)	1 each	V6751
V&P Scientific Heating Block (110V, North America use only)	1 each	V6761
1.2ml, Round-Bottom Deep Well Plate	50 /case	V6771
2.2ml, Square-Well Deep Well Plate	50 /case	V6781
Pyramid-Bottom Reservoir, 12 Column	25 /case	V6791
Pyramid-Bottom Reservoir	25 /case	V6801
U-Bottom Microplate	50 /case	V6811
1.1ml, Square-Well, V-Bottom Deep Well Plate	25 /case	V6821
10ml, 24-Well Deep Well Plate	25 /case	V6831

Cat.# V3691, V6741 For Laboratory Use.

Description: The Genetic Identity Automation Hardware can be used on automated platforms in conjunction with Promega Genetic Identity products. Please contact Technical Services for specific application and platform information.

Plexor® HY System

Product	Size	Cat.#
Plexor® HY System	200 reactions	DC1001
	800 reactions	DC1000
Available Separately	**Size**	**Cat.#**
Plexor® Calibration Kit, Set A	1 each	DC1500
Water, Amplification Grade	6,250 µl (5 × 1,250 µl)	DW0991

Cat.# DC1000, DC1001, DC1500 Not for Medical Diagnostic Use. Cat.# DW0991 For Laboratory Use.

Description: The Plexor® HY System is a real-time PCR assay to determine the concentration of total human DNA and male human DNA simultaneously in one reaction. The kit contains an internal PCR control (IPC) to test for false-negative results that may occur in the presence of PCR inhibitors and a melt curve function to confirm that the correct product was amplified.

Plexor® HY is a sensitive multiplex kit that routinely detects approximately 6.4pg total DNA. PCR setup is performed at room temperature and is compatible with automated platforms.

The Plexor® Systems work by measuring a reduction in fluorescent signal during amplification. Amplification of each target uses only two primers, one of which contains both a fluorescent tag and a modified base. As amplification proceeds, fluorescence is reduced by site-specific incorporation of a fluorescent quencher opposite the complementary modified base. The quencher is in close proximity to a fluorescent dye located on the end of the primer, resulting in a reduction of fluorescent signal. After PCR, a melt analysis can be performed to provide an internal control for the final assay design or to expedite troubleshooting.

The Plexor® HY System is optimized for use on the Applied Biosystems 7500 and 7500 FAST real-time PCR systems and Stratagene Mx3005P® and Mx3000P® qPCR systems. For information about use with other qPCR instrumentation, contact Promega Technical Services.

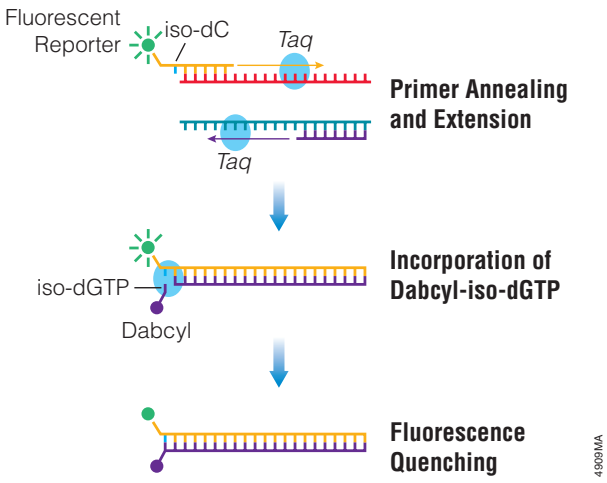
The Plexor® Analysis Software is available for free download at: **www.promega.com/plexorhy/**. The unique functions of this software allow you to quickly and easily review data and create reports. Replicate samples are automatically averaged, template amounts are calculated and the necessary volume of DNA is displayed for your optimized STR amplification conditions.

Features:

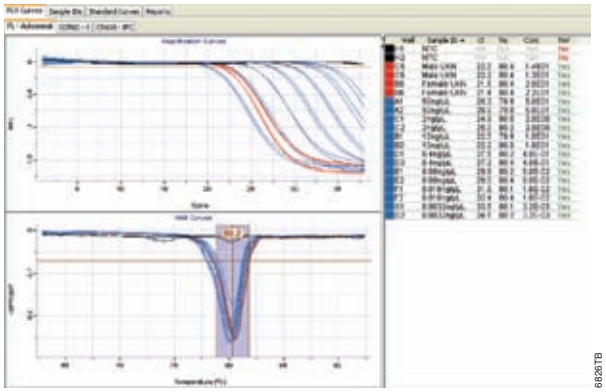
- **Simultaneous Quantification of Autosomal and Y-Chromosome DNA:** Less variability, less time, more valuable data.
- **Consistent and Reproducible Detection of 6.4pg of DNA:** If you can't detect it with Plexor® HY, you can't detect it with your STR system.
- **Internal Positive Control and Melt-Curve Analysis:** Guard against false-negative and false-positive results, allowing you to be confident in your data.

Protocol	Part#
Plexor® HY System for the Applied Biosystems 7500 and 7500 FAST Real-Time PCR Systems Technical Manual	TM293
Plexor® HY System for the Stratagene Mx3000P® and Mx3005P™ Quantitative PCR Systems Technical Manual	TM294
Plexor® HY System for the Bio-Rad iQ™5 Real-Time PCR Detection System Technical Manual	TM296
Plexor® HY System for the Corbett Rotor-Gene 6000 Series Detection System Technical Manual	TM299
Plexor® HY System for the Roche LightCycler® 480 Technical Manual	TM302

Storage Conditions: Store at –20°C.



Schematic diagram illustrating the Plexor® real-time PCR process.



Autosomal amplification curves and melt curves from a Plexor® HY amplification.

PowerPlex® S5 System

Product		Size	Cat.#
PowerPlex® S5 System		100 reactions	DC6951
		400 reactions	DC6950
Available Separately	**Size**	**Conc.**	**Cat.#**
9947A DNA	250 ng	10 ng/µl	DD1001
PowerPlex® Matrix Standards, 3100/3130	25 µl (each dye)		DG4650
PowerPlex® Matrix Standards, 310	50 µl (each dye)		DG4640

Not for Medical Diagnostic Use.

Description: The PowerPlex® S5 System is a mini-STR kit that allows co-amplification and detection of four STR markers (D18S51, D8S1179, TH01 and FGA) plus Amelogenin. One primer specific for each of the Amelogenin, D18S51 and D8S1179 loci is labeled with fluorescein (FL), and one primer specific for each of the TH01 and FGA loci is labeled with 6-carboxy-4′,5′-dichloro-2′,7′-dimethoxy-fluorescein (JOE). All five loci are amplified simultaneously in a single tube and analyzed in a single injection. The four STR loci are included in the CODIS and European databases. The amplicons for all loci are smaller than 260bp. It was the first Promega STR kit to include hot-start *Taq* DNA polymerase, which is included in the PowerPlex® S5 5X Master Mix. The PowerPlex® S5 System is primarily a screening tool, but it also can be used as a mini-STR casework kit.

Features:

- **Sensitive:** Generate full DNA profiles with as little as 50pg of DNA.
- **Easy to Use:** The PowerPlex® S5 System comes complete with premixed primer pairs, a master mix with *Taq* DNA polymerase and internal lane standard. The simplified thermal cycling protocol requires no ramping, and the system is compatible with a number of instrument platforms, including ABI PRISM® 310, 3100 and 3100-*Avant* and Applied Biosystems 3130 and 3130*xl* Genetic Analyzers.
- **Robust:** The PowerPlex® S5 System is more tolerant of DNA degradation and less sensitive to inhibitors. Full DNA profiles can be achieved in the presence of 130µM hematin, 200ng tannic acid or 150ng humic acid.

Protocol	Part#
PowerPlex® S5 System Technical Manual	TMD021
PowerPlex® Matrix Standards, 310, Technical Bulletin	TBD021
PowerPlex® Matrix Standards, 3100/3130, Technical Bulletin	TBD022

Storage Conditions: Store at –20°C.

PowerPlex® 16 System

Product	Size	Cat.#
PowerPlex® 16 System	100 reactions	DC6531
	400 reactions	DC6530
Available Separately	**Size**	**Conc.**	**Cat.#**
9947A DNA	250 ng	10 ng/µl	DD1001
PowerPlex® Matrix Standards, 310	50 µl (each dye)		DG4640
PowerPlex® Matrix Standards, 3100/3130	25 µl (each dye)		DG4650
PowerTyper™ Macros (Release 2.0)	1 CD-ROM		DG3470
Blue Dextran Loading Solution	3 × 1 ml		DV4351

Cat.# DV4351 For Laboratory Use. All other Cat.# Not for Medical Diagnostic Use.

Description: The PowerPlex® 16 System is a multiplex STR system for use in DNA typing, including paternity testing, forensic DNA analysis, human identity testing and tissue culture strain identification. The system allows co-amplification and three-color detection of sixteen loci (fifteen STR loci and Amelogenin): Penta E, D18S51, D21S11, TH01, D3S1358, FGA, TPOX, D8S1179, vWA, Amelogenin, Penta D, CSF1PO, D16S539, D7S820, D13S317 and D5S818. One primer for each of the Penta E, D18S51, D21S11, TH01 and D3S1358 loci is labeled with fluorescein (FL); one primer for each of the FGA, TPOX, D8S1179, vWA and Amelogenin loci is labeled with carboxy-tetramethylrhodamine (TMR); and one primer for each of the Penta D, CSF1PO, D16S539, D7S820, D13S317 and D5S818 loci is labeled with 6-carboxy-4′,5′-dichloro-2′,7′-dimethoxy-fluorescein (JOE). All sixteen loci are amplified simultaneously in a single tube and analyzed in a single injection or gel lane.

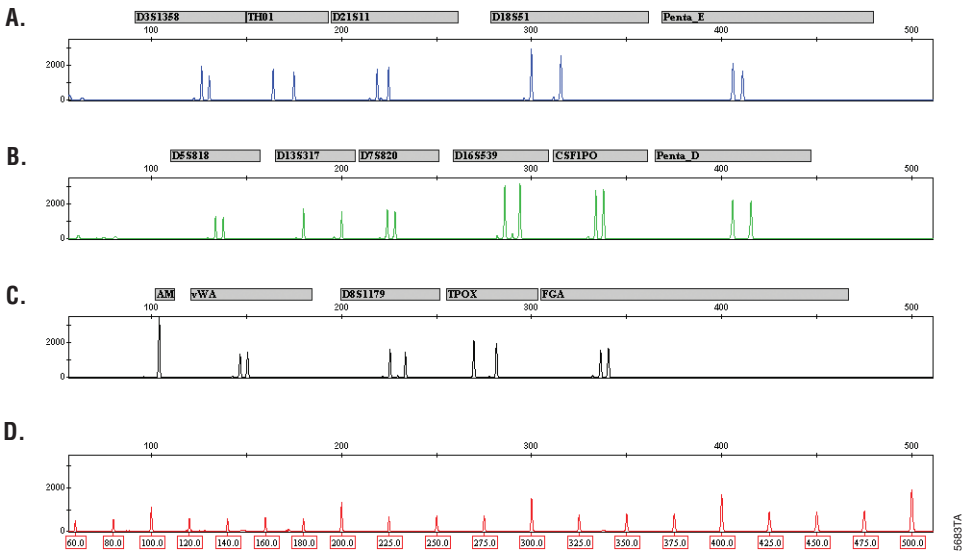
The PowerPlex® 16 System is compatible with the ABI PRISM® 310, 3100 and 3100-*Avant* Genetic Analyzers, Applied Biosystems 3130 and 3130*xl* Genetic Analyzers and ABI PRISM® 377 DNA Sequencer.

Features:

- **Single Amplification:** Amplify all 13 CODIS STR loci in one reaction.
- **Maximum Sensitivity:** Analyze minute quantities of DNA. The system is optimized for use with 1ng of DNA; however, each lot is performance tested to provide reproducible results with 0.5ng of DNA. Additional studies show interpretable results can be obtained with <0.5ng of DNA.
- **Internal Lane Standard 600:** This marker offers the greatest precision available for DNA typing. It is used in each gel lane or capillary injection to increase the precision of analysis.
- **Allelic Ladders:** Allelic ladders for all 16 loci are provided to simplify interpretation.
- **Automatic Assignment of Genotypes:** Panel and bin files are required for use with GeneMapper® *ID* software and are available for download at: **www.promega.com/geneticidtools/**. The PowerTyper™ 16 Macro facilitates data analysis, allowing automatic assignment of genotypes using the Genotyper® software. The PowerTyper™ Macros can be downloaded from: **www.promega.com/geneticidtools/** or ordered on CD-ROM.
- **Validation Studies:** The PowerPlex® 16 System has undergone extensive validation efforts including internal validation, CODIS database validation and forensic casework validation.

Protocol	Part#
PowerPlex® 16 System Technical Manual	TMD012
PowerPlex® Matrix Standards, 310, Technical Bulletin	TBD021
PowerPlex® Matrix Standards, 3100/3130, Technical Bulletin	TBD022

Storage Conditions: Store at –20°C. The fluorescent primer pairs and allelic ladders are light-sensitive; therefore, minimize light exposure.



The PowerPlex® 16 System. A single DNA template (0.5ng) was amplified using the PowerPlex® 16 10X Primer Pair Mix. Amplification products were mixed with Internal Lane Standard 600 and analyzed using an Applied Biosystems 3130 Genetic Analyzer and a 3kV, 5-second injection. Results were analyzed using GeneMapper® *ID* software, version 3.2. **Panel A.** Peaks of the fluorescein-labeled loci: D3S1358, TH01, D21S11, D18S51 and Penta E. **Panel B.** Peaks of the JOE-labeled loci: D5S818, D13S317, D7S820, D16S539, CSF1PO and Penta D. **Panel C.** Peaks of the TMR-labeled loci: Amelogenin, vWA, D8S1179, TPOX and FGA. **Panel D.** An electropherogram showing the Internal Lane Standard 600 fragments.

PowerPlex® 16 HS System

Product	Size	Cat.#
PowerPlex® 16 HS System	100 reactions	DC2101
	400 reactions	DC2100

Available Separately	Size	Conc.	Cat.#
9947A DNA	250 ng	10 ng/µl	DD1001
PowerPlex® Matrix Standards, 310	50 µl (each dye)		DG4640
PowerPlex® Matrix Standards, 3100/3130	25 µl (each dye)		DG4650
Water, Amplification Grade	6,250 µl (5 × 1,250 µl)		DW0991

Cat.# DW0991 For Laboratory Use. All other Cat.# Not for Medical Diagnostic Use.

Description: The PowerPlex® 16 HS System is a multiplex STR system for use in DNA typing. This system co-amplifies the loci D18S51, D21S11, TH01, D3S1358, Penta E (labeled with fluorescein); FGA, TPOX, D8S1179, vWA and Amelogenin (labeled with TMR); CSF1PO, D16S539, D7S820, D13S317, D5S818 and Penta D (labeled with JOE). This multiplex includes all 13 CODIS STR markers, Amelogenin for gender determination and two low-stutter, highly discriminating pentanucleotide STR markers. All sixteen loci are amplified simultaneously in a single tube and analyzed in a single injection. The PowerPlex® 16 HS System is compatible with ABI PRISM® 310, 3100 and 3100-*Avant* Genetic Analyzers and Applied Biosystems 3130, 3130*xl*, 3500 and 3500*xl* Genetic Analyzers.

For more information, visit: **www.promega.com/powerplex16hs/product.htm**

Features:

- **Robustness:** The PowerPlex® 16 HS System is more tolerant of PCR inhibitors than competing STR systems and the previous version of PowerPlex® 16. Generate profiles with samples that previously failed to amplify. Avoid costly and time-consuming sample cleanup.
- **Sensitivity:** Each lot is quality tested to produce full profiles from 100pg of DNA. Gain confidence in analysis of limited samples.
- **High Discrimination:** The loci included in PowerPlex® 16 HS are more discriminating than competitive systems and are ideal for resolving partial matches or challenging familial cases.
- **Proven Design:** Primer sequences, dyes and ladders are all unchanged from PowerPlex® 16. Expect concordance with existing databases.
- **Complete System:** PowerPlex® 16 HS includes size standard, amplification-grade water and *Taq* DNA polymerase already in the master mix. Simple to order, easy to use.

Protocol	Part#
Technical Manual	TMD022

Storage Conditions: Store at –20°C.

PowerPlex® 16 BIO System

Product	Size	Cat.#
PowerPlex® 16 BIO System	100 reactions	DC6541
	400 reactions	DC6540

Available Separately	Size	Conc.	Cat.#
9947A DNA	250 ng	10 ng/µl	DD1001

Not for Medical Diagnostic Use.

Description: The PowerPlex® 16 BIO System is a multiplex STR system for use in DNA typing, including paternity testing, forensic DNA analysis, human identity testing and tissue culture strain identification. The system allows co-amplification and three-color detection of sixteen loci (fifteen STR loci and Amelogenin): Penta E, D18S51, D21S11, TH01, D3S1358 (labeled with fluorescein); FGA, TPOX, D8S1179, vWA, Amelogenin (labeled with Rhodamine Red™-X); and Penta D, CSF1PO, D16S539, D7S820, D13S317, D5S818 (labeled with JOE). The amplified STRs are detected using the Hitachi FMBIO® II Fluorescence Imaging System.

High-throughput analysis is achieved by using the available software to compare amplified DNA fragments directly with the allelic ladder provided for each locus. The allelic ladder mixture consists of most or all known alleles in the population and is included to allow rapid and precise assignment of alleles. The size marker (Internal Lane Standard 600 BIO) provided with the system is tagged with a fourth fluorescent dye. This marker is included as a standard to increase precision of analysis when using the PowerPlex® 16 BIO System.

The Matrix 16 BIO is provided to help generate a color matrix for color separation using a filter set* and the Hitachi FMBIO® Analysis Software. The Matrix 16 BIO consists of fragments labeled with four different fluorescent dyes: Fluorescein, JOE, Rhodamine Red™-X and Texas Red®-X. These four dyes are the same as those used in the PowerPlex® 16 BIO System.

*Filter sets (MiraiBio Cat.# 11999-246-00) are available for purchase from MiraiBio, Inc., at 888-615-9600 (USA) or 650-615-7600.

Features:

- **One Amplification:** Amplify all 13 CODIS STR loci in one reaction.
- **Sensitivity:** The system is designed to amplify 0.5–1ng of DNA.
- **Internal Lane Standard (ILS) 600 BIO:** The marker is used in each gel lane to increase precision in analyses. The ILS 600 BIO contains 21 evenly balanced fragments of 80–600bp.
- **Allelic Ladder Mix:** Allelic ladders for all 16 loci, labeled with three fluorescent dyes, are provided to simplify interpretation of alleles.
- **Matrix 16 BIO:** This tool is provided to aid color separation.

Protocol	Part#
Technical Manual	TMD016

Storage Conditions: Store at –20°C. The fluorescent primer pairs and allelic ladders are light-sensitive; therefore, minimize light exposure.

PowerPlex® Y System

Product	Size	Cat.#
PowerPlex® Y System	50 reactions	DC6761
	200 reactions	DC6760
Available Separately	**Size**	**Conc.**	**Cat.#**
9948 Male DNA	250 ng	10 ng/µl	DD2061
9947A DNA	250 ng	10 ng/µl	DD1001
PowerPlex® Matrix Standards, 310	50 µl (each dye)		DG4640
PowerPlex® Matrix Standards, 3100/3130	25 µl (each dye)		DG4650
PowerTyper™ Macros (Release 2.0)	1 CD-ROM		DG3470
Blue Dextran Loading Solution	3 × 1 ml		DV4351

Cat.# DV4351 For Laboratory Use. All other Cat.# Not for Medical Diagnostic Use.

Description: The PowerPlex® Y System allows co-amplification and three-color detection of twelve loci. The system contains primers for the loci DYS19, DYS385a/b, DYS389I/II, DYS390, DYS391, DYS392, DYS393, DYS437, DYS438 and DYS439. One primer specific for each of the DYS389I/II, DYS391 and DYS439 loci is labeled with fluorescein (FL). One primer specific for each of the DYS385a/b, DYS390 and DYS393 loci is labeled with carboxy-tetramethylrhodamine (TMR). One primer specific for each of the DYS19, DYS392, DYS437 and DYS438 loci is labeled with 6-carboxy-4′,5′-dichloro-2′,7′-dimethoxy-fluorescein (JOE).

All twelve loci are amplified simultaneously in a single tube and analyzed in a single injection or gel lane. Fragment sizing is provided by an internal size standard (Internal Lane Standard 600) labeled with carboxy-X-rhodamine (CXR). Color deconvolution can be performed using matrix standards available from Promega.

The PowerPlex® Y System is compatible with the ABI PRISM® 310, 3100 and 3100-*Avant* Genetic Analyzers, Applied Biosystems 3130 and 3130*xl* Genetic Analyzers and ABI PRISM® 377 DNA Sequencer.

Features:

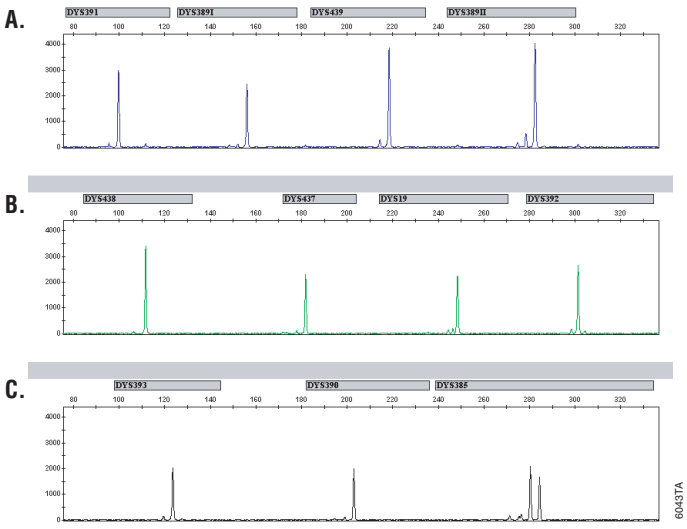
- **Amplification of 12 Y-STR Loci in One Reaction:** Amplify the nine loci defined as the European minimal haplotype and endorsed by ISFG (International Society of Forensic Genetics)* plus the two loci added to this panel by SWGDAM (Scientific Working Group on DNA Analysis Methods).

Note: The data in the PowerPlex® Y Haplotype Database have been added to the US Y-STR Database, a searchable listing of 11- to 17-locus Y-STR haplotypes. For Y-STR haplotype searches, visit the US Y-STR Database at: **www.usystrdatabase.org** or Y-Chromosome Haplotype Reference Database at: **www.yhrd.org**
*ISFG-endorsed loci are DYS19, DYS385a/b, DYS389I/II, DYS390, DYS391, DYS392, DYS393. SWGDAM-endorsed loci are DYS19, DYS385a/b, DYS389I/II, DYS390, DYS391, DYS392, DYS393, DYS438 and DYS439.

- **Maximum Sensitivity:** The system is optimized for use with 0.5–1ng of template DNA. However, each lot is performance tested to provide reproducible results with 0.25ng of DNA. Additional studies show interpretable results can be obtained with <0.25ng of DNA.
- **Superior Specificity—You Amplify Only Male DNA:** Each lot is performance tested to ensure no amplification with up to 100ng of female DNA. Additional studies show no amplification with >100ng of female DNA. This makes the PowerPlex® Y System a great choice to analyze "heavy" mixture samples.
- **Short PCR Products:** Amplification products are all less than 335 bases, which increases the likelihood of obtaining a full profile from degraded samples.
- **No Locus Overlap:** Primers were designed so that alleles for each locus do not overlap with a neighboring locus, reducing the likelihood of misinterpretation.
- **Typing of Samples with Greater Confidence:** Allelic ladders for all 12 loci are provided to simplify interpretation through the use of the PowerTyper™ Macro. The wide span of alleles included in the ladder allow typing of more rare alleles.
- **Automatic Assignment of Genotypes:** The PowerTyper™ Y Macro automatically labels fragments from Genotyper® data. The PowerTyper™ Macros can be downloaded from: **www.promega.com/geneticidtools/** or ordered on CD-ROM. Panel and bin files for use with GeneMapper® *ID* are available for download at: **www.promega.com/geneticidtools/**

Protocol	Part#
PowerPlex® Y System Technical Manual	TMD018
PowerPlex® Matrix Standards, 310, Technical Bulletin	TBD021
PowerPlex® Matrix Standards, 3100/3130, Technical Bulletin	TBD022

Storage Conditions: Store at –20°C. The fluorescent primer pairs and allelic ladders are light-sensitive; therefore, minimize light exposure.



The PowerPlex® Y System. A single template DNA (0.5ng) was amplified using the PowerPlex® Y 10X Primer Pair Mix. Amplification products were analyzed using an Applied Biosystems 3130 Genetic Analyzer and a 3kV, 5-second injection. Results were analyzed using GeneMapper® *ID* software, version 3.2. **Panel A.** An electropherogram showing the fluorescein-labeled loci: DYS389I/II, DYS391 and DYS439. **Panel B.** An electropherogram showing the JOE-labeled loci: DYS19, DYS392, DYS437 and DYS438. **Panel C.** An electropherogram showing the TMR-labeled loci: DYS385a/b, DYS390 and DYS393.

PowerPlex® ESX and ESI Systems

Product	Size	Cat.#
PowerPlex® ESX 16 System	100 reactions	DC6711
	400 reactions	DC6710
PowerPlex® ESX 17 System	100 reactions	DC6721
	400 reactions	DC6720
PowerPlex® ESI 16 System	100 reactions	DC6771
	400 reactions	DC6770
PowerPlex® ESI 17 System	100 reactions	DC6781
	400 reactions	DC6780
PowerPlex® ESX 17 System and PowerPlex® ESI 17 System Bundle	400 reactions each	DC6790
	100 reactions each	DC6791
PowerPlex® ESX 16 System and PowerPlex® ESI 16 System Bundle	400 reactions each	DC6792
	100 reactions each	DC6793

Available Separately	Size	Conc.	Cat.#
PowerPlex® 5-Dye Matrix Standards, 310	50 µl (each dye)		DG4600
PowerPlex® 5-Dye Matrix Standards, 3100/3130	25 µl (each dye)		DG4700
CC5 Internal Lane Standard 500	150 µl		DG1521
9947A DNA	250 ng	10 ng/µl	DD1001
Water, Amplification Grade	6,250 µl (5 × 1,250 µl)		DW0991

Cat.# DC6711, DC6710, DC6721, DC6720, DC6771, DC6770, DC6781, DC6780, DC6790, DC6791, DC6792, DC6793 Not for Medical Diagnostic Use. For in vitro use only. Cat.# DG4600, DG4700, DD1001 Not for Medical Diagnostic Use. Cat.# DG1521, DW0991 For Laboratory Use.

Description: The PowerPlex® ESX and ESI Systems meet the new ENFSI (European Network of Forensic Science Institutes) recommendations for DNA profile sharing across Europe. They are based on five-color technology and allow co-amplification and detection of D3S1358, D8S1179, D18S51, D21S11, FGA, TH01, vWA, D2S441, D10S1248, D22S1045, D1S1656, D12S391, D2S1338, D16S539, D19S433, SE33 and Amelogenin. These kits are available in multiple formats, including the option to detect SE33, to accommodate various requirements and preferences. The kits have increased tolerance to common inhibitors and increased sensitivity to obtain full profiles from low-level DNA and are robust enough to genotype degraded DNA samples through the use of mini-STR loci. This system is compatible with ABI PRISM® 310, 3100 and 3100-*Avant* and Applied Biosystems 3130, 3130*xl*, 3500 and 3500*xl* Genetic Analyzers.

Features:

- **More Kit Configurations:** Satisfy your specific needs for processing database and casework samples.
- **More Loci:** Amplify all ENFSI-recommended loci, with or without SE33.
- **More Mini-STRs:** Obtain more complete profiles from degraded DNA.
- **More Robust Buffer:** Achieve better results with inhibited samples.
- **More Complete Kit:** Complete system includes *Taq* DNA polymerase in a convenient master mix, size standard and amplification-grade water.

Protocol	Part#
PowerPlex® ESX 16 System Technical Manual	TMD023
PowerPlex® ESX 17 System Technical Manual	TMD024
PowerPlex® ESI 16 System Technical Manual	TMD027
PowerPlex® ESI 17 System Technical Manual	TMD028
PowerPlex® 5-Dye Matrix Standards, 310, Technical Bulletin	TBD023
PowerPlex® 5-Dye Matrix Standards, 3100/3130, Technical Bulletin	TBD024

Storage Conditions: Store at –20°C.

PowerPlex® ES System

Product	Size	Cat.#
PowerPlex® ES System	100 reactions	DC6731
	400 reactions	DC6730

Available Separately	Size	Conc.	Cat.#
9947A DNA	250 ng	10 ng/µl	DD1001
PowerPlex® Matrix Standards, 310	50 µl (each dye)		DG4640
PowerPlex® Matrix Standards, 3100/3130	25 µl (each dye)		DG4650
PowerTyper™ Macros (Release 2.0)	1 CD-ROM		DG3470

Not for Medical Diagnostic Use.

Description: The PowerPlex® ES System is a multiplex STR system for use in DNA typing, including paternity testing, forensic DNA analysis, human identity testing and tissue culture strain identification. This system co-amplifies the loci D3S1358, TH01, D21S11, D18S51 (labeled with fluorescein), Amelogenin, vWA, D8S1179, FGA (labeled with TMR) and SE33 (also known as ACTBP2, labeled with JOE).

The PowerPlex® ES System includes the seven core STR loci for ENFSI as well as Amelogenin. The PowerPlex® ES System types all loci necessary for the German National Database (DAD).

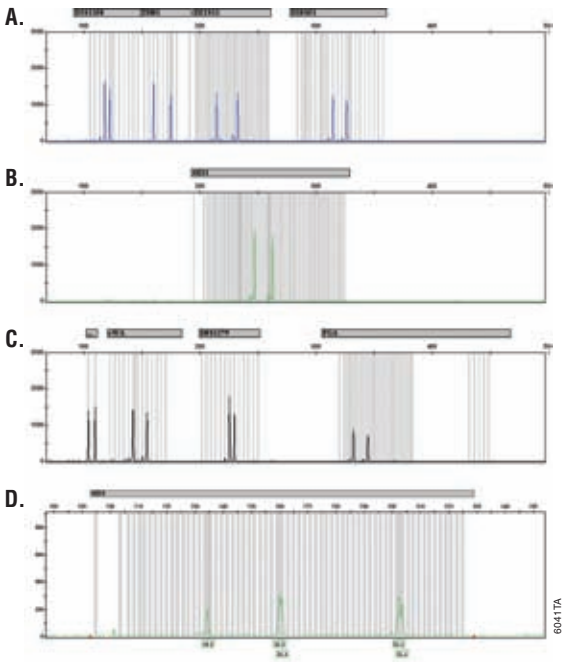
The PowerPlex® ES System is compatible with the ABI PRISM® 310, 3100 and 3100-*Avant* Genetic Analyzers, Applied Biosystems 3130 and 3130*xl* Genetic Analyzers and ABI PRISM® 377 DNA Sequencer.

Features:

- **Single Amplification:** Amplify 7 ENFSI loci, SE33 (ACTBP2) and Amelogenin in one reaction.
- **Sensitivity:** The system is optimized for use with 0.5–1ng template DNA. Additional studies show that interpretable and reproducible results can be obtained with <0.5ng template DNA.
- **Internal Lane Standard 600:** This marker offers the greatest precision available for DNA typing. The CXR-labeled marker consists of 22 bands of 60–600bp. Fragments that are multiples of 100 bases have fluorescence intensities approximately twice that of other fragments to simplify size assignment.
- **Allelic Ladders:** Allelic ladders for all nine loci, including the most frequently occurring microvariants, are provided to simplify interpretation.
- **Separation Control:** This component contains five SE33 alleles used to document the resolution of closely migrating fragments.
- **Automatic Assignment of Genotypes:** Panel and bin files are required for use with GeneMapper® *ID* and are available for download at: **www.promega.com/geneticidtools/**. The PowerTyper™ ES Macro facilitates data analysis, allowing automatic assignment of genotypes using the Genotyper® software. The PowerTyper™ Macros can be downloaded from: **www.promega.com/geneticidtools/** or ordered on CD-ROM.

Protocol	Part#
PowerPlex® ES System Technical Manual	TMD017
PowerPlex® Matrix Standards, 310, Technical Bulletin	TBD021
PowerPlex® Matrix Standards, 3100/3130, Technical Bulletin	TBD022

Storage Conditions: Store at –20°C. The fluorescent primer pairs and allelic ladders are light-sensitive; therefore, minimize light exposure.



The PowerPlex® ES System. A single DNA template (1ng) was amplified using the PowerPlex® ES 10X Primer Pair Mix. Amplification products were detected using an ABI PRISM® 310 Genetic Analyzer and a 2-second injection. Results were analyzed using GeneMapper® *ID* software. **Panel A.** An electropherogram showing the fluorescein-labeled loci D3S1358, TH01, D21S11 and D18S51. **Panel B.** An electropherogram showing the JOE-labeled locus SE33. **Panel C.** An electropherogram showing the TMR-labeled loci Amelogenin, vWA, D8S1179 and FGA. **Panel D.** An electropherogram showing the PowerPlex® ES Separation Control.

PowerPlex® 16 and ES Monoplex Systems

Product	Size	Cat.#
PowerPlex® 16 Monoplex System, Penta E (Fluorescein)	100 reactions	DC6591
PowerPlex® 16 Monoplex System, Penta D (JOE)	100 reactions	DC6651
PowerPlex® ES Monoplex System, SE33 (JOE)	100 reactions	DC6751
PowerPlex® 16 Monoplex System D3S1358 (Fluorescein)	100 reactions	DC6551
PowerPlex® 16 Monoplex System TH01 (Fluorescein)	100 reactions	DC6561
PowerPlex® 16 Monoplex System D21S11 (Fluorescein)	100 reactions	DC6571
PowerPlex® 16 Monoplex System D18S51 (Fluorescein)	100 reactions	DC6581
PowerPlex® 16 Monoplex System D5S818 (JOE)	100 reactions	DC6601
PowerPlex® 16 Monoplex System D13S317 (JOE)	100 reactions	DC6611
PowerPlex® 16 Monoplex System D7S820 (JOE)	100 reactions	DC6621
PowerPlex® 16 Monoplex System D16S539 (JOE)	100 reactions	DC6631
PowerPlex® 16 Monoplex System CSF1PO (JOE)	100 reactions	DC6641
PowerPlex® 16 Monoplex System vWA (TMR)	100 reactions	DC6661
PowerPlex® 16 Monoplex System D8S1179 (TMR)	100 reactions	DC6671
PowerPlex® 16 Monoplex System TPOX (TMR)	100 reactions	DC6681
PowerPlex® 16 Monoplex System FGA (TMR)	100 reactions	DC6691

Not For Medical Diagnostic Use.

Description: The PowerPlex® 16 and ES Monoplex Systems are available to amplify the Penta E, Penta D, SE33, D3S1358, TH01, D21S11, D18S51, D5S818, D13S317, D7S820, D16S539, CSF1PO, vWR, D8S1179, TPOX or FGA locus. Each monoplex system allows amplification of a single locus to confirm results obtained with the PowerPlex® 16 or PowerPlex® ES System. The monoplex systems also can be used to re-amplify DNA samples when one or more of the loci do not amplify initially due to suboptimal amplification conditions or poor DNA quality.

The PowerPlex® 16 and PowerPlex® ES Monoplex Systems contain primer pairs that have the same sequence as those used in the PowerPlex® 16 HS (Cat.# DC2100, DC2101), PowerPlex® 16 (Cat.# DC6530, DC6531), PowerPlex® 2.1 (Cat.# DC6470, DC6471), PowerPlex® 16 BIO (Cat.# DC6540, DC6541) and PowerPlex® ES Systems (Cat.# DC6730, DC6731).

Allelic ladders are only provided in the following PowerPlex® Monoplex Systems: DC6751, DC6591 and DC6651 [SE33 (JOE); Penta E (fluorescein) and Penta D (JOE), respectively].

Allelic ladders that are not provided are available by custom order. Please contact Technical Services for allelic ladder options based on the platform used.

The PowerPlex® 16 and ES Monoplex Systems were developed for use with ABI PRISM® 310, 3100 and 3100-*Avant* Genetic Analyzers and Applied Biosystems 3130 and 3130*xl* Genetic Analyzers and are compatible with the Hitachi FMBIO® II Fluorescence Imaging System.

Protocol	Part#
PowerPlex® 16 and PowerPlex® ES Monoplex Systems Technical Bulletin	TBD017
PowerPlex® 16 System Technical Manual	TMD012
PowerPlex® ES System Technical Manual	TMD017

Storage Conditions: Store at –20°C. The fluorescent primer pair is light-sensitive; therefore, minimize light exposure.

PowerPlex® 1.1 System

Product	Size	Cat.#
PowerPlex® 1.1 System	100 reactions	DC6091
	400 reactions	DC6090
Available Separately	**Size**	**Cat.#**
PowerPlex® 1.1 and 2.1 Systems	100 reactions	DC6501
	400 reactions	DC6500
K562 DNA High Molecular Weight	30 µg	DD2011
D16S539 Add-In for PowerPlex® 1.1	250 µl	DK3131

Cat. DC6091, DC6090, DC6501, DC6500, DK3131 Not for Medical Diagnostic Use.

Description: The PowerPlex® 1.1 System allows co-amplification and two-color detection of eight STR loci. This system contains D16S539, D7S820, D13S317 and D5S818 primers labeled with fluorescein and CSF1PO, TPOX, TH01 and vWA primers labeled with carboxy-tetramethylrhodamine (TMR). All eight loci are amplified simultaneously in a single tube and analyzed in a single gel lane. Loci amplified using the PowerPlex® 1.1 System also may be co-amplified with the fluorescent *GenePrint*® Sex Identification System—Amelogenin (TMR) (Cat.# DC6171), allowing detection of nine loci in a single gel lane. The PowerPlex® 1.1 System is customized for use with Hitachi FMBIO® and FMBIO® II Fluorescence Imaging Systems.

The D16S539 Add-In for PowerPlex® 1.1 is a primer pair provided to alleviate an allele dropout in the D16S539 locus of the PowerPlex® 1.1 System when amplified with AmpliTaq Gold® and Gold ST★R 10X Buffer.

Features:

- **High-Throughput Analysis:** Analysis is achieved by comparing amplified DNA fragments directly with the allelic ladder provided for each locus.
- **Size Marker:** The Fluorescent Ladder (CXR), 60–400 Bases, is provided with the system and is tagged with a third fluorescent dye. This marker may be included as a standard in each gel lane and visualized separately to monitor fluctuations in sample mobility.
- **Allelic Ladders:** Allelic ladders for all loci are provided to simplify interpretation.
- **Flexibility:** The primer sets are compatible with the use of AmpliTaq Gold® [when used with Gold ST★R 10X Buffer (Cat.# DM2411), sold separately] or other sources of *Taq* DNA polymerase, generating few or no amplification artifacts with either enzyme.

Protocol	Part#
PowerPlex® 1.1 System Technical Manual	TMD008
D16S539 Add-In for PowerPlex® 1.1 Promega Product Information	9CADK313

Storage Conditions: Store at –20°C. The fluorescent primer pairs and allelic ladders are light-sensitive; therefore, minimize light exposure.

PowerPlex® 2.1 System

Product	Size	Cat.#
PowerPlex® 2.1 System	100 reactions	DC6471
	400 reactions	DC6470
Available Separately	**Size**	**Cat.#**
PowerPlex® 1.1 and 2.1 Systems	100 reactions	DC6501
	400 reactions	DC6500
K562 DNA High Molecular Weight	30 µg	DD2011

Cat.# DC6471, DC6470, DC6501, DC6500 Not for Medical Diagnostic Use.

Description: The PowerPlex® 2.1 System allows co-amplification and two-color detection of nine STR loci. The PowerPlex® 2.1 System amplifies the loci Penta E, D18S51, D21S11, TH01, D3S1358, FGA, TPOX, D8S1179 and vWA. One primer specific for each of the Penta E, D18S51, D21S11, TH01 and D3S1358 loci is labeled with fluorescein (FL), and one primer specific for each of the FGA, TPOX, D8S1179 and vWA loci is labeled with carboxy-tetramethyl-rhodamine (TMR). All nine loci are amplified simultaneously in a single tube and analyzed in a single gel lane.

The PowerPlex® 2.1 System is customized for use with Hitachi FMBIO® and FMBIO® II Fluorescence Imaging Systems.

Features:

- **Efficiency:** An allelic ladder mixture consisting of most known alleles in the population is included to allow rapid and precise assignment of alleles.
- **High-Throughput Analysis:** Analysis is achieved by comparing amplified DNA fragments directly with the allelic ladder provided for each locus.
- **Internal Lane Standard 600:** This marker (Cat.# DG2611) offers the greatest precision available for DNA typing. It is used in each gel lane to increase the precision of analysis. The CXR-labeled marker consists of 22 bands of 60–600bp. Fragments that are multiples of 100 bases have fluorescence intensities approximately twice that of other fragments to simplify size assignment.
- **Allelic Ladders:** Allelic ladders for all loci are provided to simplify interpretation.
- **Penta E:** The low level of stutter associated with Penta E simplifies the interpretation of results from samples containing more than one source of human DNA. In addition, it is a highly polymorphic locus and has no known microvariant alleles. These combined properties make the Penta E locus an ideal marker for forensic analysis.

Protocol	Part#
PowerPlex® 2.1 System Technical Manual	TMD011

Storage Conditions: Store at –20°C. The fluorescent primer pairs and allelic ladders are light-sensitive; therefore, minimize light exposure.

PowerPlex® 1.2 System

Product	Size	Cat.#
PowerPlex® 1.2 System	100 reactions	DC6101
Available Separately	**Size**	**Cat.#**
K562 DNA High Molecular Weight	30 µg	DD2011
PowerPlex® Matrix Standards, 310/377	50 µl (each dye)	DG3640
PowerTyper™ Macros (Release 2.0)	1 CD-ROM	DG3470

Cat.# DC6101, DG3640, DG3470 Not for Medical Diagnostic Use.

Description: The PowerPlex® 1.2 System allows co-amplification and two-color detection of nine loci (eight STR loci and Amelogenin). The PowerPlex® 1.2 System contains D16S539, D7S820, D13S317 and D5S818 primers labeled with fluorescein and CSF1PO, TPOX, TH01, vWA and Amelogenin primers labeled with carboxy-tetramethylrhodamine (TMR). All nine loci are amplified simultaneously in a single tube and analyzed in a single gel lane or capillary.

The PowerPlex® 1.2 System is designed for use with the ABI PRISM® 310 and 3100 Genetic Analyzers and ABI PRISM® 377 DNA Sequencer. When using the ABI PRISM® 3100 Genetic Analyzer, use the PowerPlex® Matrix Standards, 3100—Custom (Cat.# X3121) for spectral calibration.

Features:

- **High-Throughput Analysis:** Analysis is achieved by comparing amplified DNA fragments directly with the allelic ladder provided for each locus.
- **Size Marker:** Fluorescent Ladder (CXR), 60–400 Bases, is provided with the system and is tagged with a third fluorescent dye. This marker is included as an internal lane standard in each lane to increase precision of analysis.
- **Allelic Ladders:** Allelic ladders for all loci are provided to simplify interpretation through the use of the PowerTyper™ 1.2 Macro.
- **Efficiency:** The PowerPlex® 1.2 System allows simultaneous amplification and analysis of eight STR loci plus Amelogenin for gender identification. The Amelogenin primers allow amplification of sequences specific to the X (212-base) and Y (218-base) chromosomes.
- **Automatic Assignment of Genotypes:** Panel and bin files are required for use with GeneMapper® *ID* software and are available for download at: **www.promega.com/geneticidtools/**. The PowerTyper™ 1.2 Macro facilitates data analysis, allowing automatic assignment of genotypes using the Genotyper® software. The PowerTyper™ Macros can be downloaded from: **www.promega.com/geneticidtools/** or ordered on CD-ROM.

Protocol	Part#
PowerPlex® 1.2 System Technical Manual	TMD009
PowerPlex® Matrix Standards, 310/377, Technical Bulletin	TBD018

Storage Conditions: Store at –20°C. The fluorescent primer pairs and allelic ladders are light-sensitive; therefore, minimize light exposure.

PowerTyper™ Macros (Release 2.0)

Product	Size	Cat.#
PowerTyper™ Macros (Release 2.0)	1 CD-ROM	DG3470

Not For Medical Diagnostic Use.

Description: The PowerTyper™ Macros facilitate analysis of data generated using the PowerPlex® 16, 1.2, ES and Y Systems, PowerPlex® 16 Monoplex Systems, Penta E and Penta D, and PowerPlex® ES Monoplex System, SE33. The macros allow automatic assignment of genotypes using the Genotyper® software. After samples are amplified using the PowerPlex® Systems and detected using the ABI PRISM® 310, 3100 or 3100-*Avant* Genetic Analyzer, Applied Biosystems 3130 or 3130*xl* Genetic Analyzer or ABI PRISM® 377 DNA Sequencer, GeneScan® sample files are imported into the Genotyper® program and analyzed using the appropriate PowerTyper™ Macro. Compatibility information and download capabilities for the PowerTyper™ Macros can be found at: **www.promega.com/geneticidtools/**; the macros also can be ordered on CD-ROM.

Panel and bin files for use with GeneMapper® *ID* are available for download at: **www.promega.com/geneticidtools/**

Features:

- **Convenient:** The PowerTyper™ Macros allow automatic assignment of genotypes using the Genotyper® software.

Protocol	Part#
PowerPlex® 16 System Technical Manual	TMD012
PowerPlex® Y System Technical Manual	TMD018
PowerPlex® ES System Technical Manual	TMD017
PowerPlex® 1.2 System Technical Manual	TMD009

Gold ST★R 10X Buffer

Product	Size	Cat.#
Gold ST★R 10X Buffer	1.2 ml	DM2411

For Laboratory Use.

Description: Gold ST★R 10X Buffer can be used to amplify STR loci using AmpliTaq Gold® DNA polymerase. Gold ST★R Buffer can be substituted for the STR 10X Buffer that is supplied with PowerPlex® System and *GenePrint*® STR Systems, allowing the use of either AmpliTaq® or AmpliTaq Gold® DNA polymerase. This buffer includes BSA for a more robust reaction and improved results under nonoptimal conditions. The combination of Gold ST★R 10X Buffer and AmpliTaq Gold® DNA polymerase can result in greater sensitivity and reduced amplification artifacts.

Storage Conditions: Store at –20°C.

New Internal Lane Standard 600

Product	Size	Cat.#
Internal Lane Standard 600	150 µl	DG1071
For Laboratory Use.		

Description: The Internal Lane Standard 600 (ILS 600) consists of 22 bands ranging in size from 60bp to 600bp. Fragments of 60–200bp are spaced at 20bp intervals, fragments of 200–500bp are spaced every 25 bases, and fragments of 500–600bp are spaced every 50 bases. Fragments that are multiples of 100 bases have fluorescence intensities approximately twice that of other fragments to simplify size assignment. The DNA ladder is double-stranded and asymmetrically labeled with carboxy-X-rhodamine (CXR). The Internal Lane Standard 600 is used to assign sizes to DNA fragments separated by electrophoresis and detected using a variety of fluorescence-detection instruments (e.g., Hitachi FMBIO® Fluorescence Imaging System and ABI PRISM® 310, 3100, 3100-*Avant* and Applied Biosystems 3130, 3130*xl* and 3500 Genetic Analyzers). ILS 600 is commonly used as an internal size marker for other applications and can be visualized by detecting fluorescent emission at 597nm after excitation at 576nm.

In addition, the Internal Lane Standard 600 contains additives that prevent the formation of two artifacts ("split peak" and "n–10") at the vWA locus in the PowerPlex® 16 and PowerPlex® ES Systems when using ABI PRISM® 3100, 3100-*Avant* and Applied Biosystems 3130, 3130*xl*, 3500 and 3500*xl* Genetic Analyzers.

Storage Conditions: Store at –20°C. Avoid multiple freeze-thaw cycles or exposure to frequent temperature changes. These fluctuations can greatly alter product stability. The Internal Lane Standard 600 is light-sensitive; therefore, minimize light exposure.

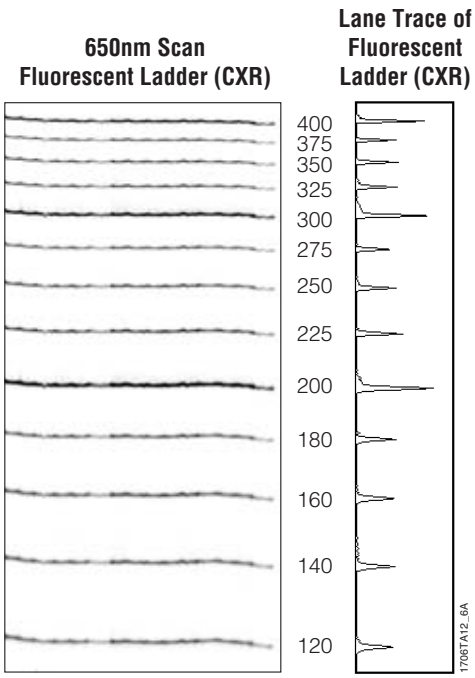
Fluorescent Ladder (CXR), 60–400 Bases

Product	Size	Cat.#
Fluorescent Ladder (CXR), 60–400 Bases	65 µl	DG6221

Description: The Fluorescent Ladder (CXR), 60–400 Bases, consists of 16 DNA fragments with sizes of 60, 80, 100, 120, 140, 160, 180, 200, 225, 250, 275, 300, 325, 350, 375 and 400 bases. Each fragment is 5´-end labeled with the fluorescent dye carboxy-X-rhodamine (CXR). The ladder can be used as a single-stranded DNA size marker in denaturing gels. The 100, 200, 300 and 400bp fragments have fluorescence intensities approximately twice that of the other bands and serve as visible reference indicators.

The Fluorescent Ladder (CXR) is intended to be used in assigning sizes to DNA fragments separated by electrophoresis and detected using a variety of fluorescence-detection instruments (e.g., Hitachi FMBIO® Fluorescence Imaging System, ABI PRISM® 377 DNA Sequencer and ABI PRISM® 310, 3100, 3100-*Avant* and Applied Biosystems 3130 and 3130*xl* Genetic Analyzers).

Storage Conditions: Store at –20°C. Avoid multiple freeze-thaw cycles or exposure to frequent temperature changes. These fluctuations can greatly alter product stability. The Fluorescent Ladder is light-sensitive; therefore, minimize light exposure.



The Fluorescent Ladder (CXR), 60–400 Bases. Following separation, the Fluorescent Ladder (CXR) was detected using a 650nm filter with the Hitachi FMBIO® II Fluorescence Imaging System. Fragment sizes are shown in the middle, and a lane trace is provided. The 100, 200, 300 and 400bp fragments have fluorescence intensities approximately twice that of the others.

GenePrint® Fluorescent STR Systems

Product	Size	Cat.#
GammaSTR® Multiplex (Fluorescein) D16S539, D7S820, D13S317, D5S818	100 reactions	DC6071
	400 reactions	DC6070
CSF1PO, TPOX, TH01, vWA Multiplex (Fluorescein)	100 reactions	DC6301
	400 reactions	DC6300
F13A01, FESFPS, F13B, LPL Multiplex (Fluorescein)	100 reactions	DC6311
	400 reactions	DC6310
Available Separately	**Size**	**Cat.#**
CTTv Allelic Ladder Mix (Fluorescein)	150 µl	DG2121
FFFL Allelic Ladder Mix (Fluorescein)	150 µl	DG2131
GammaSTR® Allelic Ladder Mix (Fluorescein)	150 µl	DG3291

Cat.# DC6071, DC6070, DC6301, DC6300, DC6311, DC6310 Not for Medical Diagnostic Use.

Description: The *GenePrint*® Fluorescent STR Systems were developed for use with the Hitachi FMBIO® Fluorescence Imaging Systems and ABI PRISM® 377 DNA Sequencer. One primer for each locus is labeled with fluorescein to allow fluorescent detection. Fluorescein has an excitation maximum at 488nm and an emission maximum at 532nm. Therefore, the systems are compatible with a variety of fluorescence-detection instruments, including the ABI PRISM® 310, 3100 and 3100-*Avant* Genetic Analyzers and Applied Biosystems 3130 and 3130*xl* Genetic Analyzers. When using the ABI PRISM® 3100 Genetic Analyzer, use the PowerPlex® Matrix Standards, 3100—Custom (Cat.# X3121) for spectral calibration.

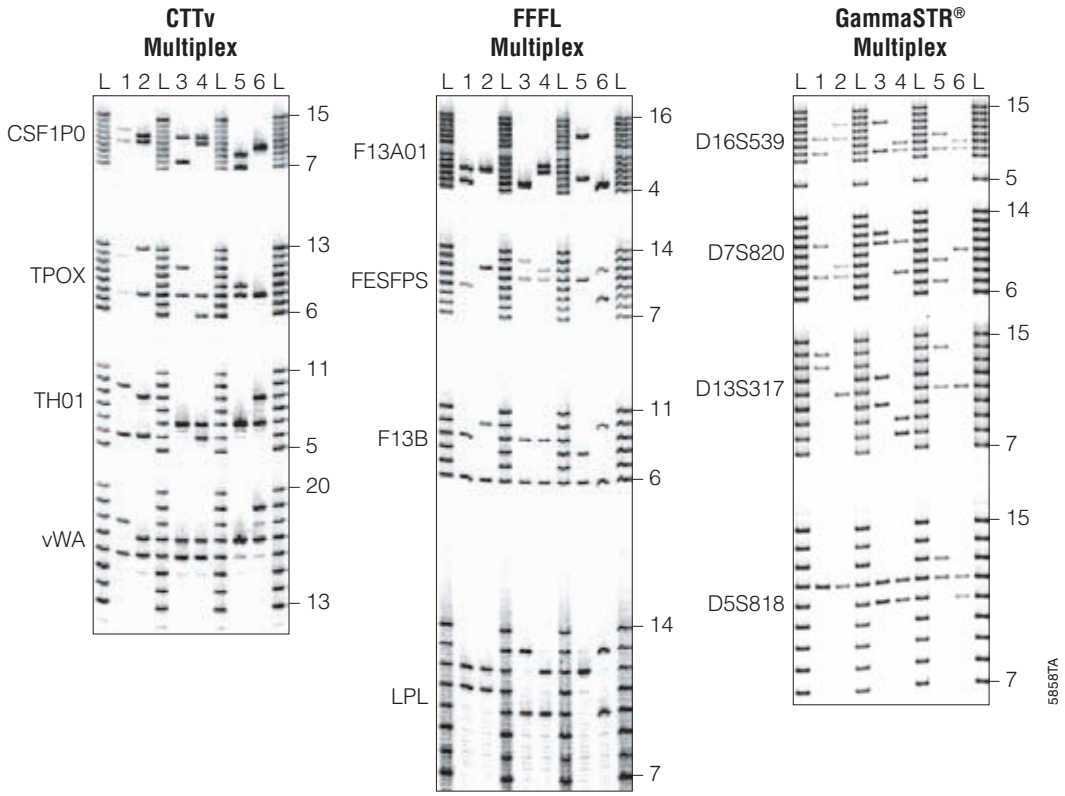
Each system provides all materials required to amplify STR regions of purified genomic DNA except *Taq* DNA polymerase. Amplification of DNA using the system components plus *Taq* DNA polymerase produces fluorescein-labeled fragments representing alleles from the template DNA. For instruments that support two-color fluorescence detection, additional precision may be achieved by including the Fluorescent Ladder (CXR), 60–400 Bases (Cat.# DG6221), or the Internal Lane Standard 600 (Cat.# DG2611) in each gel lane.

Features:

- **High-Throughput Analysis:** Analysis is achieved by comparing amplified DNA fragments directly with the allelic ladder provided for each locus.
- **Efficiency:** The fluorescent STR multiplex systems support simultaneous single-tube amplification of four polymorphic STR loci with nonoverlapping allele size ranges.
- **Allelic Ladders:** Comparing amplified alleles with allelic ladders provided with each system allows rapid and reliable allele assignment.

Protocol	Part#
Technical Manual	TMD006

Storage Conditions: Store at –20°C. The fluorescent primer pairs and allelic ladders are light-sensitive; therefore, minimize light exposure.



***GenePrint*® CTTv, FFFL and GammaSTR® Multiplex Systems**. STR analyses were performed using the *GenePrint*® CTTv, FFFL and GammaSTR® Fluorescent STR Systems. For each system, six DNA samples were amplified (lanes 1–6) and are shown with allelic ladders for the corresponding loci (lanes L). Numbers to the right of each image indicate the smallest and largest number of repeat units present in corresponding fragments of each allelic ladder. All materials were separated on 4% denaturing polyacrylamide gels. Amplification products were detected using the Hitachi FMBIO® II Fluorescence Imaging System.

GenePrint® STR Systems (Silver Stain Detection)

Product	Size	Cat.#
GenePrint® SilverSTR® III System (D7S820, D13S317, D16S539)	100 reactions	DC6451
	400 reactions	DC6450
CSF1PO, TPOX, TH01 Multiplex	100 reactions	DC6001
	400 reactions	DC6000
F13A01, FESFPS, vWA Multiplex	100 reactions	DC6031
	400 reactions	DC6030
CSF1PO	100 reactions	DC4011
F13A01	100 reactions	DC4041
F13B	100 reactions	DC4001
FESFPS	100 reactions	DC4021
HPRTB	100 reactions	DC4061
LPL	100 reactions	DC4071
TH01	100 reactions	DC1191
TPOX	100 reactions	DC4051
vWA	100 reactions	DC4031
Available Separately	**Size**	**Cat.#**
CTT Allelic Ladder Mix	150 µl	DG2101
FFv Allelic Ladder Mix	150 µl	DG2141

Cat.# DC6451, DC6450, DC6001, DC6000, DC6031, DC6030, DC4011, DC4041, DC4001, DC4021, DC4061, DC4071, DC1191, DC4051, DC4031 Not For Medical Diagnostic Use.

Description: The *GenePrint*® Silver STR Systems provide a rapid, non-radioactive method to evaluate small amounts (e.g., 1ng) of human DNA. The systems provide all of the materials required to amplify STR regions of purified genomic DNA except for *Taq* DNA polymerase and sample DNA. The amplified STR fragments then are separated by polyacrylamide gel electrophoresis and detected by silver staining.

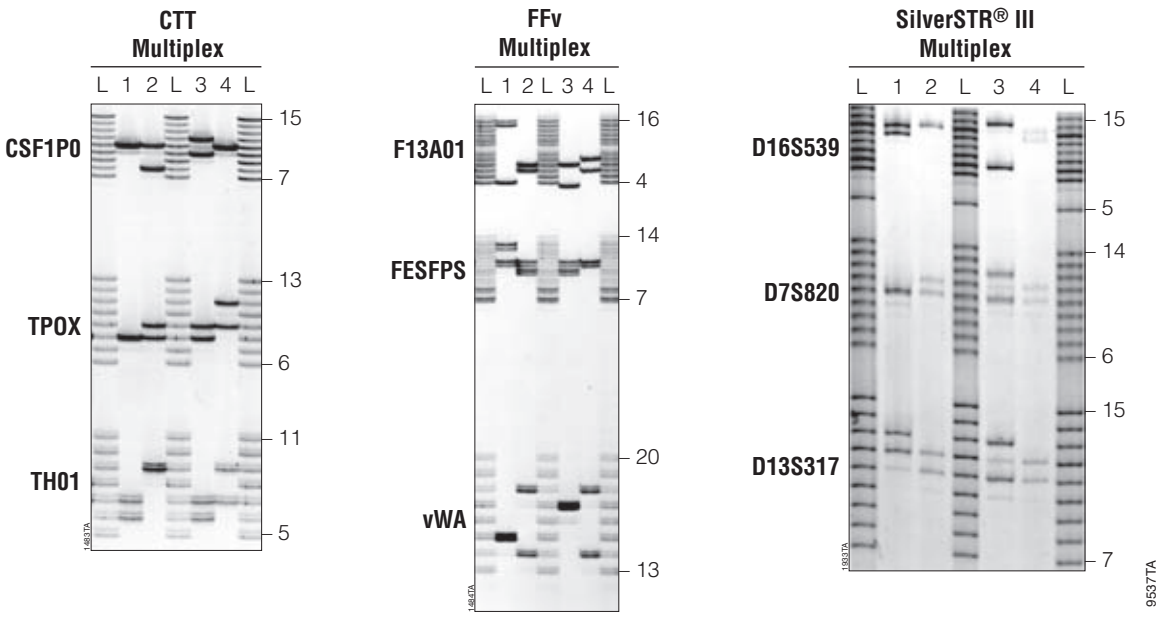
The combination of SilverSTR® III, CTT and FFv provides access to seven of the thirteen core loci that comprise the Combined DNA Index System (CODIS) database.

Features:

- **Economical:** The *GenePrint*® Silver STR Systems do not require fluorescence-detection equipment for analysis. The systems are ideal for labs that are starting STR analysis or do not wish to purchase expensive fluorescence-detection equipment.
- **Efficient:** Analysis requires less than one day. Each multiplex allows simultaneous amplification of three nonoverlapping STR loci for high discrimination power.

Protocol	Part#
Technical Manual	TMD004

Storage Conditions: Store at –20°C.



***GenePrint*® Silver STR Systems**. Individual genomic DNA samples (lanes 1–4) were amplified using the *GenePrint*® Silver STR Systems. Amplification products from the CTT and FFv Multiplexes were separated on a 4% polyacrylamide denaturing gel. Amplification products from the SilverSTR® III Multiplex (D16S539, D7S820, D13S317) were separated on a 6% polyacrylamide denaturing gel. Lanes labeled L contain allelic ladders for the respective loci. Numbers to the right of each image indicate the smallest and largest number of repeat units present in corresponding fragments of each allelic ladder.

GenePrint® Sex Identification Systems

Product	Size	Cat.#
Amelogenin (Silver Detection)	100 reactions	DC4081
Amelogenin (Fluorescein Detection)	100 reactions	DC5171
Amelogenin (TMR)	100 reactions	DC6171

Not For Medical Diagnostic Use.

Description: The Amelogenin Systems can be used for sex determination. When used under reaction conditions recommended in the Technical Manuals (#TMD004, #TMD006 and #TMD008), a specific segment of the human X chromosome generates a 212bp product, while the corresponding human Y-chromosomal DNA segment produces a 218bp fragment. The Amelogenin locus may be co-amplified and co-analyzed with a compatible multiplex system by mixing the Amelogenin primers with those of the appropriate multiplex system prior to use.

Protocol	Part#
GenePrint® STR Systems Technical Manual	TMD004
GenePrint® Fluorescent STR Systems Technical Manual	TMD006
PowerPlex® 1.1 System Technical Manual	TMD008

Storage Conditions: Store at –20°C. The fluorescent primer pair and ladder are light-sensitive; therefore, minimize light exposure.

Selection Guide to Match Amelogenin Choice with Appropriate Multiplex System.

Amelogenin Label	Cat.#	Multiplex System	Cat.#
Silver Detection	DC4081	CTT	DC6001, DC6000
Fluorescein Detection	DC5171	CTTv	DC6301, DC6300
TMR Detection	DC6171	PowerPlex® 1.1	DC6091, DC6090

9489LA

Biochemical Reagents

Product	Size	Cat.#
SILVER SEQUENCE™ Staining Reagents	10 gels	Q4132
STR 10X Buffer	1.2 ml	DM2211
Gold ST★R 10X Buffer	1.2 ml	DM2411
Agarose	1 kg	DV3123
STR 2X Loading Solution	3 ml	DV4331
Blue Dextran Loading Solution	3 × 1 ml	DV4351
Gel Tracking Dye	4 × 250 µl	DV4361
Bromophenol Blue Loading Solution	3 × 1 ml	DV4371
Mineral Oil	12 ml	DY1151

Cat.# DV4351, DV4361, DV4371 For Laboratory Use.

Description: Promega offers supporting reagents for separation, hybridization and detection of specific loci in the human genome. These quality-tested reagents are optimized for use with Promega genetic identity systems.

Storage Conditions: Store Cat.# DM2211, DM2411, DV4331, DV4351, DV4361 and DV4371 at –20°C. Store Cat.# Q4132, DV3123 and DY1151 at 22–25°C.

ART® Aerosol-Resistant Tips

Product	Size	Cat.#
ART® 10 Ultramicro Pipet Tip, 0.5–10µl	960 /pk	DY1051
ART® 20E Ultramicro Pipet Tip, 0.5–10µl	960 /pk	DY1061
ART® 20P Pipet Tip, 20µl	960 /pk	DY1071
ART® GEL Gel Loading Pipet Tip, 100µl	960 /pk	DY1081
ART® 100 Pipet Tip, 100µl	960 /pk	DY1101
ART® 100E Pipet Tip, 100µl	960 /pk	DY1111
ART® 200 Pipet Tip, 200µl	960 /pk	DY1121
ART® 1000E Pipet Tip, 1,000µl	800 /pk	DY1131

Description: ART® Aerosol-Resistant Tips offer a safe alternative to using positive displacement pipettors. ART® Tips are designed for applications such as PCR, pipetting radioactive materials and genetic studies. In addition, ART® Tips are excellent for tissue culture, gel loading, serological assays and forensic studies. ART® Tips prevent aerosol contaminants and liquids from reaching the pipette barrel due to a nonwicking self-sealing barrier. All ART® Tips are presterilized, DNase- and RNase-free, racked and ready-to-use.

Storage Conditions: Store at room temperature.

Genetic Reporters and Transfection Systems

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Section Contents

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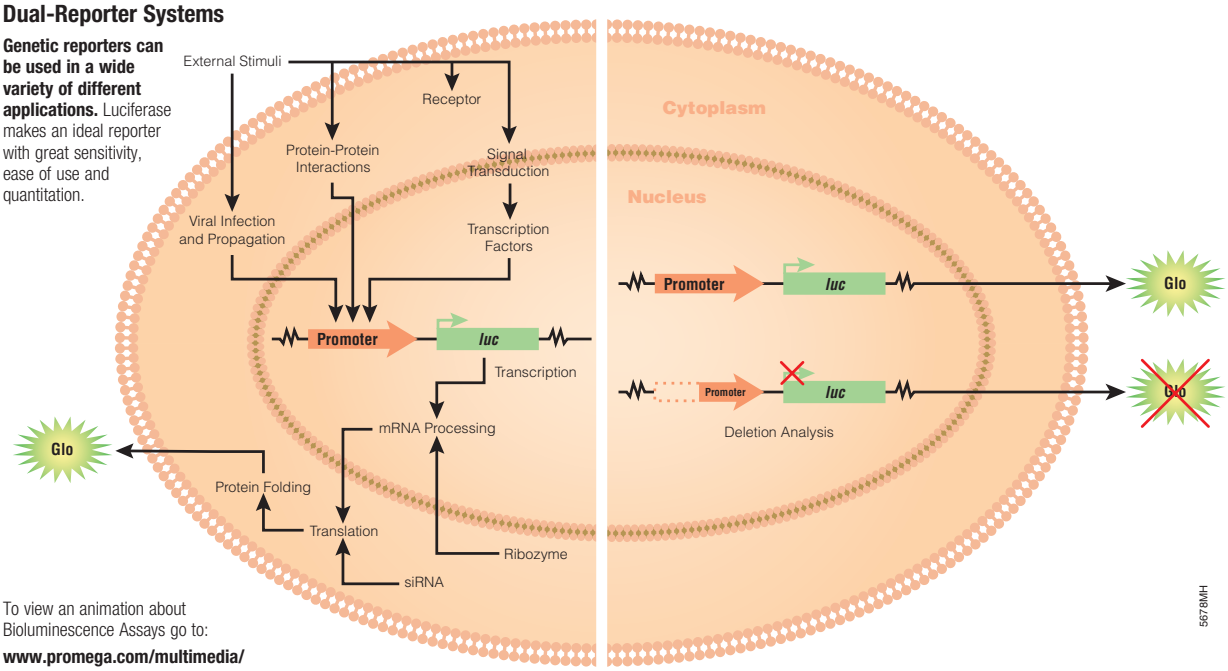
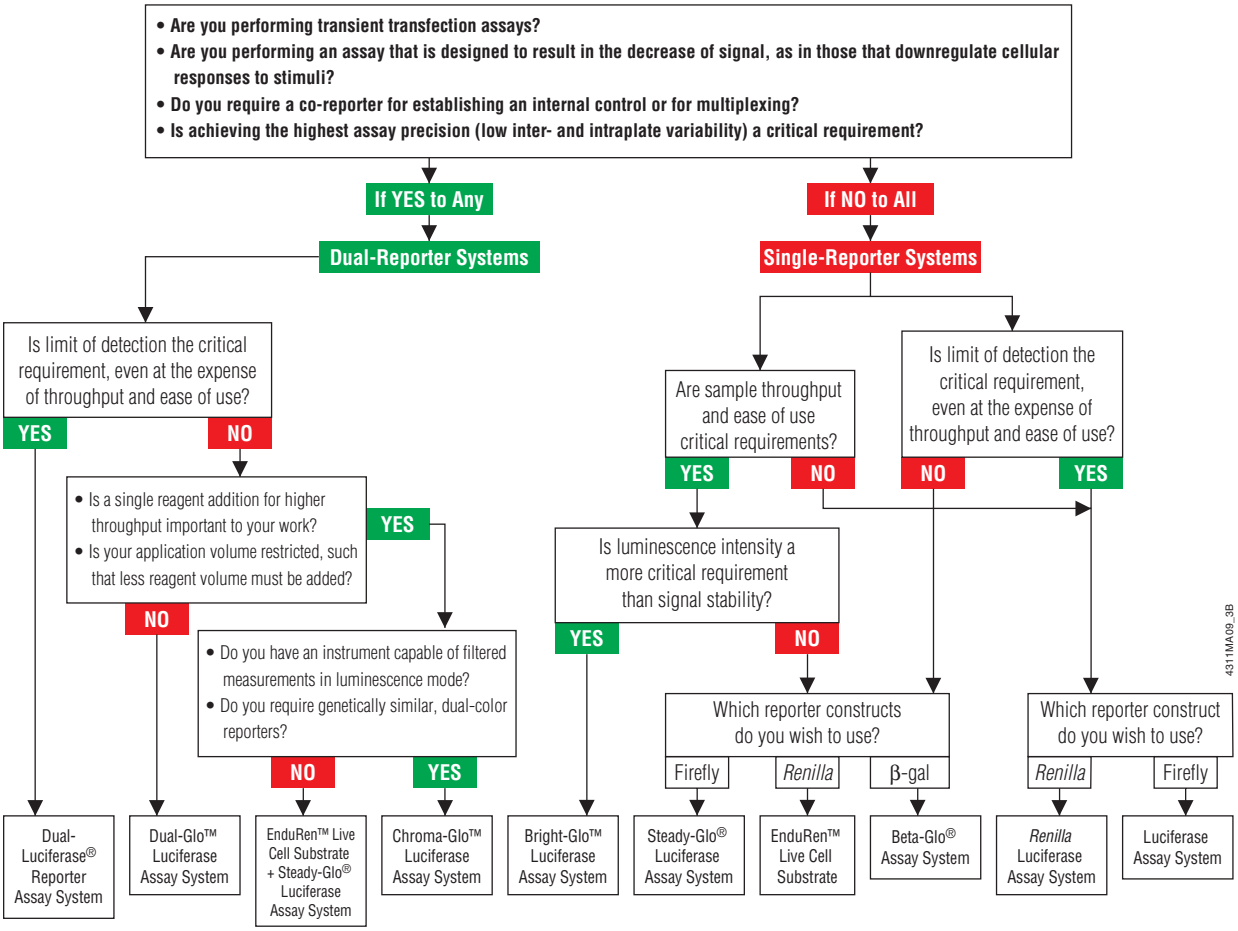
Dual-Reporter Systems

Genetic reporters can be used in a wide variety of different applications. Luciferase makes an ideal reporter with great sensitivity, ease of use and quantitation.



To view an animation about Bioluminescence Assays go to:
www.promega.com/multimedia/

Genetic Reporters Selection Guide



This chart and the chart on page 141 provide a general guide for selecting from Promega luminescence reporter assays and firefly and *Renilla* luciferase genes. These charts contain general questions about your application needs and preferences and are designed to recommend a product based on your answer(s). Selection of a particular reporter assay or gene may require additional considerations and preferences not addressed by these charts. In these instances, please contact Technical Services for assistance.

Dual-Glo® Luciferase Assay System

Product	Size	Cat.#
Dual-Glo® Luciferase Assay System	10 ml	E2920
	100 ml	E2940
	10 × 100 ml	E2980

Please contact Promega for information on bulk purchases.

Description: The Dual-Glo® Luciferase Assay System is a homogeneous reagent system that enables fast and simple quantitation of a stable luminescent signal from two reporter genes in a single sample. This convenient "add-and-read" system generates both firefly and *Renilla* luciferase luminescence signals from cells that have not been preconditioned or prelysed. The Dual-Glo® Luciferase Assay System provides high Z´-factors for cell-based, high-throughput screening applications. With the Dual-Glo® System, internal controls can be established to minimize sample variability by reducing false positive and false negative readings caused by nonspecific factors such as cytotoxicity. In the Dual-Glo® Luciferase Assay, the activity of the primary reporter is correlated with the effect of specific stimuli, and the activity of the co-transfected control reporter provides an internal control to normalize results. The system is optimized for batch processing both 96- and 384-well plates and is compatible with a wide variety of mammalian cell culture media.

Features:

- **Increased Precision and Accuracy:** Normalize primary reporter results with an internal control, a co-reporter that minimizes effects of cell number and health, transfection efficiency and nonspecific cellular responses.
- **Homogeneous Format:** Perform fewer steps. Assay cells directly in growth medium for both reporters. No centrifugation or lysis step is required.
- **Stable Signal:** Obtain flexibility for either batch or continuous processing of 96- and 384-well plates. Each luminescent signal may be measured for up to 2 hours after reagent addition.
- **Convenience:** Screen efficiently with this simple, two-step assay that is ideal for any luminometer. On-board injectors not required.
- **Wide Dynamic Range:** Analyze high and low reporter activity without sample dilution. Linear over at least 6 logs of enzyme concentration for each reporter.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Manual	TM058

Storage Conditions: Store Dual-Glo® Substrates at –20°C. Store Dual-Glo® Buffers below 25°C.

Dual-Luciferase® Reporter Assay System

Product	Size	Cat.#
Dual-Luciferase® Reporter Assay System	100 assays	E1910
Dual-Luciferase® Reporter Assay System 10-Pack	1,000 assays	E1960
Dual-Luciferase® Reporter 1000 Assay System	1,000 assays	E1980
Available Separately	**Size**	**Cat.#**
Passive Lysis 5X Buffer	30 ml	E1941

Description: The Dual-Luciferase® Reporter (DLR™) Assay System provides an efficient means of performing two reporter assays. In the DLR™ Assay, the activities of firefly (*Photinus pyralis*) and *Renilla* (*Renilla reniformis* or sea pansy) luciferases are measured sequentially from a single sample. The firefly luciferase reporter is measured first by adding Luciferase Assay Reagent II (LAR II) to generate a luminescent signal lasting at least one minute. After quantifying the firefly luminescence, this reaction is quenched, and the *Renilla* luciferase reaction is initiated simultaneously by adding Stop & Glo® Reagent to the same sample. Both assays can be completed in about 4 seconds using a luminometer with reagent auto-injectors. In the DLR™ Assay System, both reporters yield linear assays with attomole ($<10^{-18}$) sensitivities and no endogenous activity in the experimental host cells. Furthermore, the integrated format of the DLR™ Assay provides rapid quantitation of both reporters either in transfected cells or in cell-free transcription/translation reactions.

For best results with the Dual-Luciferase® Assay, Promega recommends using a luminometer that has been validated for use with the assay. These luminometers are qualified as DLR*eady*™. For a listing of qualified instruments, please visit the DLR*eady*™ Validated Luminometers page.

The pGL4 Luciferase Reporter Vectors are designed for use with the DLR™ Assay Systems. A *Renilla* luciferase vector with constitutive expression may be used in combination with any experimental firefly luciferase vector to co-transfect mammalian cells.

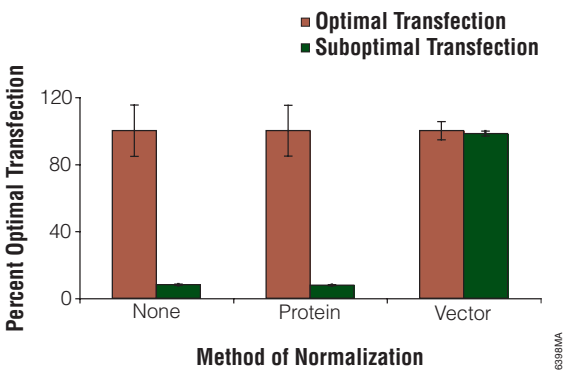
Notice for Cat.# E1960 and E1980: Sufficient Passive Lysis Buffer is provided to perform 1,000 assays with cells grown in 96-well plates (typically 20µl of 1X PLB per well). For applications requiring more lysis reagent (e.g., >100µl/well), additional Passive Lysis Buffer may be purchased separately.

Features:

- **Greater Accuracy:** *Renilla* luciferase internal control allows for more accurate results.
- **Convenient:** Samples don't have to be split, which saves plates and time.
- **Sensitive:** Allows study of weak promoters, low-level expression/regulation, and expression in cells that transfect poorly.
- **Linear:** Range extends 7 logs; very active samples typically do not need dilution.

Protocol	Part#
Dual-Luciferase® Reporter Assay System Technical Manual	TM040
Dual-Luciferase® Reporter 1000 Assay System Technical Manual	TM046

Storage Conditions: Store at –20°C.



Effect of transfection conditions on reporter results analyzed using different normalization methods. HEK293 cells were transfected with pGL4.13[*luc2*/SV40] Vector expressing firefly luciferase and pGL4.74[*hRluc*/TK] Vector expressing *Renilla* luciferase. Transfections were performed using both optimal and suboptimal lipid:DNA ratios (indicated as Optimal and Suboptimal Transfection conditions). Firefly and *Renilla* luciferase activities were measured using the Dual-Luciferase® Reporter Assay System (Cat.# E1960). Protein concentrations were determined using the Coomassie® Plus Bradford Reagent (Pierce). Firefly luciferase data were either not normalized (None), normalized to total protein (Protein), or normalized to *Renilla* luciferase activity (Vector). Data represent the average ± standard deviation of triplicate samples and are expressed as a percent of the optimal transfection for each normalization condition.

Chroma-Glo™ Luciferase Assay System

Product	Size	Cat.#
Chroma-Glo™ Luciferase Assay System	10 ml	E4910
	100 ml	E4920

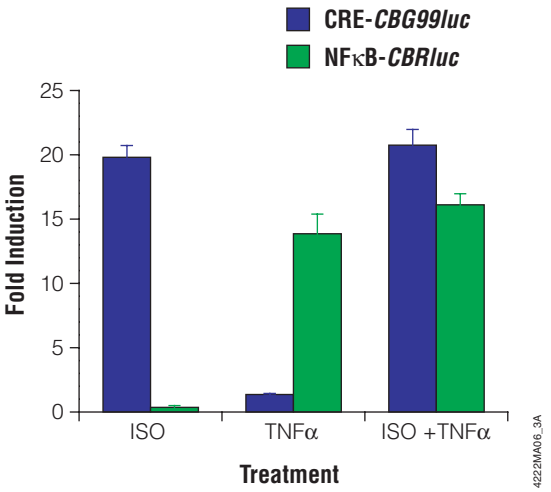
Description: The Chroma-Glo™ Luciferase Assay System and the Chroma-Luc™ Vectors provide a method to generate red and green (dual-color) luminescence from a single sample upon a single-reagent addition. Filtered measurement of the dual-color luminescence produced by the Chroma-Luc™ luciferases permits each reporter to be measured independently and virtually simultaneously. The Chroma-Glo™ Assay is in a homogeneous format that generates luminescence with >30-minute signal half-lives for each of the Chroma-Luc™ luciferases, thereby enabling the processing of many plates without prior sample handling. Use the high-homology Chroma-Luc™ luciferases to establish an ideal internal control for normalizing cytotoxicity in downregulation applications and for decreasing inter- and intrasample variability. You can also use the reporters to multiplex experimental reporters to increase the data content from cell-based assays.

Features:

- **Measure Dual Reporters Using a Single Substrate Addition:** Increase your accuracy and precision through normalization, or use both reporters to multiplex experimental measurements. Use filters to spectrally separate the luminescent signals.
- **Establish the Ideal Control or Multiplexed System:** Use the high-homology red and green luciferases to minimize potential RNA and protein effects on reporter expression.
- **Increase Your Throughput:** Use the stable luminescence for batch or continuous processing of multiple plates.
- **Perform Fewer Steps:** Add Chroma-Luc™ Reagent directly to cells in medium, then measure.
- **Automate This Assay:** Validated automated methods available at: **www.promega.com/automethods/**
- **Choose Your Configuration:** Learn more about our custom options for this product at:

Protocol	Part#
Technical Manual	TM062

Storage Conditions: Store the Chroma-Glo™ Substrate at –20°C. Store the Chroma-Glo™ Assay Buffer below 25°C.



Using the Chroma-Luc™ Technology to monitor two independent experimental signals from the same sample. DNA segments containing either CRE or the NFB consensus sequence were cloned into pCBG99-Basic (Cat.# E1431) or pCBR-Basic (Cat.# E1411). The resulting constructs, pCRE-CBG99-*luc* and pNFB-CBR*luc*, were cotransfected into 293 cells. At 24 hours post transfection, the cells received one of three treatments: ISO (1µM)/RO(100µM), TNFα (0.1µg/ml)/RO(100µM), or ISO(1µM)/RO(100µM) plus TNFα (0.1µg/ml). Only RO(100µM) was added to the Control wells. At six hours post treatment, cells were harvested and assayed with the Chroma-Glo™ Reagent. Relative light units were measured using the Mithras LB940 (Berthold Technologies) configured with a red filter (610 long pass) and a green filter (510/60). The red and green signals were deciphered by using the Chroma-Luc™ Calculator (available as a downloadable file at: **www.promega.com/chromacalc/**). Fold inductions were calculated by dividing the three treatments by the RO Control.

GloResponse™ Luciferase Reporter Cell Lines

Product	Size	Cat.#
GloResponse™ CRE-*luc2P* HEK293 Cell Line	2 vials	E8500
GloResponse™ NFAT-RE-*luc2P* HEK293 Cell Line	2 vials	E8510
GloResponse™ NF-B-RE-*luc2P* HEK293 Cell Line	2 vials	E8520
GloResponse™ 9X*GAL4*UAS-*luc2P* HEK293 Cell Line	2 vials	E8530

Each item contains two vials of approximately 2 × 10^6 cells in Freezing Media.

Description: The GloResponse™ Luciferase Reporter Cell Lines contain optimized, state-of-the-art luciferase reporter technology integrated into a cell line. This allows the rapid development of a reporter assay based on the pathway of interest regulating the luciferase gene. Assays configured using the GloResponse™ Cell Lines are amenable for high-throughput screening. These assays typically have greater response dynamics (fold of induction) than other assay formats and good quality as indicated by the high Z′ values. GloResponse™ Cell Lines were developed to study a variety of signaling pathways. Activators of these pathways may be native to the HEK293 cell line. Activity of non-native activators can be studied after they have been introduced by transfection.

GPCRs regulate a wide-range of biological functions and are one of the most important target classes for drug discovery. GPCR signaling pathways can be categorized into three classes based on the G protein -subunit involved: G_s, $G_{i/o}$ and G_q. The GloResponse™ CRE-*luc2P* HEK293 Cell Line can be used to study and configure screening assays for G_s- and $G_{i/o}$-coupled GPCRs, which signal through cAMP and the cAMP Response Element (CRE). For G_q-coupled GPCRs, which signal through calcium ion release and activate the Nuclear Factor of Activated T-Cells response element (NFAT-RE), the GloResponse™ NFAT-RE-*luc2P* HEK293 Cell Line should be used.

NF-κB-REs are the DNA binding sequences for the NF-κB transcription factor complex, which is responsible for regulating inflammation, immune response, cell growth and apoptosis. The GloResponse™ NF-κB-RE-*luc2P* HEK293 Cell Line is designed for rapid and convenient analysis of any cellular response that results in modulation of NF-κB activities.

The GloResponse™ 9X*GAL4*UAS-*luc2P* HEK293 Cell Line contains nine repeats of GAL4 UAS (Upstream Activator Sequence) driving the transcription of the luciferase reporter gene *luc2P* in response to binding of a fusion protein containing the GAL4 DNA Binding Domain, such as the Estrogen Receptor Ligand Binding Domain in pBIND-ER Vector (Cat.# E1390) when activated by a ligand. This makes the cell line suitable for the study of nuclear receptors or can be used to study other types of protein:protein and protein:DNA interactions. The GAL4 DNA Binding Domain partner must be introduced to this cell line by transfection or other similar techniques.

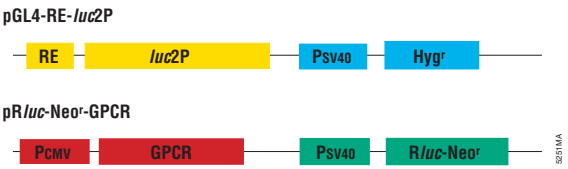
The GloResponse™ Cell Lines were generated by clonal selection of HEK293 cells stably transfected with pGL4-based vectors carrying specific response elements for the pathway of interest. These cell lines incorporate the improvements developed for the pGL4 family of reporter vectors for enhanced performance. The destabilized *luc2P* luciferase reporter is used for improved responsiveness to transcriptional dynamics. The *luc2P* gene is codon optimized for enhanced expression in mammalian cells, and the pGL4 plasmid backbone was engineered to reduce background reporter expression. The result is a cell line with very high induction levels when the pathway of interest is activated.

Features:

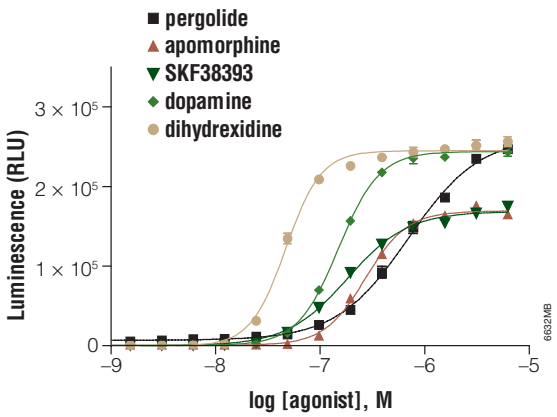
- **Convenient:** Prebuilt, optimized luciferase reporter cell lines.
- **Robust:** Large assay window provided by high levels of induction and low background expression.
- **Faster Results:** Improved responsiveness to transcriptional dynamics with destabilized luciferase.

Protocol	Part#
Glo-Response™ CRE-*luc2P* HEK293 Cell Line Technical Bulletin	TB362
GloResponse™ NFAT-RE-*luc2P* HEK293 Cell Line Technical Bulletin	TB363
GloResponse™ NF-κB-RE-*luc2P* HEK293 Cell Line Technical Bulletin	TB380
Glo-Response™ 9X*GAL4*UAS-*luc2P* HEK293 Cell Line Technical Bulletin	TB552

Storage Conditions: Place frozen cells in storage at less than or equal to –140°C (mechanical deep freeze or vapor phase liquid nitrogen) until you are ready to thaw and propagate them. We strongly recommend that the cells be propagated, using the provided procedure, as soon as possible. This will ensure the optimal cell viability and assay performance.



Two plasmids involved in the dual-luciferase GPCR assay. RE, response element/promoter; ***luc2P***, destabilized firefly luciferase with PEST sequence; P_{SV40}, SV40 promoter; Hyg^r, hygromycin resistance gene; P_{CMV}, CMV promoter; ***Rluc***-neo^r, ***Renilla*** luciferase and neomycin resistance gene fusion. PEST sequences are associated with rapidly degraded proteins.



Ranking compound potency and detection of DRD1 partial agonists. A GloResponse™ CRE-*luc2P* clone stably expressing dopamine receptor D1 was plated at 10,000 cells/well in a 96-well plate. Each agonist was serially diluted 1:2, then added to wells in replicates of four, beginning with 50µM. Cells were incubated with agonist for four hours, harvested and analyzed using the Dual-Glo™ Luciferase Assay System (Cat.# E2920). Luciferase activity was measured on the GloMax® 96 Microplate Luminometer (Cat.# E6501).

ONE-Glo™ Luciferase Assay System

Product	Size	Cat.#
ONE-Glo™ Luciferase Assay System	10 ml	E6110
	100 ml	E6120
	1 L	E6130

For Laboratory Use. Please contact Promega for information on bulk purchases and custom configurations.

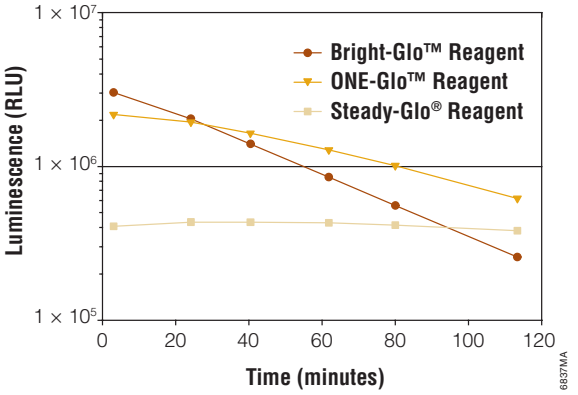
Description: The ONE-Glo™ Luciferase Assay System provides a highly sensitive, robust, homogeneous assay for detection of firefly luciferase reporter gene expression in mammalian cells. Ideally suited for high- and ultrahigh-throughput applications, the ONE-Glo™ Assay contains a new luciferase substrate, resulting in a reagent that is more stable, more tolerant to sample components, and has less odor than standard luciferase assay reagents. These features ensure that the ONE-Glo™ Assay provides robust performance and also eliminates many of the handling inconveniences experienced using other reporter assays in a high-throughput setting.

Features:

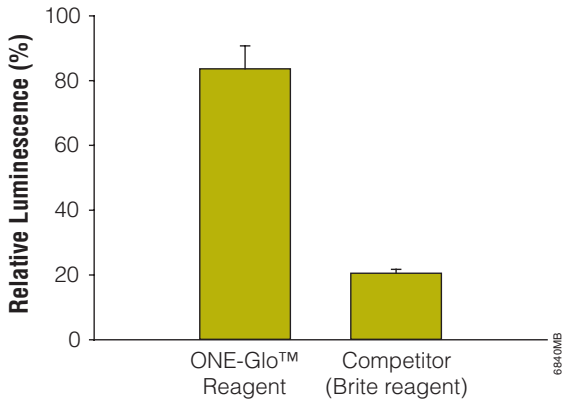
- **Simplify Your Assay Optimization:** The robust performance, reduced odor, improved storage, and larger available sizes make assay optimization even easier and more efficient.
- **Store at Room Temperature or at 4°C:** The extended stability of the ONE-Glo™ Reagent at room temperature and 4°C makes it more convenient for everyday use.
- **Improve Assay Precision:** Because the ONE-Glo™ Reagent is less sensitive to mixing and dispensing conditions, reproducibility is enhanced. The assay is ideal for use in high-density (384- and 1536-well) microplates.
- **Get a Brighter, Longer-Lasting Signal:** Optimized for batch and continuous-process handling, the extended bright light output of the ONE-Glo™ Assay allows high sensitivity, especially when extended incubation is required prior to reading results.
- **Reduce Unwanted Effects from Sample Components:** The novel chemistry used in the ONE-Glo™ Assay is less sensitive to culture media, phenol red, and luciferase inhibitors than other luciferase assays.

Protocol	Part#
Technical Manual	TM292

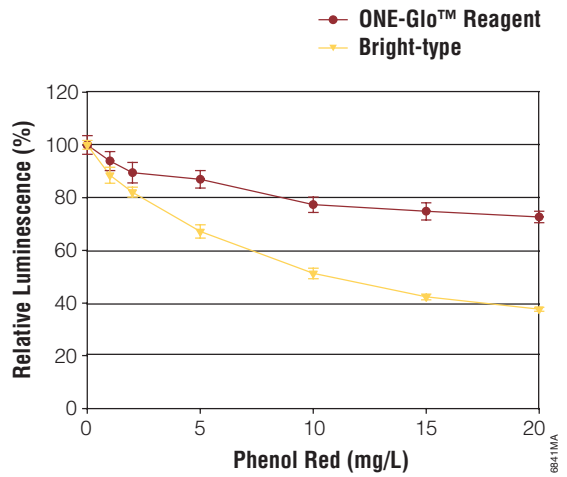
Storage Conditions: Store the ONE-Glo™ Luciferase Assay System components at –20°C. Please refer to the Technical Manual, #TM292, for other storage options, including room temperature storage.



ONE-Glo™ Reagent generates bright and stable luminescence that can easily be measured for multiple hours. Samples in 96-well plates consisted of 50µl of purified firefly luciferase (14.9ng/ml with 0.1% Prionex®) combined with 50µl of the respective reagent. Luminescence was measured (1.0 second integration/well) at 3 minutes and periodically for almost 2 hours. All coefficients of variation were < 3%; n = 3.



ONE-Glo™ Reagent protects the luciferase reaction in the presence of resveratrol, a known luciferase inhibitor (Bakhtiarova, A. *et al.*(2006) *Biochem. Biophys. Res. Comm.* **351**, 481–4). Luciferase reactions generated by ONE-Glo™ Reagent or another bright-type reagent were initiated in the presence or absence of 10µM resveratrol. Luminescence was initiated and measured by the method noted in the figure above. The relative luminescence is the luminescence from reactions containing resveratrol/luminescence from reactions without resveratrol x 100; n = 3.



ONE-Glo™ Reagent is more tolerant of phenol red than luciferin-based reagents. Luciferase reactions composed of 14.9ng/ml luciferase in phenol red-free RPMI medium (with 0.1% Prionex®) and ONE-Glo™ Reagent or another bright-type reagent were initiated in the presence of varied amounts of phenol red. Relative luminescence is the luminescence of reactions containing phenol red/luminescence from reactions without phenol red x 100; n = 3.

Steady-Glo® Luciferase Assay System

Product	Size	Cat.#
Steady-Glo® Luciferase Assay System	10 ml	E2510
	100 ml	E2520
	10 × 100 ml	E2550

Please contact Promega for information on bulk purchases.

Description: High-throughput quantitation of firefly (*Photinus pyralis*) luciferase expression in mammalian cells is commonly performed by batch processing of 96- and 384-well plates. Steady-Glo® Luciferase Assay System is designed for this purpose by providing long-lived luminescence when added to cultured cells. The homogeneous assay provides signal half-lives of over 5 hours in commonly used cell culture media without prior sample processing. Throughput rates of several thousand samples per hour may be achieved with high reproducibility under standard laboratory conditions.

Features:

- **Greater Light Output:** Results in greater assay sensitivity than other leading extended-lifetime firefly luciferase assay reagents.
- **Improved Assay Precision:** Less sensitive to mixing conditions for improved assay reproducibility in multiwell plates. This is particularly useful in 384-well plates.
- **Convenient:** Simply mix buffer with lyophilized substrate and use; there is no need for thawing or measuring before use.
- **Quick:** No need to remove culture medium or wash cells prior to adding assay reagent. Cells may be grown and assayed directly within the same multiwell plate.
- **Easy to Use:** Simply add reagent, which contains a cell lysis component, wait 5 minutes and measure luminescence.
- **Robust:** Compatible with many tissue culture media, including those containing up to 10% serum.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Manual	TM051

Storage Conditions: Store Steady-Glo® Luciferase Assay Substrate at –20°C. Store Steady-Glo® Luciferase Assay Buffer below 25°C.

Bright-Glo™ Luciferase Assay System

Product	Size	Cat.#
Bright-Glo™ Luciferase Assay System	10 ml	E2610
	100 ml	E2620
	10 × 100 ml	E2650

For Laboratory Use. Please contact Promega for information on bulk purchases.

Description: High-throughput quantitation of firefly (*Photinus pyralis*) luciferase expression in mammalian cells requires highly sensitive reagents that can adapt to continuous-process robotic systems. Bright-Glo™ Luciferase Assay System is designed specifically to meet the needs of continuous-process systems by providing robust, homogeneous assay chemistry that achieves high assay sensitivity and approximately 30-minute signal half-life without prior sample processing. These attributes also benefit scientists who are using fewer samples but still require high sensitivity and ease of use.

Features:

- **Quick:** No need to remove culture medium or wash cells prior to adding assay reagent. Cells may be grown and assayed directly within the same multiwell plate.
- **Sensitive:** Up to tenfold more light intensity than other homogeneous luciferase assay reagents results in increased sensitivity.
- **Improved Assay Precision:** Less sensitive to mixing conditions, sample evaporation and pipetting errors, resulting in improved assay reproducibility.
- **Convenient:** Simply mix buffer with lyophilized substrate and add to cells in culture medium; no need for thawing or measuring before use.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Manual	TM052

Storage Conditions: Store Bright-Glo™ Luciferase Assay Substrate at –20°C. Store Bright-Glo™ Luciferase Assay Buffer below 25°C.

Glo Lysis Buffer, 1X

Product	Size	Cat.#
Glo Lysis Buffer, 1X	100 ml	E2661

Description: Glo Lysis Buffer (GLB), 1X, is a proprietary formulation developed to promote rapid lysis (within 5 minutes) of cultured mammalian cells without scraping or performing freeze-thaw cycles. It is fully compatible with Bright-Glo™ and Steady-Glo® Assay Reagents and the Luciferase Assay Reagent for analysis of firefly luciferase expression. The half-life of these reagents remains the same with or without use of GLB, >5 hours for Steady-Glo® Reagent and >24 minutes for Bright-Glo™ Reagent.

Features:

- **Convenient:** No need for cell scraping or freeze-thaw cycles.
- **Fast:** Cell lysis within 5 minutes.
- **Versatile:** Use with Bright-Glo™ and Steady-Glo® Reagents to provide nonhomogeneous assay formats or with other reporter applications.
- **Robust:** Firefly luciferase enzyme in Glo Lysis Buffer is stable at room temperature for at least 48 hours.

Protocol	Part#
Promega Product Information	9PIE266

Storage Conditions: Store Glo Lysis Buffer at 4°C. For long-term storage, Glo Lysis Buffer can be frozen at –20°C or –70°C.

Luciferase Assay System

Product	Size	Cat.#
Luciferase Assay System	100 assays	E1500
Luciferase Assay System with Reporter Lysis Buffer	100 assays	E4030
Luciferase Assay System, 10-Pack	1,000 assays	E1501
Luciferase Assay System Freezer Pack	1,000 assays	E4530
Luciferase 1000 Assay System	1,000 assays	E4550
Available Separately	**Size**	**Cat.#**
Luciferase Assay Reagent	1,000 assays	E1483
Luciferase Cell Culture Lysis 5X Reagent	30 ml	E1531
Reporter Lysis 5X Buffer	30 ml	E3971
Glo Lysis Buffer, 1X	100 ml	E2661

Description: The Luciferase Assay System is an extremely sensitive and rapid reagent for quantitation of firefly luciferase. Linear results are seen over at least eight orders of magnitude of enzyme concentration, and patented technology incorporated in the formulation has allowed for less than 10^{-20} moles of luciferase to be measured under optimal conditions. Generally, 100-fold greater sensitivity can be achieved over the chloramphenicol acetyltransferase (CAT) assay. The Luciferase Assay Reagent generates light that is nearly constant for at least 1 minute and so is compatible with measuring firefly luciferase in a single-tube luminometer or in a multiwell plate luminometer with an autoinjector.

The Luciferase Assay System is a nonhomogeneous assay system; the cells containing the luciferase must be lysed before reagent addition. Glo Lysis Buffer (Cat.# E2661), Cell Culture Lysis Reagent (Cat.# E1531), Passive Lysis Buffer (Cat.# E1941) and Reporter Lysis Buffer (Cat.# E3971) may be used with the Luciferase Assay System for reporter quantitation in mammalian cells. The Luciferase Assay System may also be used for quantitation in plant and bacterial cells, but only Cell Culture Lysis Reagent is suitable for these applications. Reporter Lysis Buffer allows for firefly luciferase, CAT and β-galactosidase assays to be performed from the same cell extract. In some kits the lysis buffer is included, and in others it must be purchased separately.

Features:

- **Linear:** Eight or more orders of magnitude of enzyme concentration.
- **Sensitive:** To 10^{-20} moles of luciferase.
- **Fast:** Perform cell lysis, sample preparation and assays in as little as 5 minutes.
- **Convenient:** Reporter Lysis Buffer allows luciferase, CAT and β-galactosidase assays to be performed from the same cell extract.
- **Simple Assay Procedure:** Eliminates the need for autoinjection devices and rapid mixing protocols when using single-tube luminometers.
- **Versatile:** Luminometer preferred, but not required; adaptable to scintillation counters.
- **Safe:** Non-radioactive.
- **Superior:** High performance compared to competitors' luciferase assays.

Protocol	Part#
Technical Bulletin	TB281

Storage Conditions: Store system at –20°C. Store Cat.# E1483 at –70°C. Reporter Lysis Buffer (Cat.# E3971) may be stored at room temperature. Store Cat.# E2661 at 4°C. For long-term storage, Cat.# E2661 can be frozen at –20°C or –70°C.

QuantiLum® Recombinant Luciferase

Product	Size	Conc.	Cat.#
QuantiLum® Recombinant Luciferase	1 mg	10–15 mg/ml	E1701
	5 mg	10–15 mg/ml	E1702

Description: QuantiLum® Recombinant Luciferase is a luciferase expressed from a cloned gene from the North American firefly (*Photinus pyralis*) that provides the reliability and dependability needed for performing research or producing kits using bioluminescence reagents to detect ATP or luciferin substrates. A recombinant source eliminates the possibility of seasonal and regional variability that may be found in luciferase purified from natural sources.

Features:

- **Value:** Product available in bulk for large orders to suit individual needs and requirements.
- **Reliable:** Long-term supply assurance.
- **Consistent:** Excellent lot-to-lot consistency.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Storage Conditions: Store at –70°C. Avoid multiple freeze-thaw cycles.

Beetle Luciferin, Potassium Salt

Product	Size	Cat.#
Beetle Luciferin, Potassium Salt	5 mg	E1601
	50 mg	E1602
	250 mg	E1603
	1 g	E1605

Description: Luciferase genes from the North American firefly (*Photinus pyralis*) and from other beetles are commonly used as reporter genes for studying transcription regulation in transient assay systems and as markers for stably transformed eukaryotic cells. Beetle luciferin (also known as D-luciferin) is synthesized as the monopotassium salt and is a substrate for the beetle luciferase reporter systems. D-luciferin is provided for those researchers who prefer to formulate their own assay reagents for monitoring in vitro or in vivo luciferase activity.

Formula: $C_{11}H_7N_2O_3S_2{\cdot}K$.

Formula Weight: 318.4Da (anhydrous).

Features:

- **Formulation:** Supplied as a potassium salt for easy preparation in aqueous buffer.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Storage Conditions: Store at –70°C.

Renilla-Glo™ Luciferase Assay System

Product	Size	Cat.#
Renilla-Glo™ Luciferase Assay System	10 ml	E2710
	100 ml	E2720
	10 × 100 ml	E2750

Description: The *Renilla*-Glo™ Luciferase Assay System is a single-addition reagent that generates a glow-type signal with *Renilla* luciferase. When reconstituted, it has the capacity to lyse cells, reduce the autoluminescence of the coelenterazine substrate, and produce a stable signal (i.e., half-life greater than 60 minutes at 22°C).

Features:

- **Simplify Your Assay Optimization:** Add-and-read simplicity for a *Renilla* luciferase reporter system.
- **Improve Assay Precision:** No need for separate lysis and reagent injection steps.
- **Get a Brighter, Longer-Lasting Signal:** Extended bright light output is optimized for batch and continuous-process handling.
- **Reduced Autoluminescence:** Low background formulation offers increased sensitivity.

Protocol	Part#
Technical Manual	TM329

Storage Conditions: Store at –20°C.

Renilla Luciferase Assay System

Product	Size	Cat.#
Renilla Luciferase Assay System	100 assays	E2810
	1,000 assays	E2820

Description: *Renilla* Luciferase Assay System is designed to provide a fast and sensitive method of detecting the luciferase from sea pansy (*Renilla reniformis*). The system is a convenient alternative to firefly (*Photinus pyralis*) reporter systems and is designed to yield reliable, linear results for a concentration range over 7 orders of magnitude. The *Renilla* Luciferase Assay System has been formulated with a proprietary composition that significantly reduces the effect of coelenterazine autoluminescence when compared to other reagents, making the reagent orders of magnitude more sensitive than published methods. This system enables measurements with wildtype and the synthetic *hRluc* genes for primary expression or internal normalization measurements of gene expression.

Features:

- **Reduced Autoluminescence:** Low background, increased sensitivity.
- **Sensitive:** 10^{-19} moles of *Renilla* luciferase detectable.
- **Linear:** Linear range extending 7 logs.
- **Unique:** The first independent assay system for *Renilla* luciferase.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Manual	TM055

Storage Conditions: Store the *Renilla* Luciferase Assay System at –20°C.

EnduRen™ Live Cell Substrate

Product	Size	Cat.#
EnduRen™ Live Cell Substrate	0.34 mg	E6481
	3.4 mg	E6482
	34 mg	E6485

Description: EnduRen™ Live Cell Substrate provides new capabilities in performing luminescent reporter assays by enabling live cell kinetic measurements, streamlining assay development and multiplexing with other lytic assays. EnduRen™ Live Cell Substrate provides the ability to measure *Renilla* luciferase luminescence for at least 24 hours after substrate addition, with up to tenfold higher signal-to-background ratios than wildtype coelenterazines.

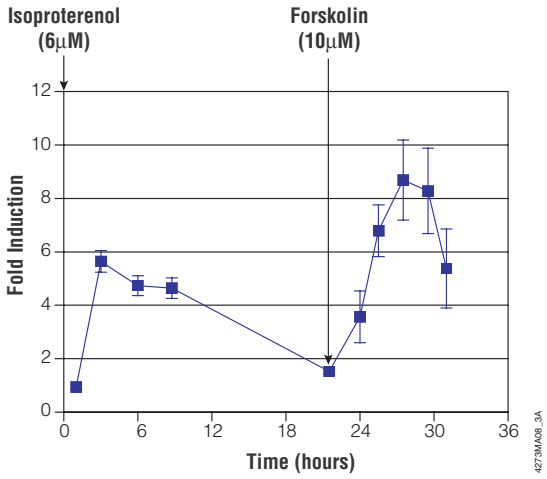
EnduRen™ Live Cell Substrate is a uniquely engineered coelenterazine with protected oxidation sites, which minimizes substrate degradation and autoluminescence (background) in cell culture, while it extends the luminescent signal to accommodate microplates without the need for auto-injectors. The result is that EnduRen™ Live Cell Substrate overcomes the key limitations of wildtype coelenterazines by providing an automation-friendly, highly sensitive substrate for *Renilla* luciferase-based gene reporter and BRET applications.

Features:

- **Live Cell Assay:** Generate kinetic profiles for reporter gene, BRET and RNAi applications.
- **Kinetic Reporter Gene Analysis:** Conserve test compounds as you create response profiles in real time to generate more data-rich results.
- **Streamlined Assay Development and Screening:** Rapidly obtain optimal assay parameters through repeat measurements using only a single cell population. Increase your sample throughput using microplates without time-consuming per-sample reagent injection steps.
- **Designed for Multiplexing:** Perform more dynamic experiments using the same sample set by pairing with any lytic assay.
- **High Signal-to-Background Ratios:** Reliably quantitate low levels of expression for reporter gene detection and BRET.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Manual	TM244

Storage Conditions: Store at –20°C.



Real-Time Live Cell Reporter Assay using the EnduRen™ Substrate. Luminescence was monitored from HEK 293 cells for >24 hours, permitting measurement of the effects of sequential treatment of the cells with isoproterenol and forskolin.

ViviRen™ Live Cell Substrate

Product	Size	Cat.#
ViviRen™ Live Cell Substrate	0.37 mg	E6491
	3.7 mg	E6492
	37 mg	E6495

Description: ViviRen™ Live Cell Substrate is a uniquely engineered coelenterazine that generates three- to fivefold brighter *Renilla* luciferase luminescence than wildtype coelenterazine. Using live cells, achieve up to 100-fold higher signal-to-noise ratios for super-sensitive quantitation of reporter gene, BRET and RNAi activity.

Cat.# E6491 is supplied as a liquid, 60mM in DMSO. Cat.# E6492 and E6495 are supplied as a lyophilized solid.

Features:

- **Three- to Fivefold Brighter** *Renilla* Luminescence than Coelenterazine: Quantitate with confidence using miniaturized formats, low-level expression and CCD imagers.
- **Low Autoluminescence:** Achieve unparalleled sensitivity with up to 100-fold higher signal-to-noise ratios than coelenterazine.
- **Live Cell Assay:** Generate kinetic profiles for reporter gene, BRET and RNAi applications.
- **Multiplex Options:** Improve accuracy and precision by combining with CellTiter-Glo® and other lytic assays.

Protocol	Part#
Technical Manual	TM064

Storage Conditions: Store Cat.# E6491 at –70°C. Store Cat.# E6492 and E6495 at –20°C.

Coelenterazines

Product	Size	Cat.#
Coelenterazine	250 µg	S2001
Coelenterazine-h	250 µg	S2011

Description: Luciferases from *Renilla*, *Aequorea* and other marine organisms are commonly used as indicators or reporters for studying cellular phenomena in expression assays in eukaryotic cells. *Renilla* luciferase is often used as a reporter of transcription regulation, whereas apoaequorin is often used as a calcium indicator. Other uses of coelenterazines include chemiluminescent detection of Reactive Oxygen Species (ROS) in cells or tissues. Promega offers several coelenterazine analogs.

Coelenterazine (native) is the luminescent substrate for *Renilla* luciferase and apoaequorin. **Formula:** $C_{26}H_{21}N_3O_3$. **Formula Weight:** 423.5.

Coelenterazine-h imparts a luminescent intensity with its aequorin complex that is reported to be 10–20 times higher than that of native coelenterazine, making this derivative a useful tool for measuring small changes in Ca^{2+} concentrations. **Formula:** $C_{26}H_{21}N_3O_2$. **Formula Weight:** 407.5.

Features:

- **Highly Pure:** 95%.
- **Custom Capabilities:** Custom packaging and sizes available.
- **Easy to Prepare:** Supplied as a dried substrate for easy preparation in methanol or ethanol.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Storage Conditions: Store at –20°C.

Form: Film.

Bioluminescence Imaging Technology

Product	Size	Cat.#
VivoGlo™ Luciferin, In Vivo Grade	50 mg	P1041
	250 mg	P1042
	1 g	P1043
VivoGlo™ Caspase-3/7 Substrate (Z-DEVD-Aminoluciferin, Sodium Salt)	50 mg	P1781
	5 × 50 mg	P1782
VivoGlo™ Luciferin–βGalactoside Substrate (6-O–βgalactopyranosyl luciferin)	50 mg	P1061
	250 mg	P1062
EnduRen™ In Vivo *Renilla* Luciferase Substrate	0.34 mg	P1111
	3.4 mg	P1112
ViviRen™ In Vivo *Renilla* Luciferase Substrate	0.37 mg	P1231
	3.7 mg	P1232
Available Separately	**Size**	**Cat.#**
pGL4.50[*luc2*/CMV/Hygro] Vector	20 g	E1310
pGL4.51[*luc2*/CMV/Neo] Vector	20 g	E1320

Description: VivoGlo™ Luciferin, in vivo grade. Luciferase genes from the North American firefly (*Photinus pyralis*) and from other beetles are commonly used as light-emitting reporters in cellular and animal models. VivoGlo™ Luciferin is the potassium salt of D-luciferin, the firefly luciferase substrate capable of generating light when a suitable model is used.

VivoGlo™ Caspase-3/7 Substrate (Z-DEVD-Aminoluciferin, Sodium Salt) is a firefly luciferase prosubstrate containing the DEVD tetrapeptide sequence recognized by caspase-3 and -7. Upon activation of caspase-3 or -7, the DEVD peptide is cleaved, and the liberated aminoluciferin reacts with luciferase to generate measurable light. Cleavage has been shown in in cellulo and in vivo systems. For mice, activity of a related salt was demonstrated when 10mg of the substrate in 150µl of saline was injected intraperitoneally. Other references suggest that doses as low as 1.5mg per mouse (50mg/kg) can be used. We recommend conducting a preliminary dose-response study using no more than 500mg/kg.

VivoGlo™ Caspase-3/7 Substrate (Z-DEVD-Aminoluciferin, Sodium Salt) has a minimum solubility of 500mg/ml in PBS, and the resulting solution is stable for at least 3 days at room temperature. Injection is usually done via the intraperitoneal route, and imaging is generally started 10 minutes after injection.

VivoGlo™ Luciferin-β-Galactoside Substrate (6-O-β-Galactopyranosyl-luciferin). Luciferin β-galactoside is a substrate for the commonly used reporter enzyme β-galactosidase. The substrate is cleaved by β-galactosidase to form luciferin and galactose. When used in a model system expressing firefly luciferase, the luciferin is then utilized in a firefly luciferase reaction to generate light.

EnduRen™ in vivo *Renilla* Luciferase Substrate is a uniquely engineered coelenterazine-based compound with protected oxidation sites. These modifications are designed to minimize substrate degradation and autoluminescence. It is reported that EnduRen™ Substrate may have a longer kinetic output when compared to the native coelenterazine substrate when used in an in vivo imaging application in a mouse model.

ViviRen™ in vivo *Renilla* Luciferase Substrate is a uniquely engineered coelenterazine-based compound with protected oxidation sites. These modifications are designed to minimize substrate degradation and autoluminescence. It is reported that the ViviRen™ Substrate demonstrates brighter output when compared to the native coelenterazine substrate when used in an in vivo imaging application in a mouse model.

VivoGlo™ In Vivo Imaging Substrates are provided in cooperation with Xenogen Corporation and Caliper Life Sciences for use in in vivo bioluminescence imaging applications.

XENOGEN now part of Caliper Life Sciences Caliper LifeSciences

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Features:

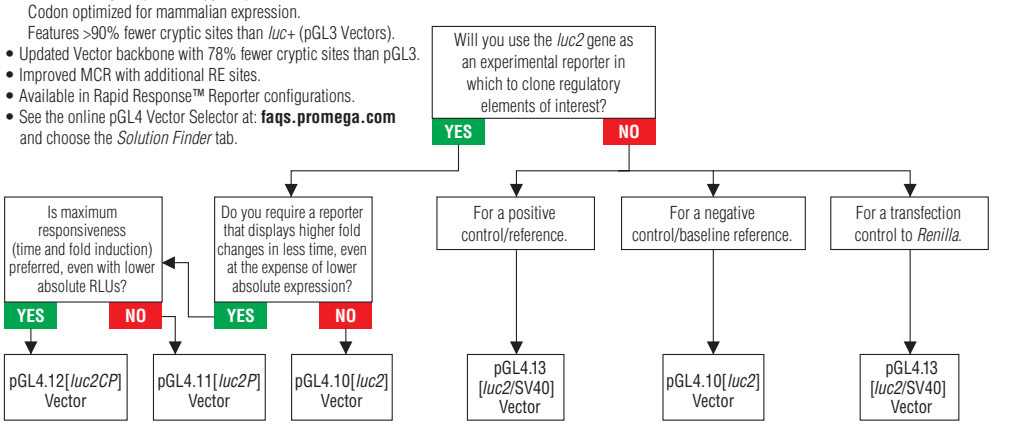
- **Highest Quality Substrates:** Eliminate potential interference in assays due to the presence of endotoxins.
- **Assured Product Integrity:** Most products are packaged in amber vials with septa to ensure product integrity as well as offer ease of dilution and use for imaging experiments. Product is packaged with fine tolerances to minimize the need to weigh substrates.
- **Flexibility and Convenience:** Available in multiple sizes to accommodate a variety of experimental settings.

Protocol	Part#
VivoGlo™ Caspase-3/7 Substrate (Z-DEVD-Aminoluciferin, Sodium Salt) Quick Protocol Card	FB107

Storage Conditions: Store these products at –20°C.

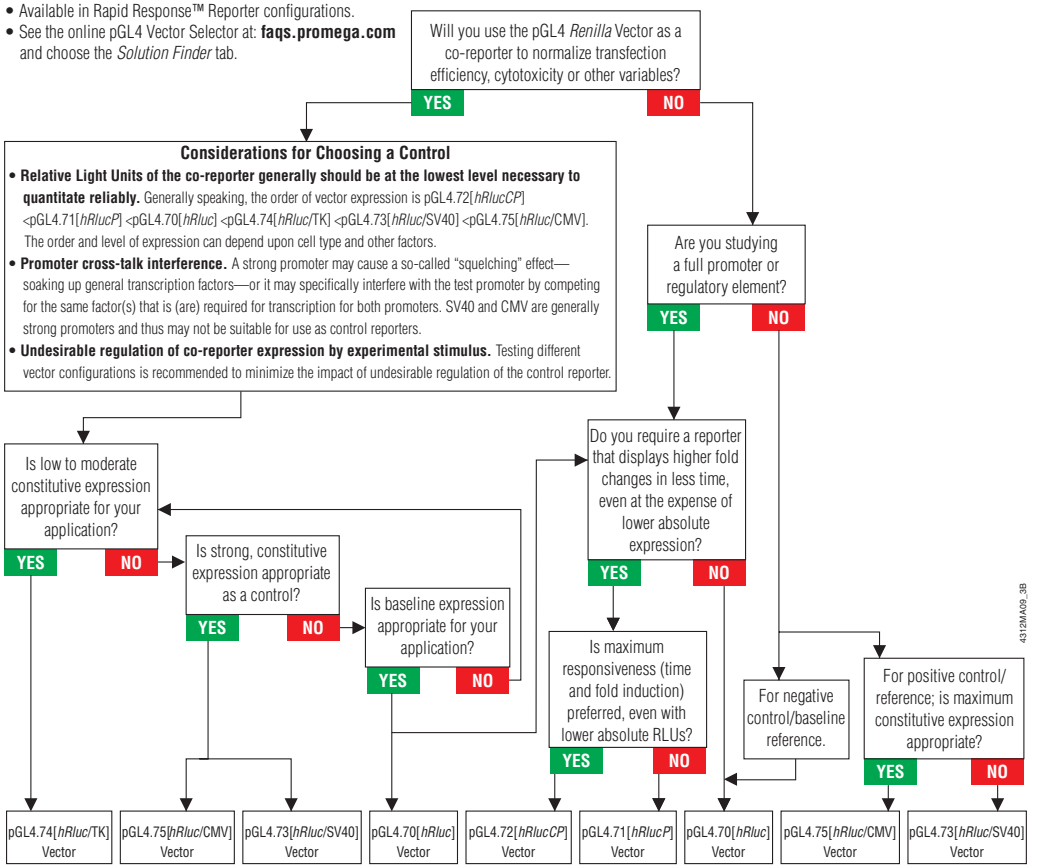
pGL4 Firefly Luciferase Reporter Vectors Selection Guide

- Contains *luc2* gene (*Photinus pyralis*):
  Codon optimized for mammalian expression.
  Features >90% fewer cryptic sites than *luc+* (pGL3 Vectors).
- Updated Vector backbone with 78% fewer cryptic sites than pGL3.
- Improved MCR with additional RE sites.
- Available in Rapid Response™ Reporter configurations.
- See the online pGL4 Vector Selector at: **faqs.promega.com** and choose the *Solution Finder* tab.



pGL4 *Renilla* Luciferase Reporter Vectors Selection Guide

- Contains *hRluc* gene (*Renilla reniformis*).
- Updated Vector backbone with 78% fewer cryptic sites than pGL3/phRG.
- Available in Rapid Response™ Reporter configurations.
- See the online pGL4 Vector Selector at: **faqs.promega.com** and choose the *Solution Finder* tab.



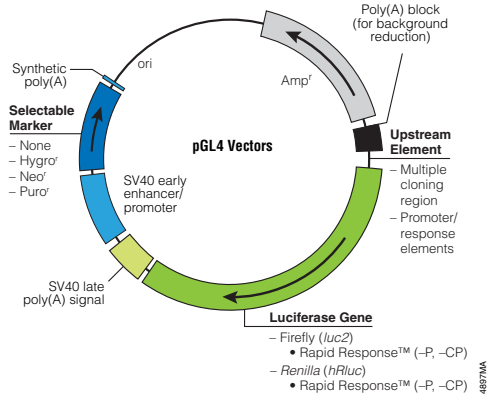
pGL4 Luciferase Reporter Vectors

Product	Size	Cat.#
pGL4.10[*luc2*] Vector	20 µg	E6651
pGL4.11[*luc2P*] Vector	20 µg	E6661
pGL4.12[*luc2CP*] Vector	20 µg	E6671
pGL4.13[*luc2*/SV40] Vector	20 µg	E6681
pGL4.14[*luc2*/Hygro] Vector	20 µg	E6691
pGL4.15[*luc2P*/Hygro] Vector	20 µg	E6701
pGL4.16[*luc2CP*/Hygro] Vector	20 µg	E6711
pGL4.17[*luc2*/Neo] Vector	20 µg	E6721
pGL4.18[*luc2P*/Neo] Vector	20 µg	E6731
pGL4.19[*luc2CP*/Neo] Vector	20 µg	E6741
pGL4.20[*luc2*/Puro] Vector	20 µg	E6751
pGL4.21[*luc2P*/Puro] Vector	20 µg	E6761
pGL4.22[*luc2CP*/Puro] Vector	20 µg	E6771

Product	Size	Cat.#
pGL4.23[*luc2*/minP] Vector	20 µg	E8411
pGL4.24[*luc2P*/minP] Vector	20 µg	E8421
pGL4.25[*luc2CP*/minP] Vector	20 µg	E8431
pGL4.26[*luc2*/minP/Hygro] Vector	20 µg	E8441
pGL4.27[*luc2P*/minP/Hygro] Vector	20 µg	E8451
pGL4.28[*luc2CP*/minP/Hygro] Vector	20 µg	E8461
pGL4.29[*luc2P*/CRE/Hygro] Vector	20 µg	E8471
pGL4.30[*luc2P*/NFAT-RE/Hygro] Vector	20 µg	E8481
pGL4.31[*luc2P/GAL4*UAS/Hygro] Vector	20 µg	C9351
pGL4.32[*luc2P*/NF-B-RE/Hygro] Vector	20 µg	E8491
pGL4.33[*luc2P*/SRE/Hygro] Vector	20 µg	E1340
pGL4.34[*luc2P*/SRF-RE/Hygro] Vector	20 µg	E1350
pGL4.35[*luc2P*/9X*GAL4*UAS/Hygro] Vector	20 µg	E1370
pGL4.36[*luc2P*/MMTV/Hygro] Vector	20 µg	E1360
pGL4.50[*luc2*/CMV/Hygro] Vector	20 µg	E1310
pGL4.51[*luc2*/CMV/Neo] Vector	20 µg	E1320
pGL4.70[*hRluc*] Vector	20 µg	E6881
pGL4.71[*hRlucP*] Vector	20 µg	E6891
pGL4.72[*hRlucCP*] Vector	20 µg	E6901
pGL4.73[*hRluc*/SV40] Vector	20 µg	E6911
pGL4.74[*hRluc*/TK] Vector	20 µg	E6921
pGL4.75[*hRluc*/CMV] Vector	20 µg	E6931
pGL4.76[*hRluc*/Hygro] Vector	20 µg	E6941
pGL4.77[*hRlucP*/Hygro] Vector	20 µg	E6951
pGL4.78[*hRlucCP*/Hygro] Vector	20 µg	E6961
pGL4.79[*hRluc*/Neo] Vector	20 µg	E6971
pGL4.80[*hRlucP*/Neo] Vector	20 µg	E6981
pGL4.81[*hRlucCP*/Neo] Vector	20 µg	E6991
pGL4.82[*hRluc*/Puro] Vector	20 µg	E7501
pGL4.83[*hRlucP*/Puro] Vector	20 µg	E7511
pGL4.84[*hRlucCP*/Puro] Vector	20 µg	E7521

Description: Numerous pGL4 Vectors are available, including those with synthetic firefly *luc2* (*Photinus pyralis*) or *Renilla hRluc* (*Renilla reniformis*) genes, codon optimized for more efficient expression in mammalian cells. The reporter genes and vector backbone, the ampicillin (Amp^r) gene and mammalian selectable marker genes for hygromycin (Hyg^r), neomycin (Neo^r) and puromycin (Puro^r) have been engineered to reduce the number of consensus transcription factor binding sites, reducing background and the risk of anomalous transcription.

The pGL4 Vector backbone is provided with either the *luc2* or *hRluc* gene and, in certain vectors, the two Rapid Response™ Reporter genes. The proteins encoded by the Rapid Response™ Luciferase genes respond more quickly and with greater magnitude to changes in transcriptional activity than their more stable counterparts.



Generic pGL4 Vector map showing the variety of genes, selectable markers, promoters and response elements available.

Vectors are available with a minimal promoter upstream of the *lucP* reporter gene, useful for cloning in a response element of choice. Vectors also are available with a minimal promoter plus choice of response elements or other promoter elements that can be used to study cellular signaling and other events. These vectors can be used in transient transfections or to generate a stable cell line. Please see the table below for available vectors.

Response Element pGL4 Vectors.

Vector Name	Response Element	Signaling Pathway
pGL4.29[*luc2P*/CRE/Hygro] Vector Cat.# E8471	cyclic AMP Response Element	cAMP/PKA
***pGL4.30[luc2P*/NFAT-RE/Hygro] Vector** Cat.# E8481	Nuclear Factor of Activated T-Cells (NFAT) Response Element	Calcium/Calcineurin
***pGL4.31[luc2P/GAL4*UAS/Hygro] Vector** Cat.# C9351	GAL4 upstream activating sequence	Varies (requires binding and activation by GAL4-DNA Binding Domain)
***pGL4.32[luc2P*/NF-κB-RE/Hygro] Vector** Cat.# E8491	Nuclear Factor κB Response Element	NF-κB
***pGL4.33[luc2P*/SRE/Hygro] Vector** Cat.# E1340	Serum Response Element	MAP/ERK
***pGL4.34[luc2P*/SRF-RE/Hygro] Vector** Cat.# E1350	Serum Response Factor Response Element	RhoA
***pGL4.35[luc2P*/9X *GAL4*UAS/Hygro] Vector** Cat.# E1370	GAL4 upstream activating sequence	Varies (requires binding and activation by GAL4-DNA Binding Domain)
***pGL4.36[luc2P*/MMTV/Hygro] Vector** Cat.# E1360	Murine Mammary Tumor Virus Long Terminal Repeat	Several nuclear receptors including androgen receptor and glucocorticoid receptor

9538LA

Promega has formed a strategic partnership with SwitchGear Genomics to promote the use of reporter systems for detailed analyses of regulatory pathways in living cells. Using Promega pGL4 technology, SwitchGear has created ready-to-screen constructs representing thousands of promoters, UTRs and other regulatory elements across the human genome.

Features:

Improved Sensitivity and Biological Relevance Due to:

- **Increased Reporter Gene Expression:** Codon optimization of synthetic genes for mammalian expression.
- **Reduced Background and Risk of Expression Artifacts:** Removal of cryptic DNA regulatory elements and transcription factor binding sites.
- **Improved Temporal Response:** Rapid Response™ technology available using destabilized luciferase genes.

Additional Advantages Include:

- **Flexible Detection Options:** Choice of synthetic *luc2* (*Photinus pyralis*) or *hRluc* (*Renilla reniformis*) reporter genes.
- **Easy Transition from Transient to Stable Cells:** Choice of mammalian selectable markers.
- **Easy Transfer from Vector to Vector:** Common multiple cloning site and a unique SfiI transfer scheme.

Protocol	Part#
Technical Manual	TM259
pGL4.29[*luc2P*/CRE/Hygro] Vector Product Information	9PIE847
pGL4.30[*luc2P*/NFAT-RE/Hygro] Vector Product Information	9PIE848
pGL4.32[*luc2P*/NF-κB-RE/Hygro] Vector Product Information	9PIE849
pGL4.33[*luc2P*/SRE/Hygro] Vector Product Information	9PIE134
pGL4.34[*luc2P*/SRF-RE/Hygro] Vector Product Information	9PIE135
pGL4.35[*luc2P*/9XGAL4UAS/Hygro] Vector Product Information	9PIE137
pGL4.36[*luc2P*/MMTV/Hygro] Vector Product Information	9PIE136

Storage Conditions: Store at –20°C.

Luciferase Reporter Assays to Study Nuclear Receptors

Product	Size	Cat.#
pFN26A (BIND) *hRluc*-neo Flexi® Vector	20 µg	E1380
pBIND-ERα Vector	20 µg	E1390
pBIND-GR Vector	20 µg	E1581
Available Separately	**Size**	**Cat.#**
pGL4.35[*luc2P*/9X*GAL4*UAS/Hygro] Vector	20 µg	E1370
GloResponse™ 9X*GAL4*UAS-*luc2P* HEK293 Cell Line	2 vials	E8530
pGL4.36[*luc2P*/MMTV/Hygro] Vector	20 µg	E1360

Description: Nuclear receptors are a class of ligand-regulated transcription factors that help sense the presence of steroids and other molecules inside the cell. While the nuclear receptor typically resides in the cytoplasm, often complexed with associated regulators, the binding of the ligand triggers a translocation into the nucleus, whereby the receptor binds specifically targeted segments of genomic DNA through its DNA-binding domain. DNA binding leads to upregulation of the adjacent gene, thereby inducing a cellular response to the presence of the ligand.

One method used to study the cellular activity of nuclear receptors in mammalian cells is with the use of luciferase reporter assays in a "one-hybrid" system. In this system, the nuclear receptor ligand binding domain (LBD) is fused to the DNA-Binding Domain of the yeast GAL4 transcription factor. The hybrid fusion-protein nuclear receptor is then used to activate the *luc2P* luciferase reporter under the control of nine repeats of the GAL4 Upstream Activator Sequence (UAS). See overview figure for an illustration.

The pBIND-ERα Vector (Cat.# E1390) contains the yeast Gal4 DNA-Binding Domain (Gal4-DBD) and an estrogen receptor-ligand binding domain (ER-LBD) gene fusion. The pBIND-GR Vector (Cat.# E1581) contains the yeast Gal4 DNA-Binding Domain (Gal4-DBD) and glucocorticoid receptor-ligand binding domain (GR-LBD) gene fusion.

The pFN26A (BIND) *hRluc*-neo Flexi® Vector (Cat.# E1380) is designed to functionally express a fusion protein comprised of a DNA-binding domain of the yeast GAL4 gene, a linker segment and an in-frame protein-coding sequence flanked by SgfI and PmeI sites at the 5′ and 3′ ends, respectively, under the control of the human cytomegalovirus (CMV) immediate early promoter. This vector can be used to clone and test putative transcriptional activation domains for protein sequences of interest, such as the ligand binding domain of many nuclear receptors.

After transfection into a cell and upon binding a ligand, each of the fusion proteins expressed by these vectors can induce the transcription of luciferase when under control of an upstream Gal4 Upstream Activator Sequence (UAS) such as that of the pGL4.35[*luc2P*/9X*GAL4*UAS/Hygro] Vector (Cat.# E1370) or GloResponse™ 9X*GAL4*UAS-*luc2P* HEK293 cells. These products use the destabilized and optimized *luc2P*, allowing greater sensitivity and shorter induction times than more native reporter enzymes.

Each BIND vector also contains a *Renilla* luciferase/neomycin resistance co-reporter. This feature allows the use of dual-luciferase normalization or the construction of a double-stable cell line without the need for additional cloning.

The sequence for the estrogen receptor-ligand binding domain contained in the pBIND-ERα plasmid has a single amino acid difference, which corresponds to amino acid 420 of the GenBank® reference sequence for the full-length estrogen receptor. For information on how the GenBank® sequence performs compared to this sequence, please contact Promega Technical Services.

Features:

- **Robust:** GAL4-based system removes background signals from endogenous receptors.
- **More Sensitive:** Optimized 9X Gal4 gives improved responses, better signal:noise ratio.
- **Adaptable:** Combination *Renilla*/Neomycin marker allows normalization with Dual-Luciferase® Assay or selectable markers for generating stable cell lines, all with one vector.
- **Consistent:** Compare or profile all nuclear receptors with a single experimental system.
- **Faster Results:** Destabilized and optimized *luc2P* luciferase gene allows greater sensitivity and shorter induction times.

Protocol	Part#
pBIND-ER Vector Product Information	9PIE139
pBIND-GR Vector Product Information	9PIE158

Storage Conditions: Store at –20°C.

pmirGLO Dual-Luciferase miRNA Target Expression Vector

Product	Size	Cat.#
pmirGLO Dual-Luciferase miRNA Target Expression Vector	20 µg	E1330

Description: The pmirGLO Vector is designed to quantitatively evaluate microRNA (miRNA) activity by the insertion of miRNA target sites downstream or 3′ of the firefly luciferase gene (*luc2*). Firefly luciferase is the primary reporter gene; reduced firefly luciferase expression indicates the binding of endogenous or introduced miRNAs to the cloned miRNA target sequence. This vector is based on Promega dual-luciferase technology, with firefly luciferase (*luc2*) used as the primary reporter to monitor mRNA regulation and *Renilla* luciferase (*hRluc-neo*) acting as a control reporter for normalization and selection.

Features:

- Measure the function of miRNA and miRNA binding sites.
- *luc2* luciferase gene provides highest expression.
- Combination *Renilla*/Neomycin marker allows for normalization with Dual-Luciferase® Assay or selectable markers for generating stable cell lines, all with one vector.
- Moderate-strength PGK promoter provides sensitive analysis not possible with strong promoters.

Protocol	Part#
Promega Product Information	9PIE133

Storage Conditions: Store at –20°C.

Chroma-Luc™ Vectors

Product	Size	Cat.#
pCBR-Basic Vector	20 µg	E1411
pCBR-Control Vector	20 µg	E1421
pCBG68-Basic Vector	20 µg	E1431
pCBG68-Control Vector	20 µg	E1441
pCBG99-Basic Vector	20 µg	E1451
pCBG99-Control Vector	20 µg	E1461

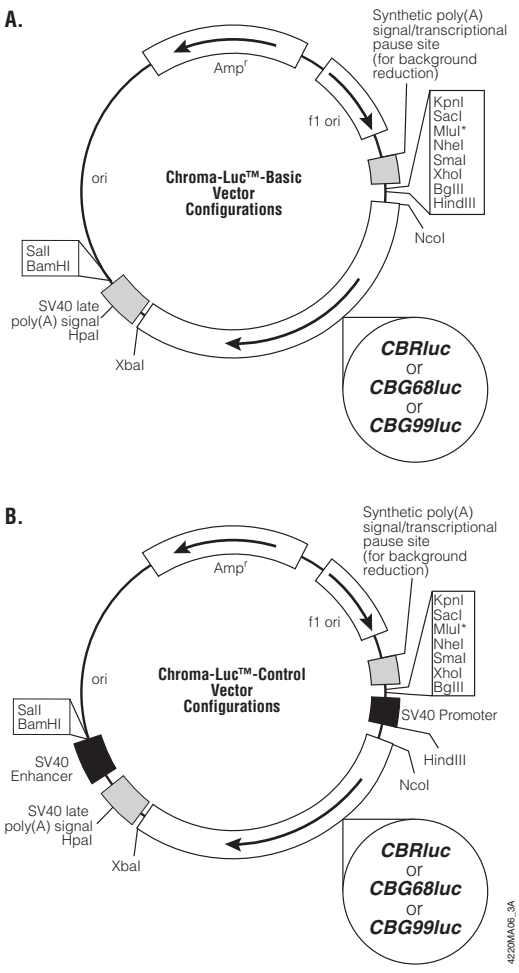
Description: The Chroma-Glo™ Luciferase Assay System and the Chroma-Luc™ Vectors can be used to generate red and green (dual-color) luminescence from a single sample upon addition of a single reagent. The Chroma-Luc™ Vectors consist of 6 plasmids containing synthetic versions of a red or one of two green click beetle luciferase genes; *CBRluc* contains a red-emitting luciferase gene, while *CBG68luc* and *CBG99luc* contain green-emitting luciferase genes. Filtered measurement of the dual-color luminescence produced by the Chroma-Luc™ luciferases permits each reporter to be measured independently and virtually simultaneously. Besides their different luminescence colors, the three Chroma-Luc™ genes differ as follows: *CBG99luc* and *CBRluc* possess 99% DNA and 98% protein homology and are the ideal choice for use when working with transient expression assays; *CBG68luc* and *CBRluc* possess 68.9% DNA homology while retaining a high degree of protein homology (>98%) and thus are the preferred pair for use with stable expression assays. Each of these genes is provided either in a Basic Vector configuration containing a multiple cloning site (MCS) or a Control Vector containing an SV40 promoter and enhancer. The Chroma-Glo™ Assay has a homogeneous format that generates luminescence with >30-minute signal half-lives for each of the Chroma-Luc™ Luciferases, thereby enabling the processing of many plates without prior sample preparation. Two reporter gene measurements can be efficiently and reproducibly determined from each well in a typical high-throughput screen.

Features:

- **Two Reporter Signals by Single Substrate Addition:** Increase your accuracy and precision through normalization, or use both reporters to multiplex experimental measurements. Use filters to spectrally separate the luminescent signals.
- **Ideal Control or Multiplexed Reporter System:** Use the high-homology red and green luciferases to minimize potential RNA and protein effects on reporter expression.
- **Flexible:** Use the Basic Vectors for cloning regulatory elements of interest, or use the Control Vectors as an internal control.
- **High Expression with Minimal Anomalous Transcription Behavior:** Use the synthetic gene design to obtain results easily and reliably.

Protocol	Part#
Technical Manual	TM059

Storage Conditions: Store at –20°C.



The Chroma-Luc™-Basic and -Control Vectors. These vectors contain *CBRluc* or *CBG68luc* or *CBG99luc*; Amp^r, a gene conferring ampicillin resistance in *E.coli*; ori, origin of plasmid replication in *E.coli*. Arrows within the Chroma-Luc™ and Amp^r genes indicate the direction of functionality. *MluI should not be used in the vector configuration containing *CBG99luc*, as this gene also contains the MluI site.

pRL Family of *Renilla* Luciferase Control Reporter Vectors

Product	Size	Cat.#
pRL-SV40 Vector	20 µg	E2231
pRL-TK Vector	20 µg	E2241
pRL-CMV Vector	20 µg	E2261
pRL-null Vector	20 µg	E2271

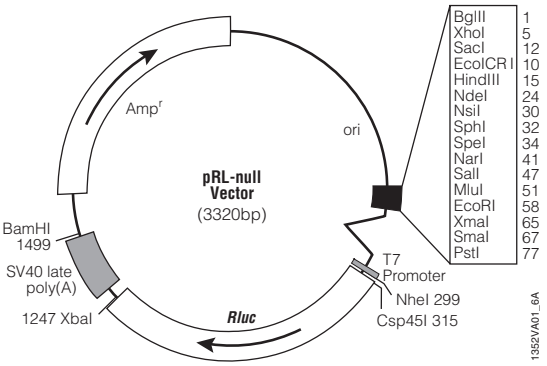
Description: The pRL Vectors are a family of wildtype *Renilla* luciferase (*Rluc*) control reporter vectors. The pRL Vectors, which provide constitutive expression of *Renilla* luciferase, may be used in combination with a firefly luciferase vector to cotransfect mammalian cells. Expression of *Renilla* luciferase provides an internal control value to which expression of the experimental firefly luciferase reporter gene may be normalized. The pRL Vectors contain the cDNA encoding *Renilla* luciferase (*Rluc*) cloned from the anthozoan coelenterate *Renilla reniformis* (sea pansy). Four different promoter configurations are available. The HSV-thymidine kinase promoter (pRL-TK) is relatively weak and may be particularly useful in providing neutral constitutive expression of the *Renilla* luciferase control reporter. The early SV40 enhancer/promoter region (pRL-SV40) and the CMV immediate early enhancer/promoter region (pRL-CMV) typically provide high-level transcription and, therefore, may be less suitable for co-reporter applications involving experimental vectors with robust regulatory elements. In general, we recommend validating the performance of specific co-reporter combinations in the desired target cells. In addition to the modified *Rluc* reporter gene, all pRL Vectors are isolated from a *dam*–/*dcm*– *E. coli* K host strain, allowing digestion with restriction enzymes that are sensitive to *dam* and *dcm* methylation.

Features:

- A T7 promoter is located immediately upstream of *Rluc*, allowing in vitro synthesis of *Renilla* luciferase.
- The SV40 late poly(A) signal sequence is positioned downstream of *Rluc* to provide efficient transcription termination and mRNA polyadenylation.
- A prokaryotic origin of replication and -lactamase gene allow selected propagation of the pRL vectors in *E. coli* host strains.
- To avoid DNA methylation, all pRL Vectors are isolated from a *dam*–/*dcm*– *E. coli* K host strain.

Protocol	Part#
Technical Bulletin	TB550

Storage Conditions: Store vectors at –20°C.



pGL3 Luciferase Reporter Vectors

Product	Size	Cat.#
pGL3-Control Vector	20 µg	E1741
pGL3-Basic Vector	20 µg	E1751
pGL3-Promoter Vector	20 µg	E1761
pGL3-Enhancer Vector	20 µg	E1771

Description: The pGL3 Luciferase Reporter Vectors provide a basis for the quantitative analysis of factors that potentially regulate mammalian gene expression. These may be *cis*- or *trans*-acting factors. The backbone of the pGL2 Luciferase Reporter Vectors was redesigned for the pGL3 Vectors for increased expression, with a modified coding region for firefly (*Photinus pyralis*) luciferase that has been optimized for monitoring transcriptional activity in transfected eukaryotic cells. The assay of this genetic reporter is rapid, sensitive and quantitative. In addition, the Luciferase Reporter Vectors contain numerous features aiding in the structural characterization of the putative regulatory sequences under investigation.

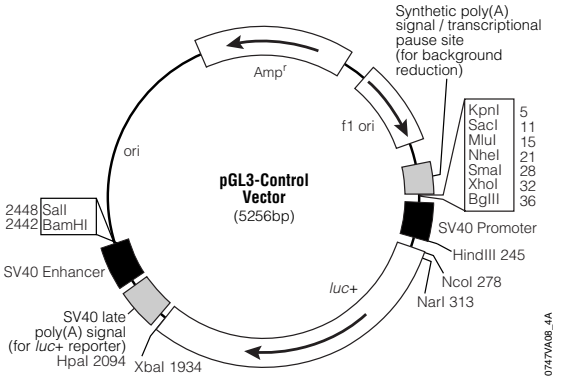
For the most advanced reporter vectors and widest selection of features, please see the pGL4 series of vectors.

Features:

- **Easy to Use:** NcoI site located at 5′ end of *luc*+ gene allows creation of fusions with reporter gene using a unique NcoI site.
- **Flexible:** Placement of SmaI site in the MCS allows blunt-ended inserts to be ligated into the MCS and restricted on either side by other restriction endonucleases.
- **Versatile:** XbaI site just downstream of *luc*+ gene facilitates insertions into the 3′ untranslated region of mRNA or subcloning of the luciferase gene.

Protocol	Part#
Technical Manual	TM033

Storage Conditions: Store vectors at –20°C.



pGL2 Luciferase Reporter Vectors

Product	Size	Cat.#
pGL2-Control Vector	20 µg	E1611
pGL2-Enhancer Vector	20 µg	E1621
pGL2-Promoter Vector	20 µg	E1631
pGL2-Basic Vector	20 µg	E1641

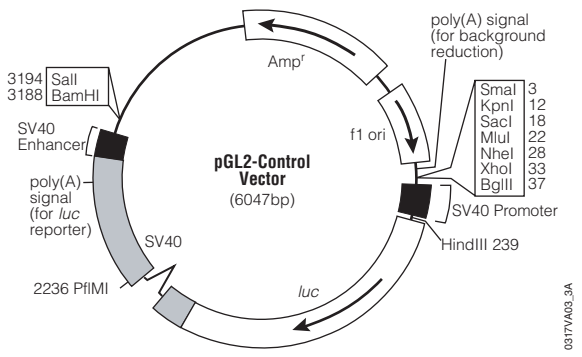
Description: The pGL2 Luciferase Reporter Vectors provide a basis for the quantitative analysis of factors that potentially regulate mammalian gene expression. These factors may be *cis*-acting, such as promoters and enhancers, or *trans*-acting, such as various DNA-binding factors. The pGL2 Vectors carry the coding region for firefly (*Photinus pyralis*) luciferase, which is used to monitor transcriptional activity in transfected eukaryotic cells. The assay of this genetic reporter is rapid, sensitive and quantitative. In addition, the pGL2 Vectors contain numerous features that aid in the characterization and mutagenesis of the putative regulatory sequences.

Features:

- **Versatile:** Deletions and site-directed mutations can be made directly to inserted DNAs without subcloning.
- **Convenient:** All vectors contain the firefly luciferase reporter gene, which enables sensitive and rapid quantitation of reporter activity.
- **Low Background:** Upstream polyadenylation signal minimizes spurious transcription of the reporter gene.

Protocol	Part#
Technical Manual	TM003

Storage Conditions: Store vector at –20°C. Store bacterial strain at –70°C.



pGEM®-*luc* DNA

Product	Size	Cat.#
pGEM®-*luc* DNA	20 µg	E1541

Description: The pGEM®-*luc* Vector is a cassette vector designed to be a source of the *luc* gene encoding firefly luciferase, which is found in the pGL2 Vectors. The plasmid is not intended for the expression of luciferase in eukaryotic or prokaryotic cells.

The pGEM®-*luc* Vector was constructed by positioning the luciferase gene (*luc*) in the center of the multiple cloning region of the pGEM®-11Zf(–) Vector, providing a number of unique restriction sites at both ends of the gene. Sites that are surrounded by parentheses are not unique, as additional sites for each also exist in the luciferase gene. Note also that using HindIII or NsiI to clone the luciferase gene will include upstream ATG codons, which may reduce the efficiency of expression in eukaryotes. The luciferase cassette does not contain the prokaryotic Shine-Delgarno sequence for bacterial expression.

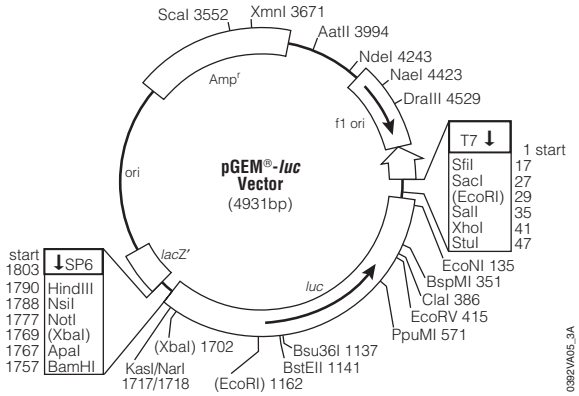
The pGEM®-*luc* Vector is supplied with a glycerol stock of bacterial strain JM109.

Features:

- **Flexibility:** Provides a luciferase cassette with several unique cloning sites at both ends for analysis of transcriptional activity, mRNA processing, protein structure/function, or labeling of cells and viruses.

Protocol	Part#
Technical Bulletin	TB104

Storage Conditions: Store at –20°C. Store bacterial strain at –70°C.



Reporter Vector and Luciferase Sequencing Primers

Product	Size	Cat.#
RVprimer3 (clockwise)	2 µg	E4481
RVprimer4 (counterclockwise)	2 µg	E4491
GLprimer1 (clockwise)	2 µg	E1651
GLprimer2 (counterclockwise)	2 µg	E1661

Description: The Reporter Vector (RV) Sequencing Primers are designed for use with the pGL3 and pGL4 Luciferase Vectors, Chroma-Luc™ Vectors and pCAT®3 Reporter Vectors. RVprimer3 binds upstream of the *luc+*, *luc2* or CAT gene, and sequencing runs clockwise across the multiple cloning region. RVprimer4 binds downstream of the *luc+*, *luc2* or CAT polyadenylation region in the Promoter and Basic Vectors and downstream of the SV40 enhancer region of the Enhancer and Control Vectors. Both primers can be used for sequencing double-stranded templates, but only RVprimer4 can be used for sequencing single-stranded templates.

The GLprimer1 sequences clockwise across the cloning sites upstream of the luciferase gene in the pGL2 Vectors. GLprimer2 sequences counterclockwise across the cloning sites upstream of the luciferase gene in pGL2 or pGL3 Vectors. Both GLprimers can be used for sequencing double-stranded DNA, but only the GLprimer2 can be used for sequencing single-stranded DNA.

Primer Sequences

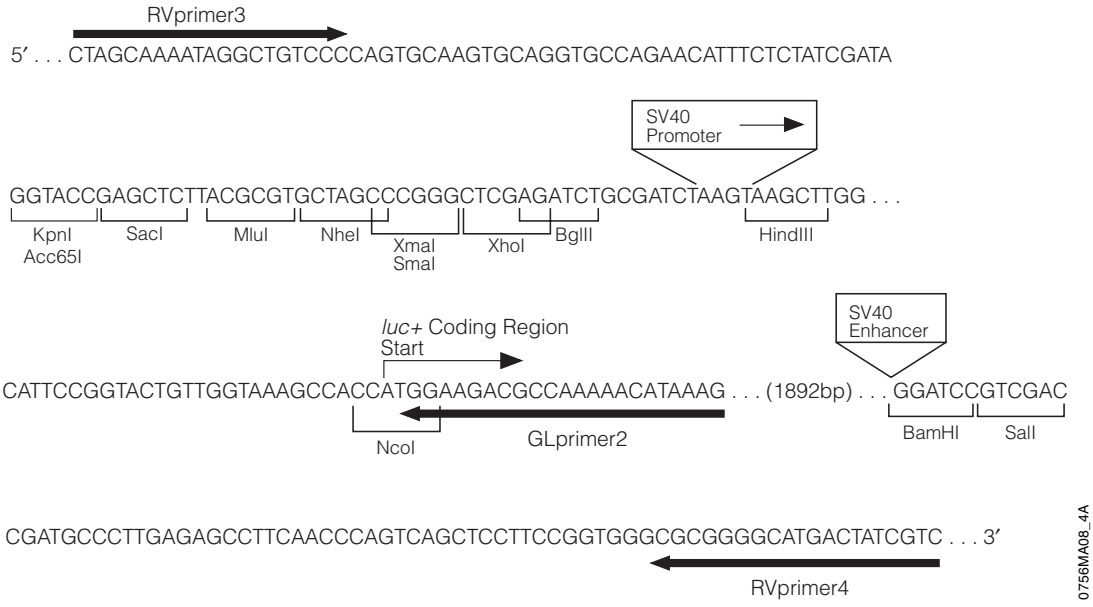
GLprimer1: 5′-d(TGTATCTTATGGTACTGTAACTG)-3′
GLprimer2: 5′-d(CTTTATGTTTTTGGCGTCTTCCA)-3′
RVprimer3: 5′-d(CTAGCAAAATAGGCTGTCCC)-3′
RVprimer4: 5′-d(GACGATAGTCATGCCCCGCG)-3′

Storage Conditions: Store at –20°C. The primers are supplied dried.

Reporter Vector Sequencing Primer Information.

	GLprimer2 Sequences from *luc* ORF into multiple cloning region. Will sequence through SV40 promoter if present.	RVprimer3 Sequences from upstream of multiple cloning region into multiple cloning region.	RVprimer4 Sequences from downstream of reporter ORF and polyadenylation sequences into SalI, BamHI multiple cloning region, which is intended for cloning enhancer elements.
pGL3 Vectors	✓	✓	✓
pCAT®3 Vectors		✓	✓
Chroma-Luc™ (Click Beetle) Vectors (pCBR, pCBG68, pCBG99)		✓	✓
pGL4 Vectors		✓	✓

9490LA



pGL3 Vector multiple cloning region.The upstream and downstream cloning sites and the locations of the sequencing primers, RVprimer3, GLprimer2 and RVprimer4, are shown.The arrows above the primers indicate the direction of sequencing.The positions of the promoter (in pGL3-Promoter and pGL3-Control) and the enhancer (in pGL3-Enhancer and pGL3-Control) are shown as insertions into the sequence of pGL3-Basic (note that the promoter replaces four bases of pGL3-Basic).The sequence shown is of the ssDNA produced using the f1 origin.

pSP-*luc*+NF Fusion Vector

Product	Size	Cat.#
pSP-*luc*+NF Fusion Vector	20 µg	E4471

Description: The pSP-*luc*+NF Fusion Vector is a luciferase cassette vector containing the engineered firefly luciferase gene, *luc*+NF. The *luc*+NF gene is related to the *luc*+ gene found in the pGL3 family of eukaryotic reporter vectors but has been further modified for maximum flexibility in constructing N-terminal fusions (NF) with luciferase. Subcloning *luc*+NF into expression vectors provides a useful genetic reporter with exceptional sensitivity. The pSP-*luc*+NF Fusion Vector is not itself intended for the expression of luciferase in eukaryotic cells, because it does not contain eukaryotic promoters, enhancers or polyadenylation signals.

A unique BstEII site has been inserted immediately downstream of the luciferase ATG translation codon, allowing cloned inserts to be positioned immediately downstream of the *luc*+NF initiation codon. This vector is recommended specifically for applications where N-terminal fusion proteins do not contain an internal ATG codon at the luciferase junction.

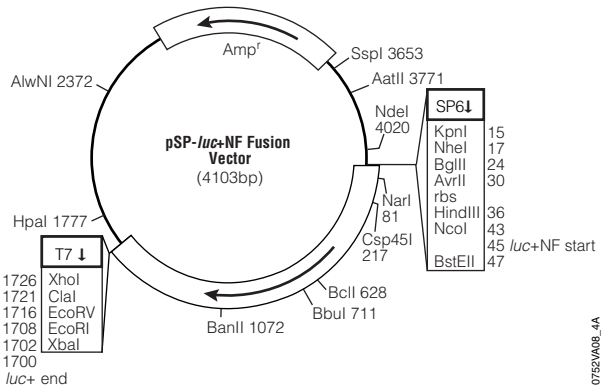
The *luc*+NF gene is positioned downstream of an SP6 promoter and a ribosome binding site. An opposing T7 promoter is located immediately downstream of *luc*+NF. Thus, the pSP-*luc*+NF Fusion Vector provides a convenient template for the in vitro synthesis of both sense and antisense luciferase transcripts for studies involving in situ hybridization, RNA processing, RNA transfection or coupled in vitro transcription/translation and protein folding. Multiple cloning regions containing recognition sequences for commonly used restriction enzymes are positioned at the 5′ and 3′ ends of *luc*+NF to provide maximum flexibility in cloning. Luciferase enzymatic activity can be assayed most efficiently using one of the Luciferase Assay Systems.

Features:

- **Flexibility:** Multiple cloning regions are positioned at the 5′ and 3′ ends of *luc* to provide maximum flexibility in cloning.
- **N-Terminal Fusions with Luciferase:** Unique BstEII site located immediately downstream of the luciferase ATG translation codon.

Protocol	Part#
Technical Bulletin	TB210

Storage Conditions: Store at –20°C.



GloMax®-Multi+ Detection System with Instinct Software

Product	Size	Cat.#
GloMax®-Multi+ Detection System with Instinct Software: Base Instrument with Shaking	1 each	E8032
GloMax®-Multi+ Detection System with Instinct Software: Base Instrument with Heating and Shaking	1 each	E9032
GloMax®-Multi+ Luminescence Module	1 each	E8041
GloMax®-Multi+ Fluorescence Module	1 each	E8051
GloMax®-Multi+ Visible Absorbance Module	1 each	E8061
GloMax®-Multi+ UV-Visible Absorbance Module	1 each	E9061
Available Separately	**Size**	**Cat.#**
GloMax® Injector Tips Replacement	1 each	E5401
GloMax® Luminometer Light Plate	1 each	E6531
Single Injector System for GloMax®-Multi Detection System	1 each	E7071
Dual Injector System for GloMax®-Multi Detection System	1 each	E7081
Cable, USB 2.0 A-B Male	1 each	E8072
DB-15 Communication Cable	1 each	E8081
GloMax®-Multi Optical Kit AFC	1 each	E8917
GloMax®-Multi Optical Kit Blue	1 each	E8921
GloMax®-Multi Optical Kit UV	1 each	E8922
GloMax®-Multi Optical Kit Green	1 each	E8923
GloMax®-Multi Optical Kit Red	1 each	E8924
Injector Inlet Tubing Assembly	1 set	E8925
Injector Outlet Tubing Assembly for Single-Injector System	1 each	E8926
Injector Outlet Tubing Assembly for Dual-Injector System	1 each	E8927
Waste Collection Tray	1 each	E8928
GloMax®-Multi Detection System 490nm Absorbance Filter Set	1 each	E8929
USB Flash Drive, 2.0, 2GB	1 each	E8935
GloMax®-Multi+ Detection System Power Supply—24V, 150W	1 each	E8942
GloMax®-Multi+ Detection System 6-384 Well Plate Adapter	1 each	E8943
GloMax®-Multi+ Detection System 96 Well Optical Crosstalk Mask	1 each	E8944
GloMax®-Multi+ Detection System 384 Well Optical Crosstalk Mask	1 each	E8945
GloMax®-Multi+ Base Instrument Service Agreement, 1 year	1 each	SA3030
GloMax® Injectors Service Agreement, 1 year	1 each	SA3040

Cat.# E8032 and E9032 cannot be sold separately and must be purchased with at least one detection module (Cat.# E8041, E8051, E8061 or E9061).

Request a quote.

Description: The GloMax®-Multi+ Detection System with Instinct Software combines the superior performance expected from single-mode instruments with the functionality of multiple modes. Detection modes include Fluorescence Intensity, Luminescence and UV-Visible Absorbance. The GloMax®-Multi+ Detection System accepts 6-, 12-, 24-, 48-, 96- and 384-well plates and is configured with a factory-installed shaker that allows for either linear or orbital shaking. The GloMax®-Multi+ Detection System may be purchased with an optional heater allowing for precise temperature control from 2°C above ambient temperature to 45°C. The GloMax®-Multi+ Detection System has a touch screen interface with an easy-to-use software program. The Instinct software puts data analysis capabilities on the touch screen. The protocol composer allows complex protocols to be easily developed by combining multiple technologies. In addition, a variety of protocols and common laboratory assays are pre-installed. The system works as a standalone in the laboratory, freeing computing resources from data capture, so more resources can be for use toward other applications.

The GloMax®-Multi+ Detection System with Instinct Software is made up of a base unit available in two formats, one with shaking (E8032) and one with heating and shaking (E9032), plus modular detection and functional units, allowing for a flexible solution that can be expanded over time. Luminescence, fluorescence and absorbance reading modules are available as well as an optional injector system (used with the luminescence detection module only).

Luminescence Module: The luminescence module can detect as little as 3×10^{-21} moles of luciferase, covering a dynamic range over 8 logs. A dual-masking system minimizes well-to-well cross-talk.

Fluorescence Module: Application-optimized Optical Kits simplify fluorescence operation while maximizing performance. Long-lived LED-based excitation lights minimize maintenance and variability in intensity. LED usage increases sensitivity by fully exciting the fluorophore and reducing nonspecific light leakage, a problem often found when using broad-spectrum light sources. The UV, Blue, Green and Red Optical Kits are included with the Fluorescence Detection Module. An AFC Optical Kit is available as an accessory.

- UV (Ex: 365nm, Em: 410–460nm)
- Blue (Ex: 490nm, Em: 510–570nm)
- Green (Ex: 525nm, Em: 580–640nm)
- Red (Ex: 625nm, Em: 660–720nm)
- AFC (Ex: 405nm, Em: 495–505nm)

Visible Absorbance Module: A 6-position filter wheel with 2 open positions ensures flexibility for a wide range of applications. An LED-based visible spectrum light source minimizes maintenance and variability. The Visible Absorbance Module has a reading range of 0–5.0 OD with an accuracy that deviates less than 2%. This module comes with filters for reading 450, 560, 600 and 750nm. A 490nm filter is available as an accessory.

UV-Visible Absorbance Module: This module comes with a 6-position filter wheel that includes filters for measuring 260, 280, 450, 560, 600 and 750nm. These filters accommodate UV DNA and protein quantitation in addition to ELISA and protein assays. Like the Visible Absorbance Module, you can customize the UV-Visible Absorbance Module by substituting a filter of your choice into either of two removable filter paddles.

Operation of the GloMax®-Multi+ Detection System can be performed entirely through the touch screen. Data can be saved on the instrument and moved via the included USB flash drive.

8839TA

GloMax®-Multi+ Detection System with Instinct Software.

Features:

- **Instinct Software:** Label samples and see analyzed data and graphs on the touch screen.
- **Measurement Techniques:** Luminescence, fluorescence and UV-Vis absorbance capabilities.
- **Flexible Modular Configuration:** Modular system grows with your needs.
- **Microplate Formats:** Reads 6-, 12-, 24-, 48-, 96- and 384-well plate formats.
- **Factory-Installed Shaker:** Enables shaking in either linear or orbital mode.
- **Optional Heater Available:** Allows precise temperature control from 2°C above room temperature to 45°C +/– 0.75 °C.
- **Dedicated Luminometer Performance:** Sensitive to approximately 3×10^{-21} moles of luciferase with over 8 logs of dynamic range.
- **Multiplex Cell-Based Assays:** Obtain more data from each experiment.
- **Engineered to Minimize Sample Cross-Talk:** Expect reliable results in all read modes.
- **Simple-to-Use Drag-and-Drop Protocol Composer:** Easily develop complex protocols.
- **Convenient, Standalone Operation:** Eliminate bottlenecks and free analysis resources.
- **Injector Systems:** Both single and dual injectors are available.

Protocol	Part#
Technical Manual	TM340

GloMax®-Multi Detection System

Product	Size	Cat.#
GloMax®-Multi Base Instrument	1 each	E7031
GloMax®-Multi Luminescence Module	1 each	E7041
GloMax®-Multi Fluorescence Module	1 each	E7051
GloMax®-Multi Absorbance Module	1 each	E7061
Available Separately	**Size**	**Cat.#**
Single Injector System for GloMax®-Multi Detection System	1 each	E7071
Dual Injector System for GloMax®-Multi Detection System	1 each	E7081
GloMax®-Multi Detection System External PC Connect Kit	1 each	E8916
GloMax® Luminometer Light Plate	1 each	E6531
GloMax®-Multi Optical Kit Blue	1 each	E8921
GloMax®-Multi Optical Kit UV	1 each	E8922
GloMax®-Multi Optical Kit Green	1 each	E8923
GloMax®-Multi Optical Kit Red	1 each	E8924
GloMax®-Multi Optical Kit AFC	1 each	E8917
Injector Inlet Tubing Assembly	1 set	E8925
Injector Outlet Tubing Assembly for Single-Injector System	1 each	E8926
Injector Outlet Tubing Assembly for Dual-Injector System	1 each	E8927
Waste Collection Tray	1 each	E8928
GloMax® Injector Tips Replacement	1 each	E5401
USB Flash Drive, 2.0, 2GB	1 each	E8935
GloMax®-Multi Detection System 490nm Absorbance Filter Set	1 each	E8929
GloMax®-Multi Base Instrument Service Agreement	1 each	SA3020

Cat.# E7031 cannot be sold separately and must be purchased with at least one detection module (Cat.# E7041, E7051 or E7061).

Request a quote.

Available Separately	Size	Cat.#
GloMax® Injectors Service Agreement, 1 year	1 each	SA3040

Cat.# E7031 cannot be sold separately and must be purchased with at least one detection module (Cat.# E7041, E7051 or E7061).

Request a quote.

Description: The GloMax®-Multi Detection System is made up of a base unit plus modular detection and functional units, allowing for a flexible solution that can be expanded over time. Luminescence, fluorescence and absorbance reading modules are available as well as an optional injector system (used with the luminescence detection module only).

Luminescence Module: An advanced photon-counting photomultiplier tube (PMT) provides unmatched signal-to-noise ratios, beating most standalone luminometers. The luminescence module can detect as little as 3×10^{-21} moles of luciferase, covering a dynamic range over 8 logs. A dual masking system minimizes well-to-well cross-talk.

Fluorescence Module: Application-optimized Optical Kits simplify fluorescence operation while maximizing performance. Long-lived LED-based excitation lights minimize maintenance and variability in intensity. The UV, Blue, Green and Red Optical Kits are included with the Fluorescence Detection Module. An AFC Optical Kit is available as an accessory.

- UV (Ex: 360nm, Em: 410–460nm)
- Blue (Ex: 490nm, Em: 510–570nm)
- Green (Ex: 525nm, Em: 580–640nm)
- Red (Ex: 625nm, Em: 660–720nm)
- AFC (Ex: 405nm, Em: 495–505nm)

Absorbance Module: A 6-position filter wheel with 2 open positions ensures flexibility for a wide range of applications. An LED-based visible spectrum light source minimizes maintenance and variability. Filters for reading 450, 560, 600 and 750nm are included. A 490nm filter is available as an accessory.

Operation of the GloMax®-Multi Detection System can be performed entirely through the touch screen. Protocols for a variety of Promega and common laboratory assays are pre-installed, and there is also the flexibility to modify the protocols or create new methods. Data can be saved on the instrument and moved via the included USB flash drive. The system works as a standalone workstation in the laboratory, freeing computing resources from data capture, so more resources can be directed toward analysis.

Features:

- **Flexible Configuration:** Modular system grows with your needs.
- **Dedicated Luminometer Performance:** Sensitive to approximately 3×10^{-21} moles of luciferase with over 8 logs of dynamic range.
- **Multiplex Cell-Based Assays:** Obtain more data from each experiment.
- **Fluorescence and Absorbance Capability:** Read standard lab assays without compromising luminescence performance.
- **Engineered to Minimize Sample Cross-Talk:** Expect reliable results in all read modes.
- **Convenient, Standalone Operation:** Eliminate bottlenecks and free analysis resources.

Protocol	Part#
Technical Manual	TM297

GloMax®-Multi Jr Single-Tube Multimode Reader

Product	Size	Cat.#
GloMax®-Multi Jr Base Instrument	1 each	E6070
GloMax®-Multi Jr with Luminescence Module	1 each	E6080
Fluorescence Optical Kit, Blue (Ex 460nm, Em 515–570nm)	1 each	E6071
Fluorescence Optical Kit, UV (Ex 365nm, Em 410–450nm)	1 each	E6072
Fluorescence Optical Kit, Green (Ex 525nm, Em 580–640nm)	1 each	E6073
Fluorescence Optical Kit, Red (Ex 625nm, Em 660–725nm)	1 each	E6074
Fluorescence Optical Kit, GFPUV (Ex 365nm, Em 515–570nm)	1 each	E6075
Absorbance Module (User Installable)	1 each	E6076
Absorbance Filter Paddle, 560nm	1 each	E6077
Absorbance Filter Paddle, 600nm	1 each	E6078
Absorbance Filter Paddle, 750nm	1 each	E6079
Available Separately	**Size**	**Cat.#**
GloMax®-Multi Jr Reader Luminescence Module Service Upgrade	1 each	E6098
Minicell Adapter Kit (for measuring 100–200l of sample)	1 each	E6094
Minicell Borosilicate Glass Cuvettes	400 each	E6091
10 × 10mm Square Polystyrene Cuvette (3.5ml capacity)	100 each	E6092
10 × 10mm Square Methacrylate Cuvette (3.5ml capacity)	100 each	E6093
AC Adapter Replacement	1 each	E6095
Thermal Serial Printer and Universal Power Cable	1 each	E2821
Thermal Printer Paper	1 each	E2851
GloMax®-Multi Jr Service Agreement	1 each	SA3080

Cat.# E6070 cannot be sold separately and must be purchased with at least one Fluorescence Optical Kit (Cat.# E6071, E6072, E6073, E6074 or E6075).
Cat.# E6076 cannot be sold separately and must be purchased with at least one Absorbance Filter Paddle (Cat.# E6077, E6078 or E6079).

Request a quote.

Description: The GloMax®-Multi Jr Single-Tube Multimode Reader is designed to provide the utmost flexibility. In addition to high performance, the GloMax®-Multi Jr blends user-friendly operation and a small footprint with flexible purchasing options. The result of this design is an instrument with superior performance that is easy to use, affordable and can be customized to your laboratory's needs.

The GloMax®-Multi Jr with a **Luminescence Module** is designed to deliver performance equivalent to dedicated single-tube luminometers while also offering the flexibility of a multimode reader. The GloMax®-Multi Jr has a sensitivity of 1×10^{-18} moles of luciferase and >5 logs of dynamic range. This dynamic range is more than adequate to cover common luminescence applications, thus reducing the need to dilute samples.

The GloMax®-Multi Jr with a **Fluorescence Module** is designed to deliver both high performance and user flexibility. To achieve high performance, each Fluorescence Module utilizes powerful light-emitting diodes (LEDs) as excitation sources. LED usage increases sensitivity by fully exciting the fluorophore and reducing nonspecific light leakage, a problem often found when using broad-spectrum light sources. Four standard fluorescence optical kits are available for purchase, or contact us to purchase a custom optical kit.

- UV (Ex 365nm, Em 410–450nm)
- Blue (Ex 460nm, Em 515–570nm)
- Green (Ex 525nm, Em 580–640nm)
- Red (Ex 625nm, Em 660–725nm)

The GloMax®-Multi Jr with the **Absorbance Module** provides measurements that are highly sensitive and cover a wide dynamic range. The absorbance channel has a large reading range of 0–4 OD with an accuracy that deviates less than 0.7%.

The GloMax®-Multi Jr has three optional filter paddles with factory-installed filters for measuring 560, 600 and 750nm. These filters accommodate the most common protein assays. Filter paddles can be exchanged easily in seconds. In addition, custom filter paddles can be made readily for nonstandard applications. The GloMax®-Multi Jr is designed to be put into use right from the box without the need to read a manual or obtain special training. To achieve this plug-and-play usability, the GloMax®-Multi Jr. combines a color touch screen with an intuitive user interface. The interface makes running samples and viewing data fast and simple while also maintaining the flexibility needed for advanced or custom protocols. The GloMax®-Multi Jr. is a modular instrument that fits easily into most budgets. Purchase the technology or modes that you need now, and add on to the system later as your needs expand. For example, the GloMax®-Multi Jr can be purchased as a Luminometer. Then Fluorescence and/or Absorbance Modules can be purchased and added later. There's no service call or downtime. With the modular design, changing technologies is as easy as snapping in a module and restarting the instrument.

Features:

- **Flexible Configuration:** Modular system grows with your needs.
- **Touch Screen Interface:** The user interface has been designed to be intuitive so that no training is required to use the instrument.
- **Easy Protocol Setup:** Promega protocols are preloaded for easy implementation.
- **Convenient Data Handling:** Record data right from the instrument or export data to an Excel® spreadsheet.

Protocol	Part#
Technical Manual	TM339

GloMax®-Multi Jr Single-Tube Multimode Reader.

GloMax® 96 Microplate Luminometer

Product	Size	Cat.#
GloMax® 96 Microplate Luminometer	1 each	E6501
GloMax® 96 Microplate Luminometer w/ Single Injector	1 each	E6511
GloMax® 96 Microplate Luminometer w/ Dual Injectors	1 each	E6521
Available Separately	**Size**	**Cat.#**
GloMax® Luminometer Light Plate	1 each	E6531
GloMax® 96 Tubing Replacement Kit for Injectors	1 each	E6541
GloMax® Injector Tips Replacement	1 each	E5401
GloMax® 96 Base Instrument Service Agreement	1 each	SA3010
GloMax® Injectors Service Agreement, 1 year	1 each	SA3040

Request a quote.

Description: The GloMax® 96 Microplate Luminometer combines instrumentation and software in a complete solution that includes bioluminescent assays, protocols and support. The GloMax® 96 is a state-of-the-art microplate luminometer that meets the needs for high sensitivity and broad dynamic range for all luminescence applications. Available with up to two reagent injectors, the GloMax® 96 Microplate Luminometer is a versatile system designed to perform both flash- and glow-type luminescence assays. The GloMax® 96 Microplate Luminometer also includes a power cable, data cable, Quick Protocol card, 5 white 96-well microplates, and software required to operate the instrument. This instrument requires the use of a computer with Microsoft® Excel.

The GloMax® 96 Microplate Luminometer provides superior sensitivity and precision for all luminescent assays. Proprietary circuitry and an advanced photon-counting photomultiplier tube (PMT) provide unmatched signal-to-noise ratios. The option of an intelligently designed internal auto-injection system is an added convenience. Connections, priming and flushing are greatly simplified because up to two reagent injectors are designed to fit next to the plate detection module. This arrangement minimizes dispensing problems, simplifies maintenance and reduces service calls. The dispensing design also includes features that help the user save valuable time and reagents, including an open architecture that enables the user to inspect all tubing and tips during operation.

The software features preloaded protocols to run Promega assays. Setup wizards guide the user through a brief process when establishing new protocols. New users can set up protocols and operate the instrument without a steep learning curve. The user can quickly select the protocol of interest and begin running assays with a minimum of modification. Direct-to-Excel-based software reports data directly to an Excel spreadsheet, where data can be analyzed quickly and easily. An Excel macro assists in data analysis for Dual-Luciferase® assays.

The GloMax® 96 Microplate Luminometer Light Plate provides a quick and easy means to verify the performance of the GloMax® 96 Microplate Luminometer. Users can check the sensitivity, reproducibility and linearity. The Light Plate consists of three highly stable light sources that simulate luminescent samples at signal levels spanning four decades. The unit is powered by a battery that is widely available and easy to replace.

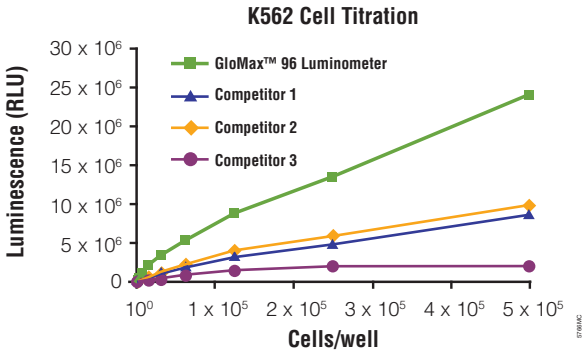
The GloMax® 96 Tubing Replacement Kit for Injectors contains parts for replacement of two complete fluid paths in the GloMax® 96 Microplate Luminometers configured with reagent injectors. Items contained within the kit include two sets of tubing and a pack of 10 injector tips.

Features:

- **Very Sensitive:** Sensitive to approximately 3×10^{-21} moles of luciferase.
- **Wide Dynamic Range:** 9-log dynamic range.
- Ideal for cell-based assays.
- Engineered to minimize sample cross-talk.
- **Convenient Data Handling:** Direct-to-Excel data importing requires Windows® PC to operate.
- **Simple Data Analysis:** Excel macros allow simple data analysis for Dual-Luciferase® Assays.

Protocol	Part#
Technical Manual	TM278

GloMax® 96 Microplate Luminometer.



The GloMax® 96 Microplate Luminometer demonstrates superior operating range compared to leading multifunction readers when using the CellTiter-Glo® Luminescent Cell Viability Assay.

GloMax® 20/20 Luminometer

Product	Size	Cat.#
GloMax® 20/20 Luminometer	1 each	E5311
GloMax® 20/20 Luminometer w/Single Auto-Injector	1 each	E5321
GloMax® 20/20 Luminometer w/Dual Auto-Injector	1 each	E5331
Available Separately	**Size**	**Cat.#**
GloMax® 20/20 Light Standard	1 each	E5341
GloMax® 20/20 Fluorescent Module, UV	1 each	E5351
GloMax® 20/20 Fluorescent Module, Blue	1 each	E5361
GloMax® 20/20 Test Tube Holder (1.5ml Microcentrifuge Tubes)	1 each	E5371
GloMax® 20/20 Replacement Tubing (2), Valves (4), Tips (30)	1 each	E5381
GloMax® 20/20 Replacement Valves	4 sets	E5391
GloMax® 20/20 Replacement Power Supply	1 each	E5411
Thermal Serial Printer and Universal Power Cable	1 each	E2821
Thermal Printer Paper	1 each	E2851
GloMax® 20/20 Base Instrument Service Agreement	1 each	SA3000
GloMax® Injectors Service Agreement, 1 year	1 each	SA3040

Request a quote.

Description: The GloMax® 20/20 Luminometer combines instrumentation and software in a complete solution that includes bioluminescent assays, protocols and support. The GloMax® 20/20 Luminometer is an ultrasensitive, versatile and affordable luminometer designed for use with any Promega bioluminescent assay. The touch screen interface provides comprehensive instrument control and data collection. Optional modules for fluorescence detection provide additional flexibility.

The option of an intelligently designed internal auto-injection system is an added convenience and meets the demands of the Dual-Luciferase® Assay. Software setup wizards guide the user through a brief process when establishing new protocols. New users can set up protocols and operate the instrument without a steep learning curve. Promega protocols are preloaded in the software to help users get started. The user can quickly select the protocol of interest and begin running assays directly to an Excel spreadsheet, where data can be analyzed quickly and easily.

Features:

- **Ultrasensitive:** Quantitate low-level luminescence samples with confidence.
- **Wide Dynamic Range:** Measure both dim and bright samples without sample dilution.
- **Easy Protocol Setup:** Promega protocols are preloaded for easy implementation.
- **Accessible Injector System:** Completely visible plumbing allows for inspection of tubing and tips.
- **Touch Screen Interface:** Simple to operate.
- **Convenient Data Handling:** Record data to a printer in real-time or export data to Excel.
- **Flexible:** Options available for up to two auto-injectors to meet your experimental needs.

Protocol	Part#
Technical Manual	TM276

GloMax® 20/20 Luminometer.

QuantiFluor™-ST and QuantiFluor™-P Single-Tube Fluorometers

Product	Size	Cat.#
QuantiFluor™-ST Handheld Fluorometer with UV/Blue Channels	1 each	E6090
QuantiFluor™-P Handheld Fluorometer with Green/Blue Channels	1 each	E6100
QuantiFluor™-P Handheld Fluorometer with UV/Blue Channels	1 each	E6105
Available Separately	**Size**	**Cat.#**
QuantiFluor™-ST Minicell Adapter Kit (for measuring 50–250l of sample)	400 each	E6112
QuantiFluor™-ST Solid Standard	1 each	E6113
QuantiFluor™-ST AC Adapter Replacement	1 each	E6096
QuantiFluor™-P Minicell Adapter Kit (for measuring 75–250l of sample)	400 each	E6111
Minicell Borosilicate Glass Cuvettes	400 each	E6091
10 × 10mm Square Polystyrene Cuvette (3.5ml capacity)	100 each	E6092
10 × 10mm Square Methacrylate Cuvette (3.5ml capacity)	100 each	E6093
Thermal Serial Printer and Universal Power Cable	1 each	E2821
Thermal Printer Paper	1 each	E2851
QuantiFluor™ Service Agreement	1 each	SA3060

Cat.# E6112 and E6111 include 400 borosilicate glass cuvettes.

Description: The QuantiFluor™-ST and QuantiFluor™-P Fluorometers are affordable, sensitive fluorometers designed for quick, easy and accurate fluorescence measurements. The QuantiFluor™ instruments provide high sensitivity for fluorescence measurements and are so easy to use that you can have results within minutes. Single-point calibration saves time, and the dual-channel design allows you to switch between two assays with the touch of a button. The QuantiFluor™-ST is a lab-ready instrument with two fluorescence channels built in: UV and Blue. The QuantiFluor™-P is a portable, battery-operated instrument with the option of either a UV/Blue configuration or Blue/Green configuration. Either instrument will have your lab running samples within a few minutes of opening the box.

Features:

- **Easy to Use:** Both QuantiFluor™ instruments are designed for fast, accurate fluorescence measurements. They are so easy to use that users will have results with a few button selections.
- **Small and Sensibly Priced:** The QuantiFluor™ instruments are smaller than a standard-sized book, saving bench space. They are sensibly priced to meet the needs for accurate fluorescence measurements on a limited budget.
- **Sample Volume Flexibility:** The QuantiFluor™ instruments can measure 2ml sample volumes or <250µl sample volumes with the optional minicell adapter.

Protocol	Part#
Technical Manual	TM338

QuantiFluor™-ST and QuantiFluor™-P Single-Tube Fluorometers.

β-Galactosidase Enzyme Assay System with Reporter Lysis Buffer

Product	Size	Cat.#
β-Galactosidase Enzyme Assay System with Reporter Lysis Buffer	10 ml	E2000
Available Separately	**Size**	**Cat.#**
Reporter Lysis 5X Buffer	30 ml	E3971

Cat.# E2000 contains sufficient reagents for 65 standard assays or 200 assays in a 96-well plate format.

Description: The β-Galactosidase Enzyme Assay System with Reporter Lysis Buffer is a convenient method for assaying β-galactosidase activity in lysates prepared from cells transfected with β-galactosidase reporter vectors such as the pSV–βGalactosidase Control Vector.

The standard assay is performed by adding a dilute sample to an equal volume of Assay 2X Buffer that contains the substrate ONPG (*o*-nitrophenyl–βD-galactopyranoside). Samples are incubated for at least 30 minutes, during which time the β-Galactosidase hydrolyzes the colorless substrate to *o*-nitrophenyl, which is yellow. The reaction may be terminated by addition of sodium carbonate, and the absorbance at 420nm is measured by spectrophotometry.

Features:

- **Safe:** Non-isotopic assay.
- **Versatile:** The assay can be used in a 96-well plate format.
- **Flexible:** Reporter Lysis Buffer allows firefly luciferase, CAT and β-galactosidase assays to be performed from the same cell extract.

Protocol	Part#
Technical Bulletin	TB097

Storage Conditions: Reporter Lysis Buffer may be stored at room temperature. Store other system components at –20°C.

Beta-Glo® Assay System

Product	Size	Cat.#
Beta-Glo® Assay System	10 ml	E4720
	100 ml	E4740
	10 × 100 ml	E4780

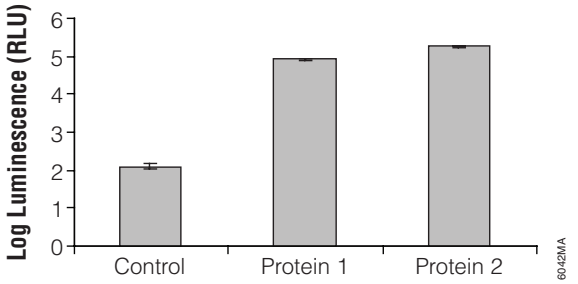
Description: The Beta-Glo® Assay System is a homogeneous method of quantitating -galactosidase expression in mammalian cells. The system provides a bright luminescent signal that is stable over several hours in commonly used cell culture medium without prior sample processing. The homogeneous assay procedure involves the addition of a single reagent directly to cells cultured in serum-supplemented medium. Throughput rates of several thousand samples per hour may be achieved with high reproducibility under standard laboratory conditions.

Features:

- **Bright Luminescent Signal:** Quantitate with confidence using low-volume formats or in samples with low-level expression.
- **Homogeneous Format:** Perform fewer steps. Add a single reagent directly to cells in growth medium.
- **Stable Signal:** Obtain flexibility and convenience when processing multiple plates.
- **Convenient:** Achieve optimal assay performance at room temperature.
- **Flexible:** Read the luminescent signal using any luminometer. Injectors are not required.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Manual	TM239

Storage Conditions: Store at –20°C.



-galactosidase activity determined using the Beta-Glo® Assay System with a yeast two-hybrid system. Please visit: **www.promega.com/enotes/** for more information.

Image kindly provided by Dr. Brad Hook, Ph.D., University of Wisconsin, Madison.

pSV–βGalactosidase Control Vector

Product	Size	Cat.#
pSV–βGalactosidase Control Vector	20 µg	E1081

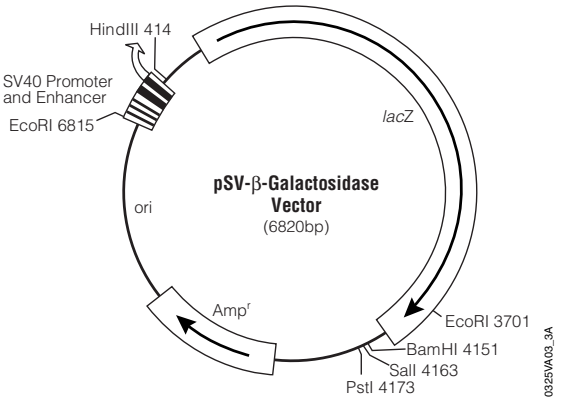
Description: The pSV–βGalactosidase Control Vector is a positive control vector for monitoring transfection efficiencies of mammalian cells. The SV40 early promoter and enhancer drive transcription of the *lac*Z gene, which encodes the -galactosidase enzyme. The pSV–βGalactosidase Control Vector can be transfected individually or co-transfected with your DNA of interest. β-galactosidase is an excellent reporter enzyme that can be assayed quickly and directly in cell extracts using spectrophotometric, fluorescent or chemiluminescent assays. This reporter enzyme is also widely used for in situ histochemical analysis using the substrate X-Gal.

The pSV–βGalactosidase Control Vector can be co-transfected with your DNA of interest. For example, co-transfection with firefly luciferase gene vectors (pGL3 Vectors) provide cell extracts that can be assayed for both luciferase and -galactosidase activities. In this manner, the pSV–βGalactosidase Vector acts as an internal control for transient expression assays. A negative control extract, prepared from mock-transfected cells, should also be assayed for the presence of endogenous β-galactosidase activity in cultured cells. In addition, co-transfection with chloramphenicol acetyltransferase reporter gene vectors (pCAT®3 Vectors) permits assaying for both CAT and β-galactosidase activities.

The pSV–βGalactosidase Vector is a modification of pRSV–βGal with SV40 and pUC18 sequences substituted for RSV and pBR322 sequences. The pSV–βGalactosidase Vector will express -galactosidase in *E. coli* due to the presence of the *E. coli* gpt promoter located upstream of the *lac*Z gene. Colonies of *E. coli* containing the pSV–βGalactosidase Vector will appear blue when plated on media containing X-Gal.

Protocol	Part#
Technical Bulletin	TB094

Storage Conditions: Store at –20°C.



CAT Enzyme Assay System

Product	Size	Cat.#
CAT Enzyme Assay System	50 reactions	E1000
Available Separately	**Size**	**Cat.#**
Reporter Lysis 5X Buffer	30 ml	E3971

Description: Chloramphenicol acetyltransferase (CAT), encoded by a bacterial drug-resistance gene, inactivates chloramphenicol by acetylating the drug at one or both of its two hydroxyl groups. This gene is not found in eukaryotes, and therefore eukaryotic cells contain no background of CAT activity. The CAT Enzyme Assay System offers two alternative methods for monitoring CAT enzyme activity in transfected cells: liquid scintillation counting (LSC) and thin layer chromatography (TLC). Either the LSC or TLC assays can be performed using the same cell extract. The TLC-based assay is less sensitive and more time-consuming to perform than the LSC assay but is useful as a visual confirmation of assay results. The resolved TLC reaction products are detected by autoradiography or phosphorimaging analysis.

Features:

- **Fast:** The assay is performed in as little as 2–3 hours.
- **Linear:** The LSC assay is linear for three orders of magnitude of enzyme activity.
- **Sensitive:** As little as 3×10^{-4} units (2pg) of CAT can be detected.
- **Robust:** Reporter Lysis Buffer allows luciferase, CAT and -galactosidase assays to be performed from the same cell extract.

Protocol	Part#
Technical Bulletin	TB084

Storage Conditions: Reporter Lysis 5X Buffer may be stored at room temperature. Store other system components at –20°C.

Chloramphenicol Acetyltransferase (CAT)

Product	Size	Conc.	Cat.#
Chloramphenicol Acetyltransferase	100 u	10–14 u/µl	E1051

Description: Chloramphenicol Acetyltransferase (CAT) catalyzes the transfer of an acetyl group from acetyl-CoA to the 3´-hydroxy position of chloramphenicol. The enzyme is suitable as a standard in CAT assays of crude cell extracts.

Storage Conditions: Store at –20°C.

n-Butyryl CoA

Product	Size	Conc.	Cat.#
n-Butyryl CoA	255 l	5 mg/ml	E1061

Description: n-Butyryl CoA is suitable for use in the chloramphenicol acetyltransferase (CAT) reaction. Transfer of the n-butyryl moiety to chloramphenicol by the CAT enzyme allows enzyme activity to be monitored using liquid scintillation counting or thin layer chromatography formats.

Storage Conditions: Store at –20°C. Avoid multiple freeze-thaw cycles.

pCAT®3 Vectors

Product	Size	Cat.#
pCAT®3-Control Vector	20 µg	E1851
pCAT®3-Promoter Vector	20 µg	E1861
pCAT®3-Basic Vector	20 µg	E1871
pCAT®3-Enhancer Vector	20 µg	E1881

Description: The pCAT®3 Reporter Vectors provide a basis for the quantitative analysis of factors that may regulate mammalian gene expression. The redesigned backbone of the pCAT®3 Reporter Vectors is similar to the pGL3 Luciferase Vectors with the exception of a chimeric intron located 5´ of the chloramphenicol acetyltransferase (CAT) gene. As with the pGL3 Vectors, the pCAT®3 Vectors contain a different polyadenylation site located 3´ of the gene. The redesigned backbone increases expression of the reporter gene, improves in vivo vector stability and provides greater flexibility in performing manipulations.

Features:

- **Efficient:** Optimal translation efficiency.
- **Robust:** Increased expression with more efficient poly(A) signal.
- **Clearer Results:** Reduced background CAT expression.
- **Compatible:** Altered multiple cloning regions make vectors compatible with the pGL3 Vectors.
- **Versatile:** Can produce ssDNA for sequencing and mutagenesis.

Protocol	Part#
Technical Manual	TM036

Storage Conditions: Store vectors at –20°C.

Monster Green® Fluorescent Protein phMGFP Vector

Product	Size	Cat.#
Monster Green® Fluorescent Protein phMGFP Vector	20 g	E6421

Description: The phMGFP Vector contains the open reading frame for the Monster Green® Fluorescent Protein cloned into a mammalian expression vector. The Monster Green® Fluorescent Protein is encoded by an improved synthetic version of the green fluorescent protein gene originally cloned from *Montastrea cavernosa* (Great Star Coral). The synthetic gene (hMGFP) expresses a 26kDa protein that shows improved fluorescence intensity compared to the native gene. Furthermore, the hMGFP gene has been codon optimized and cleared of most consensus sequence transcription factor binding sites to ensure reliability and high levels of expression.

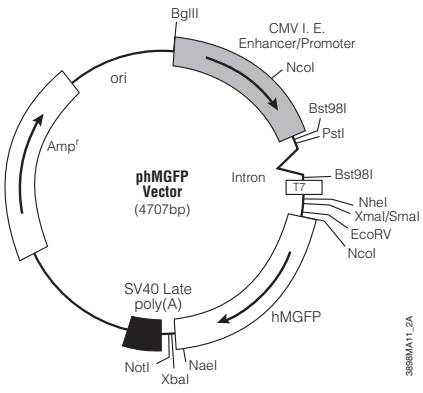
The Monster Green® Fluorescent Protein encoded by the hMGFP gene is an ideal fluorescent reporter, providing high-level fluorescence and reducing cytotoxicity. Monster Green® Fluorescent Protein generally fluoresces at least 20% brighter than other commercially available green fluorescent proteins (GFPs) and also reduces cytotoxicity, offering flexibility when working with transient and stable expression assays.

Features:

- **Brighter Fluorescence:** Visualize low-level expression in situ using fluorescence microscopy, imagers or FACS®.
- **Reduced Cytotoxicity:** Minimize cellular perturbations when working with transient or stable expression assays.
- **Flexible:** Create fusion proteins for imaging and localization studies using standard FITC detection.
- **High Purity:** Obtain high transfection efficiencies for precloning confirmation studies.

Protocol	Part#
Technical Bulletin	TB320

Storage Conditions: Store at –20°C.



FuGENE® HD Transfection Reagent

Product	Size	Cat.#
FuGENE® HD Transfection Reagent	1 ml	E2311
	5 × 1 ml	E2312

Description: FuGENE® HD Transfection Reagent is a novel, nonliposomal formulation designed to transfect DNA into a wide variety of cell lines with high efficiency and low toxicity. The protocol does not require removal of serum or culture medium and does not require washing or changing of medium after introducing the reagent/DNA complex. Additionally, the FuGENE® HD Transfection Reagent has been shown to support transfection in chemically defined media and does not contain any animal-derived components.

The cell lines listed in Table 1 have been transfected successfully by Promega Corporation or Fugent, L.L.C. For a list of conditions that were used in the transfection of these and other cell types, visit our FuGENE® HD Protocol Database at: **www.promega.com/techserv/tools/FugeneHD/**

® HD Transfection Reagent by Promega Corporation or Fugent, L.L.C. (® *HD Transfection Reagent by Promega Corporation or Fugent, L.L.C.*)

Features:

- **More Biologically Relevant:** Low toxicity, less impact on biology.
- **Simple Protocol:** No culture changes, less variability, compatible with serum.
- **Effective in Many Cell Types:** Online database with over 40 cell types, including primary and stem cells.
- **Ideal for Use with Luciferase Assays:** More expression, sensitive results.

Protocol	Part#
Technical Manual	TM328
Quick Protocol Card	FB112

Storage Conditions: Store FuGENE® HD Transfection Reagent at 4°C.

Cell lines successfully transfected using the FuGENE® HD Transfection Reagent by Promega Corporation or Fugent, L.L.C.

NIH3T3	U-937
HEK293	STSAR90
CHO-K1	AGS
CHO-S	BHK-21
SNU-16	Caco-2
A-375	Caki-1
T98G	Capan-1
HeLa	H4
HepG2	Human skeletal muscle myoblasts (HSMM)
High Five™	NCI-N87
MCF7	Panc-1
mES	SK MEL-28
hES	SK-OV-3
PC3	T-24
RAW 264.7	T-84
SCC61	U-87 MG
SQ20B	A549
STO	DMS 53
U-2 OS	T47D
COS-7	Jurkat
293F	Huh7

8684LA

TransFast™ Transfection Reagent

Product	Size	Cat.#
TransFast™ Transfection Reagent	1.2 mg	E2431

Description: The TransFast™ Transfection Reagent is composed of the synthetic cationic lipid, (+)-N,N [bis (2-hydroxyethyl)]-N-methyl-N-[2,3-di(tetradecanoyloxy)propyl] ammonium iodide and the neutral lipid, DOPE. The TransFast™ Reagent is supplied as a dried lipid film that forms multilamellar vesicles upon hydration with water. Cationic liposomes designed for transfection, such as the TransFast™ Reagent, are more versatile than many other traditional transfection methods. The advantages include flexibility in the macromolecules that are delivered, in vitro and in vivo applications, ability to more reproducibly transfect cells that are recalcitrant to other methods and suitability for transient and stable transfection. Several different types of macromolecules, including RNA and DNA of all sizes ranging from oligonucleotides to plasmids and yeast artificial chromosomes, can be delivered to cells using liposomes. The TransFast™ Transfection Reagent is designed for nucleic acid delivery to eukaryotic cells in vitro and in vivo and performs well with many cell lines. To date, we have found that TransFast™ Reagent performs particularly well for DNA delivery to NIH/3T3, CHO, 293, K562, PC12, Jurkat and insect Sf9 cells.

Features:

- **Fast:** Transfect in 1 hour. Transfection times can be decreased to as little as 30 minutes with certain cell lines.
- **Easy to Use:** Resuspend the reagent in water, freeze, thaw, mix with DNA, and add to cells.
- **Efficient:** High-efficiency transfection—transient and stable—in many cells.
- **Robust:** Requires less optimization than other systems. Allows transfection of cell types such as primary cell cultures that require continuous exposure to serum.

Protocol	Part#
Technical Bulletin	TB260

Storage Conditions: Store at –20°C.

ProFection® Mammalian Transfection System

Product	Size	Cat.#
ProFection® Mammalian Transfection System—Calcium Phosphate	40 reactions	E1200

Description: The introduction of DNA into mammalian cells is facilitated by the ProFection® Mammalian Transfection System. This system offers you a Calcium Phosphate-mediated transfection procedure. Each system contains sufficient reagents for 40 high-efficiency transfections of cells plated in 100mm tissue culture dishes.

Calcium phosphate transfection is an effective method for the production of long-term stable transfectants. This method also works well for transient expression of transfected genes and can be used with most adherent cell lines.

Features:

- **Efficient:** Components optimized for high transfection efficiencies.

Protocol	Part#
Technical Manual	TM012

Storage Conditions: Store at –20°C.

Imaging and Immunological Detection

Cellular Imaging	**164**
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Phosphorylation-Specific Protein Kinase Antibodies	**169**
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Antibody Conjugates	**180**
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HaloTag® Technology

Product	Size	Conc.	Cat.#	
HaloTag® Protein Purification System	1 each		G6280	489
HaloTag® Protein Purification System Sample Pack	1 each		G6270	165
HaloLink™ Resin	2ml (0.5ml settled resin)		G1911	72
	5ml (1.25ml settled resin)		G1912	159
HaloLink™ Magnetic Beads	40 reactions		G9311	84
HaloTag® TMR Ligand	30 µl	5 mM	G8251	513
	15 µl	5 mM	G8252	249
HaloTag® diAcFAM Ligand	30 µl	1 mM	G8272	513
	15 µl	1 mM	G8273	249
HaloTag® Coumarin Ligand	30 µl	10 mM	G8581	499
	15 µl	10 mM	G8582	249
HaloTag® Alexa Fluor® 488 Ligand	30 µl	1 mM	G1001	513
	15 µl	1 mM	G1002	249
HaloTag® Oregon Green® Ligand	30 µl	1 mM	G2801	513
	15 µl	1 mM	G2802	249
HaloTag® TMRDirect™ Ligand	30 µl	0.1 mM	G2991	294
HaloTag® R110Direct™ Ligand	30 µl	0.1 mM	G3221	294
HaloTag® Biotin Ligand	30 µl	5 mM	G8281	513
	15 µl	5 mM	G8282	249
HaloTag® PEG-Biotin Ligand	30 µl	5 mM	G8591	499
	15 µl	5 mM	G8592	249
HaloLink™ Array (TNT® T7 Quick) Two Slide System	two 50-well arrays		G6140	367
HaloLink™ Array (TNT® SP6 Wheat Germ) Two Slide System	two 50-well arrays		G6180	367
HaloLink™ Array Six Slide System	6 slides		G6190	735
HaloTag® Standard Protein	30 µg		G4491	50
HaloCHIP™ System	20 reactions		G9410	509
HaloTag® Amine (O4) Ligand	5 mg		P6741	499
HaloTag® Succinimidyl Ester (O4) Ligand	5 mg		P6751	499
HaloTag® Thiol (O4) Ligand	5 mg		P6761	499
HaloTag® Iodoacetamide (O4) Ligand	5 mg		P6771	499
HaloTag® Succinimidyl Ester (O2) Ligand	5 mg		P1691	499
HaloTag® Amine (O2) Ligand	5 mg		P6711	499
HaloTag® Iodoacetamide (O2) Ligand	5 mg		P1681	499
Anti-HaloTag® pAb	200 µg	1 mg/ml	G9281	236
pFC14A HaloTag® CMV Flexi® Vector	20 µg		G9651	193
pFC14K HaloTag® CMV Flexi® Vector	20 µg		G9661	193
pFC15A HaloTag® CMV*d1* Flexi® Vector	20 µg		G1611	193
pFC15K HaloTag® CMV*d1* Flexi® Vector	20 µg		G1601	193
pFC16A HaloTag® CMV*d2* Flexi® Vector	20 µg		G1591	193
pFC16K HaloTag® CMV*d2* Flexi® Vector	20 µg		G1571	193
pFC17A HaloTag® CMV*d3* Flexi® Vector	20 µg		G1551	193
pFC17K HaloTag® CMV*d3* Flexi® Vector	20 µg		G1321	193
pFN21A HaloTag® CMV Flexi® Vector	20 µg		G2821	193
pFN21K HaloTag® CMV Flexi® Vector	20 µg		G2831	193
pFN22A HaloTag® CMV*d1* Flexi® Vector	20 µg		G2841	193
pFN22K HaloTag® CMV*d1* Flexi® Vector	20 µg		G2851	193
pFN23A HaloTag® CMV*d2* Flexi® Vector	20 µg		G2861	193
pFN23K HaloTag® CMV*d2* Flexi® Vector	20 µg		G2871	193
pFN24A HaloTag® CMV*d3* Flexi® Vector	20 µg		G2881	193
pFN24K HaloTag® CMV*d3* Flexi® Vector	20 µg		G2981	193
HaloTag® Flexi® Vectors—CMV Deletion Series Sample Pack	9 × 2 µg		G3780	193
pFN18A HaloTag® T7 Flexi® Vector	20 µg		G2751	193
pFN18K HaloTag® T7 Flexi® Vector	20 µg		G2681	193
pFN19A HaloTag® T7 SP6 Flexi® Vector	20 µg		G1891	193
pFN19K HaloTag® T7 SP6 Flexi® Vector	20 µg		G1841	193
pFC20A HaloTag® T7 SP6 Flexi® Vector	20 µg		G1681	193
pFC20K HaloTag® T7 SP6 Flexi® Vector	20 µg		G1691	193
HaloTag® Cloning Starter System	1 each		G6050	

Description: The HaloTag® Technology is a platform technology for enabling covalent protein labeling and immobilization in vivo and in vitro. Products based on the HaloTag® Technology enable researchers to study protein function in a biochemical and cellular environment. The technology is based on the efficient formation of a covalent bond between a specially designed reporter protein encoded by a HaloTag® Vector and a specific ligand in living cells, in solution or on a solid support.

For information about the HaloTag® Protein Purification Systems, please visit the HaloTag® Protein Purification Systems online catalog page.

HaloTag® Ligands for Protein Immobilization

The HaloLink™ Resin (Cat.# G1911) is a solid support that allows covalent and oriented attachment of HaloTag® fusion proteins to a Sepharose® surface. Due to covalent linkage, HaloTag® fusion proteins cannot be eluted from the resin, allowing extensive washing to remove nonspecifically bound protein without the danger of eluting HaloTag® fusion proteins. The binding rate is very rapid and equivalent to biotin-streptavidin.

HaloLink™ Magnetic Beads (Cat.# G9311) provide a rapid and reliable method to covalently capture and immobilize HaloTag® fusion proteins to a paramagnetic particle. Immobilization through the HaloTag® Ligand provides for consistent, surface-directed orientation of the fusion protein.

HaloTag® Fluorescent Ligands for Cellular Imaging

(Please see *HaloTag® Technology: Focus on Imaging Technical Manual* TM260 for more information about the HaloTag® fluorescent ligands.)

- Cell-permeant fluorescent ligands (rapid labeling protocol):
  - HaloTag® TMR Ligand ($555_{Ex}/585_{Em}$), Cat.# G8251.
  - HaloTag® Oregon Green® Ligand ($496_{Ex}/516_{Em}$), Cat.# G2801.
  - HaloTag® diAcFAM Ligand ($494_{Ex}/526_{Em}$), Cat.# G8271.
  - HaloTag® Coumarin Ligand ($353_{Ex}/434_{Em}$), Cat.# G8581.
- Cell-impermeant fluorescent ligands for cell-surface labeling (rapid labeling protocol):
  - HaloTag® Alexa Fluor® 488 Ligand ($494_{Ex}/517_{Em}$), Cat.# G1001.
- Cell-permeant fluorescent ligands ("no wash" protocol):
  - HaloTag® TMRDirect™ Ligand ($555_{Ex}/585_{Em}$), Cat.# G2991.
  - HaloTag® R110Direct™ Ligand ($502_{Ex}/527_{Em}$), Cat.# G3221.

HaloTag® Ligands for Protein Detection

The HaloTag® Biotin Ligand (Cat.# G8281) consists of a 12-atom linker arm to biotin and is used as an affinity tag to capture the HaloTag® protein-based fusion construct using the strong biotin-streptavidin interaction.

The HaloTag® PEG-Biotin Ligand (Cat.# G8591) contains a spacer not found in the HaloTag® Biotin Ligand. This provides a significantly longer and more flexible linker between streptavidin and the HaloTag® protein, which may be advantageous in preserving the activity of a HaloTag® fusion partner protein upon immobilization or derivatization.

For information about the HaloLink™ Protein Array Systems, please visit the HaloLink™ Protein Array Systems online catalog page.

For information about the HaloCHIP™ System, please visit the HaloCHIP™ System online catalog page.

HaloTag® Vectors

The HaloTag® Vectors contain the open reading frame (ORF) for HaloTag® protein (33kDa monomeric protein), a genetically engineered derivative of a hydrolase gene. HaloTag® protein is not endogenous to mammalian cells, allowing high labeling specificity.

HaloTag® Flexi® Vectors

A variety of HaloTag® Flexi® Vectors for mammalian, *E. coli* and cell-free protein expression are available. These vectors are compatible with the Flexi® Vector System; therefore, the protein-coding sequence can be transferred easily to Flexi® Vectors that contain various expression or peptide tag options to enable downstream applications.

HaloTag® Flexi® Vectors for Mammalian Protein Expression
The pFC14, pFC15, pFC16 and pFC17 HaloTag® CMV Flexi® Vectors are designed for the expression of carboxy-terminal HaloTag® fusion proteins primarily in mammalian cells.

HaloTag® Flexi® Vectors for E. coli *Protein Expression*
The pFN18 HaloTag® T7 Flexi® Vectors are designed for the optimal expression of amino-terminal HaloTag® fusion proteins primarily in *E. coli*.

HaloTag® Flexi® Vectors for Cell-Free Protein Expression
The pFN19 HaloTag® T7 SP6 Flexi® Vectors are designed for the optimal expression of amino-terminal HaloTag® fusion proteins primarily in cell-free translation systems. The pFC20 HaloTag® T7 SP6 Flexi® Vectors are designed for the expression of carboxy-terminal HaloTag® fusion proteins primarily in cell-free translation systems.

For more information about the HaloTag® Flexi® Vectors, please visit the HaloTag® Flexi® Vectors online catalog page.

Note: pHT2 Vector (Cat.# G8241) has been discontinued and is not recommended for expression in bacterial systems.

Difference between HaloTag® protein in HaloTag® Flexi® Vectors and discontinued pHT2 Vector

The HaloTag® protein contained in the HaloTag® Flexi® Vectors is HaloTag® 7, which provides increased stability with regard to both temperature and denaturants, increased stability and faster labeling kinetics, resulting in markedly improved expression compared to the HaloTag® 2 protein contained in pHT2 HaloTag® Vector.

HaloTag® Cloning Starter System

The HaloTag® Cloning Starter System contains C8641, Flexi® System, Entry/Transfer; A9280, Wizard® SV Gel and PCR Clean-Up System; R1901, Carboxy Flexi® Enzyme Blend; and G3780, HaloTag® Flexi® Vectors—CMV Deletion Series Sample Pack.

For information about the HaloTag® Ligand Building Blocks, please visit the HaloTag® Ligand Building Blocks online catalog page.

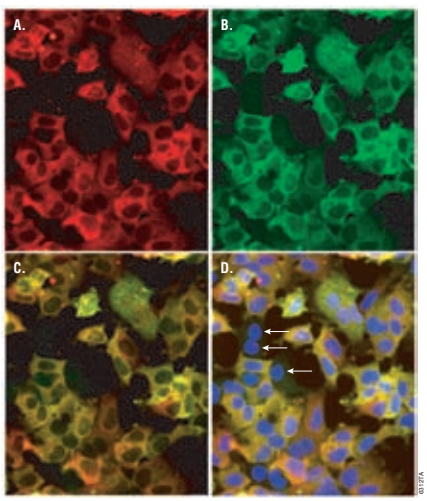
HaloTag® Polyclonal Ab

A polyclonal antibody to the HaloTag® reporter protein, Anti-HaloTag® pAb (Cat.# G9281) is available to provide an additional tool for detection of HaloTag® fusion proteins by traditional Western blotting or in situ labeling by ICC. The pAb may be used as the sole means of detecting HaloTag® fusion proteins or to confirm the specific labeling achieved by HaloTag® ligands in vivo.

Features:

- **Label in Solution or on a Solid Support:** The HaloTag® Ligands bind to the HaloTag® protein or protein fusions with high specificity and affinity.
- **Label Your HaloTag® Protein in Live Cells:** The HaloTag® TMR, diAcFAM, Coumarin and Biotin Ligands readily cross the cell membrane.
- **Pull Down Protein Complexes:** The spacer and reactive linker of the HaloTag® PEG-Biotin Ligand provide ideal pull-down capabilities. Alternatively, pull down directly with the HaloLink™ Resin.
- **Image Fixed Cells:** The covalent bond is stable, allowing imaging of fixed cells and analysis of the labeled protein under stringent conditions.
- **Introduce Novel Functionalities or Perform Sequential Labeling:** The open architecture of the technology enables the use of different ligands for multiple applications.
- **Design Only One Genetic Construct for Multiple Experiments:** Obtain new functionality by using a different HaloTag® Ligand without having to design and clone a new expression construct.
- **Analyze Labeled Fusion Proteins Using SDS-PAGE, Mass Spectrometry, etc.:** The bound ligand is stable under denaturing conditions.

Protocol	Part#
HaloTag® Technology: Focus on Imaging Technical Manual	TM260
HaloLink™ Resin Technical Manual	TM250
HaloCHIP™ System Technical Manual	TM075
Flexi® Vector Systems Technical Manual	TM254
HaloLink™ Magnetic Beads Technical Manual	TM291
HaloLink™ Protein Array Systems Technical Manual	TM310
HaloTag® Protein Purification Systems Technical Manual	TM312



Colabeling of HaloTag®-p65 fusion protein with HaloTag® TMR Ligand and the Anti-HaloTag® pAb. **Panel A.** Cytoplasmic (red) labeling of HEK293-p65-HT2 cells by HaloTag® TMR Ligand. **Panel B.** Cytoplasmic (green) labeling by Anti-HaloTag® pAb and Alexa Fluor® 488-conjugated anti-rabbit IgG (Invitrogen). **Panel C.** Colocalization of ligand and antibody binding activities. **Panel D.** Merger of red and green fluorescence with counterstaining of the nucleus by DAPI (blue). Arrows denote rare cells that show little or no expression of HaloTag®-p65. Protocols developed and performed at Promega.

Section Contents
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HaloTag® Flexi® Vectors

Product	Size	Cat.#
pFC14A HaloTag® CMV Flexi® Vector	20 µg	G9651
pFC14K HaloTag® CMV Flexi® Vector	20 µg	G9661
pFC15A HaloTag® CMV*d1* Flexi® Vector	20 µg	G1611
pFC15K HaloTag® CMV*d1* Flexi® Vector	20 µg	G1601
pFC16A HaloTag® CMV*d2* Flexi® Vector	20 µg	G1591
pFC16K HaloTag® CMV*d2* Flexi® Vector	20 µg	G1571
pFC17A HaloTag® CMV*d3* Flexi® Vector	20 µg	G1551
pFC17K HaloTag® CMV*d3* Flexi® Vector	20 µg	G1321
pFN21A HaloTag® CMV Flexi® Vector	20 µg	G2821
pFN21K HaloTag® CMV Flexi® Vector	20 µg	G2831
pFN22A HaloTag® CMV*d1* Flexi® Vector	20 µg	G2841
pFN22K HaloTag® CMV*d1* Flexi® Vector	20 µg	G2851
pFN23A HaloTag® CMV*d2* Flexi® Vector	20 µg	G2861
pFN23K HaloTag® CMV*d2* Flexi® Vector	20 µg	G2871
pFN24A HaloTag® CMV*d3* Flexi® Vector	20 µg	G2881
pFN24K HaloTag® CMV*d3* Flexi® Vector	20 µg	G2981
HaloTag® Flexi® Vectors—CMV Deletion Series Sample Pack	9 × 2 µg	G3780
pFN18A HaloTag® T7 Flexi® Vector	20 µg	G2751
pFN18K HaloTag® T7 Flexi® Vector	20 µg	G2681
pFN19A HaloTag® T7 SP6 Flexi® Vector	20 µg	G1891
pFN19K HaloTag® T7 SP6 Flexi® Vector	20 µg	G1841
pFC20A HaloTag® T7 SP6 Flexi® Vector	20 µg	G1681
pFC20K HaloTag® T7 SP6 Flexi® Vector	20 µg	G1691

Available Separately	Size	Cat.#	
HaloTag® Cloning Starter System	1 each	G6050	350
Flexi® System, Entry/Transfer	5 entry and 20 transfer reactions	C8640	223
Flexi® System, Transfer	100 transfer reactions	C8820	891
Carboxy Flexi® System, Transfer	50 transfer reactions	C9320	391
10X Flexi® Enzyme Blend (SgfI & PmeI)	25 µl	R1851	85
	100 µl	R1852	310
Carboxy Flexi® Enzyme Blend (SgfI & EcoICRI)	50 µl	R1901	84

Description: A variety of HaloTag® Flexi® Vectors for mammalian, *E. coli* and cell-free protein expression are available. These vectors are compatible with the Flexi® Vector System, a directional cloning method for protein-coding sequences. The Flexi® Vector System provides a rapid, efficient and high-fidelity way to transfer these sequences between a variety of Flexi® Vectors, which contain various expression or peptide tag options to enable expression of native or fusion proteins to study protein structure and function as well as protein:protein interactions.

HaloTag® Flexi® Vectors for Mammalian Protein Expression

The vectors listed in Table 1 are best for mammalian protein expression. All vectors designated "pFC" are configured to append HaloTag® protein to the carboxy-terminus of the protein fusion partner. All vectors designated "pFN" are configured to append HaloTag® protein to the amino-terminus of the protein fusion partner. The vectors contain either a full-length (pFC14, pFN21) or modified (pFC15, pFC16, pFC17, pFN22, pFN23, pFN24) human cytomegalovirus (CMV) promoter. The modified CMV promoter, which contains various deletions, provides varying levels of protein expression levels, depending on the protein of interest and cell type. Please see the table on the next page for general guidelines regarding protein expression levels.

Relative Mammalian Protein Expression Levels for HaloTag® Flexi® Vectors.

Vector Name	Cat.#	Expression Level*
pFC14A HaloTag® CMV Flexi® Vector	G9651	High
pFC14K HaloTag® CMV Flexi® Vector	G9661	High
pFC15A HaloTag® CMV*d1* Flexi® Vector	G1611	Medium
pFC15K HaloTag® CMV*d1* Flexi® Vector	G1601	Medium
pFC16A HaloTag® CMV*d2* Flexi® Vector	G1591	Low
pFC16K HaloTag® CMV*d2* Flexi® Vector	G1571	Low
pFC17A HaloTag® CMV*d3* Flexi® Vector	G1551	Ultra-Low
pFC17K HaloTag® CMV*d3* Flexi® Vector	G1321	Ultra-Low
pFN21A HaloTag® CMV Flexi® Vector	G2821	High
pFN21K HaloTag® CMV Flexi® Vector	G2831	High
pFN22A HaloTag® CMVd1 Flexi® Vector	G2841	Medium
pFN22K HaloTag® CMVd1 Flexi® Vector	G2851	Medium
pFN23A HaloTag® CMV*d2* Flexi® Vector	G2861	Low
pFN23K HaloTag® CMV*d2* Flexi® Vector	G2871	Low
pFN24A HaloTag® CMV*d3* Flexi® Vector	G2881	Ultra-Low
pFN24K HaloTag® CMV*d3* Flexi® Vector	G2981	Ultra-Low

*Expression level depends on the cell type and the protein fused to HaloTag® protein.

HaloTag® Flexi® Vectors for E. coli *and Cell-Free Protein Expression*

The pFN18 HaloTag® T7 Flexi® Vectors are configured to append the HaloTag® protein to the amino-terminus of the protein fusion partner and provide T7 RNA polymerase-driven protein expression in *E. coli* or in cell-free translation systems.

The pFN19 HaloTag® T7 SP6 Flexi® Vectors are configured to append the HaloTag® protein to the amino-terminus of the protein fusion partner and provide T7 RNA polymerase-driven protein expression in *E. coli* (approximately two- to fourfold lower protein expression compared to pFN18 vectors) or T7 or SP6 RNA polymerase-driven protein expression in cell-free translation systems.

The pFC20 HaloTag® T7 SP6 Flexi® Vectors are configured to append the HaloTag® protein to the carboxy-terminus of the protein fusion partner and provide T7 RNA polymerase-driven protein expression in *E. coli* or T7 or SP6 RNA polymerase-driven protein expression in cell-free translation systems.

Note: pHT2 Vector (Cat.# G8241) has been discontinued and is not recommended for expression in bacterial systems.

Difference between HaloTag® protein in HaloTag® Flexi® Vectors and discontinued pHT2 Vector.

The HaloTag® protein contained in the HaloTag® Flexi® Vectors contains HaloTag® 7, which provides increased stability with regard to both temperature and denaturants, increased stability and faster labeling kinetics, resulting in markedly improved expression compared to the HaloTag® 2 protein contained in pHT2 HaloTag® Vector.

Features:

- **Flexibility:** Choose between a variety of mammalian expression vectors to determine the vector that provides optimal protein expression for your application.
- **Time Savings:** Efficient transfer allows for direct use of recombinant clones, minimizing time wasted screening background colonies.
- **Versatility:** Express HaloTag® fusion protein in either mammalian, *E. coli* or cell-free systems.

Protocol	Part#
Flexi® Vector Systems Technical Manual	TM254
HaloTag® Technology: Focus on Imaging Technical Manual	TM260
HaloCHIP™ System Technical Manual	TM075
HaloLink™ Resin Technical Manual	TM250

Outline of Protocol for the Preparation of the HaloTag® Fusion Protein Using C-Terminal HaloTag® Flexi® Vectors

(Please see the *Flexi® Vector Systems Technical Manual* TM254 for a detailed protocol.)

Additional materials needed:

- Flexi® System, Entry/Transfer (Cat.# C8640)
- Carboxy Flexi® Enzyme Blend (SgfI and EcoICRI) (Cat.# R1901)
- N-terminal Flexi® Vector (e.g., pFN21A, pFN21K)

Note: Fusions to C-terminal tags are no longer functional for shuttling to other expression vectors To retain the capacity to transfer a protein coding sequence to multiple vectors, the protein-coding sequence must first be cloned into a Flexi ® vector with no tag or an N-terminal tag prior to transferring the protein-coding sequence to the C-terminal Flexi® Vector.

Entry Reaction

1 Design primers for the desired protein-coding region using the Flexi® Vector Primer Design Tool.
2 Amplify with a high-fidelity DNA polymerase, and purify amplification reaction.
3 Cut amplified DNA with SgfI and PmeI, clean-up digest.
4 Cut the acceptor Flexi® Vector (N-terminal Flexi® Vector) SgfI and PmeI.
5 Heat the reaction to inactivate restriction enzymes; ligate PCR product and acceptor Flexi® Vector.
6 Transform and plate on LB plates supplemented with ampicillin or kanamycin.
7 Screen colonies; sequence validate insert.

Outline of Protocol for the Preparation of the HaloTag® Fusion Protein Using N-Terminal HaloTag® Flexi® Vectors

(Please see the *Flexi® Vector Systems Technical Manual* TM254 for a detailed protocol.)

Additional materials needed:

- Flexi® System, Entry/Transfer (Cat.# C8640).

Follow the Entry Reaction protocol above.

Storage Conditions: Store vectors at –20°C.

BDNF E_{max}® ImmunoAssay Systems

Product	Size	Cat.#
BDNF E_{max}® ImmunoAssay System	2 × 96 wells	G7610
	5 × 96 wells	G7611

Description: The BDNF E_{max}® ImmunoAssay Systems provide optimized reagents and a protocol for the sensitive and specific detection of brain-derived neurotrophic factor (BDNF). After an overnight coating of a 96-well plate, the specific protein is detected using an antibody sandwich format. The systems use a horseradish peroxidase-conjugated secondary antibody and a single-component TMB substrate for the final chromogenic detection of bound neurotrophic factor. Using this system, BDNF in tissue culture supernatants, tissue homogenates, plasma and urine can be quantitated in the range of 7.8–500pg/ml. Binding and recovery from mouse brain homogenates has not been fully characterized.

Features:

- **High Value:** Optimized reagents and protocol provided.
- **Specificity:** Specific detection of BDNF; less than 3% cross-reactivity with other related neurotrophic and growth factors.
- **Sensitivity:** Detect picogram levels of factor per milliliter of sample.
- **Flexibility:** Available in sizes for two or five 96-well plates; can configure plates as desired.

Protocol	Part#
Technical Bulletin	TB257

Storage Conditions: Store the entire system in its original package protected from light at –20°C.

GDNF E_{max}® ImmunoAssay Systems

Product	Size	Cat.#
GDNF E_{max}® ImmunoAssay System	2 × 96 wells	G7620
	5 × 96 wells	G7621

Description: The GDNF E_{max}® ImmunoAssay Systems provide optimized reagents and a protocol for the sensitive and specific detection of glial cell-line-derived neurotrophic factor (GDNF). After an overnight coating of a 96-well plate, the specific protein is detected using an antibody sandwich format. The systems use horseradish peroxidase-conjugated secondary antibody and a single-component TMB substrate for the final chromogenic detection of bound neurotrophic factor. Using this system, GDNF in tissue culture supernatants or tissue homogenates can be quantitated in the range of 15.6–1,000pg/ml.

Features:

- **High Value:** Optimized reagents and protocol provided.
- **Specificity:** Specific detection of GDNF; less than 3% cross-reactivity with other related neurotrophic and growth factors.
- **Sensitivity:** Detect picogram levels of factor per milliliter of sample.
- **Flexibility:** Available in sizes for two or five 96-well plates; can configure plates as desired.

Protocol	Part#
Technical Bulletin	TB221

Storage Conditions: Store the entire system in its original package protected from light at –20°C. Once thawed, store the system (except the GDNF Standard) at 4°C.

NGF E_{max}® ImmunoAssay Systems

Product	Size	Cat.#
NGF E_{max}® ImmunoAssay System	2 × 96 wells	G7630
	5 × 96 wells	G7631

Description: The NGF E_{max}® ImmunoAssay Systems provide optimized reagents and a protocol for the sensitive and specific detection of biologically active nerve growth factor (NGF). After an overnight coating of a 96-well plate, the specific protein is detected using an antibody sandwich format. The systems use horseradish peroxidase-conjugated secondary antibody and a single-component TMB substrate for the final chromogenic detection of bound neurotrophic factor. The system can be used to quantitate NGF in tissue culture supernatants and tissue extracts in the range of 3.9–250pg/ml. Avoid using samples containing high levels of IgG such as serum, plasma and spleen.

Features:

- **High Value:** Optimized reagents and protocol provided.
- **Specificity:** Specific detection of NGF; less than 3% cross-reactivity with other related neurotrophic and growth factors.
- **Sensitivity:** Detect picogram levels of factor per milliliter of sample.
- **Flexibility:** Available in sizes for two or five 96-well plates; can configure plates as desired.

Protocol	Part#
Technical Bulletin	TB226

Storage Conditions: Store the entire system in its original package protected from light at –20°C.

NT-3 E_{max}® ImmunoAssay Systems

Product	Size	Cat.#
NT-3 E_{max}® ImmunoAssay System	2 × 96 wells	G7640
	5 × 96 wells	G7641

Description: The NT-3 E_{max}® ImmunoAssay Systems provide optimized reagents and a protocol for the sensitive and specific detection of neurotrophin-3 (NT-3). After an overnight coating of a 96-well plate, the specific protein is detected using an antibody sandwich format. The systems use horseradish peroxidase-conjugated secondary antibody and a single-component TMB substrate for the final chromogenic detection of bound neurotrophic factor. Using this system, NT-3 can be quantitated in the range of 4.7–300pg/ml.

Features:

- **High Value:** Optimized reagents and protocol provided.
- **Specificity:** Specific detection of NT-3; less than 3% cross-reactivity with other related neurotrophic and growth factors.
- **Sensitivity:** Detect picogram levels of factor per milliliter of sample.
- **Flexibility:** Available in sizes for two or five 96-well plates; can configure plates as desired.

Protocol	Part#
Technical Bulletin	TB243

Storage Conditions: Store the entire system in its original package protected from light at –20°C.

Block & Sample 5X Buffer

Product	Size	Cat.#
Block & Sample 5X Buffer	54 ml	G3311
For Laboratory Use.		

Description: The Block & Sample 5X Buffer is optimized for use in the E_{max}® ImmunoAssay Systems (for BDNF, GDNF, NGF and NT-3) providing additional buffer for further sample dilutions and manipulations. This buffer is used to block the plates and dilute the standards, samples, detection antibodies and conjugates in these E_{max}® ImmunoAssay Systems. The buffer is provided as 54ml of buffer containing gentamicin as a preservative.

Note: The Block & Sample 5X Buffer should not be used with the TGFβ_1 and TGFβ_2 E_{max}® ImmunoAssay Systems.

Storage Conditions: Store at 4°C.

TGFβ_1 E_{max}® ImmunoAssay Systems

Product	Size	Cat.#
TGFβ_1 E_{max}® ImmunoAssay System	2 × 96 wells	G7590
	5 × 96 wells	G7591

Description: The TGFβ_1 E_{max}® ImmunoAssay System provides optimized reagents and a protocol for the sensitive and specific detection of transforming growth factor β_1 (TGFβ_1). After an overnight coating of a 96-well plate, the specific protein is detected using an antibody sandwich format. The system uses horseradish peroxidase-conjugated secondary antibody and a single-component TMB substrate for the final chromogenic detection of bound TGFβ_1. Using this system, biologically active TGFβ_1 in tissue culture supernatants, plasma, serum or urine can be quantitated in the range of 15.6–1,000pg/ml.

Features:

- **High Value:** Optimized reagents and protocol provided.
- **Specificity:** Specific detection of TGFβ_1; less than 3% cross-reactivity with other related growth factors (TGFβ_2 and TGFβ_3).
- **Sensitivity:** Detect picogram levels of factor per milliliter of sample.
- **Flexibility:** Available in sizes for two or five 96-well plates; can configure plates as desired.

Protocol	Part#
Technical Bulletin	TB196

Storage Conditions: Store the entire system in its original package protected from light at –20°C.

TGFβ$_2$ E$_{max}$® ImmunoAssay System

Product	Size	Cat.#
TGFβ$_2$ E$_{max}$® ImmunoAssay System	5 × 96 wells	G7600

Description: The TGFβ$_2$ E$_{max}$® ImmunoAssay System provides optimized reagents and a protocol for the sensitive and specific detection of biologically active TGFβ$_2$. After an overnight coating of a 96-well plate, the specific protein is detected using an antibody sandwich format. The system uses horseradish peroxidase-conjugated secondary antibody and a single-component TMB substrate for the final chromogenic detection of bound TGFβ$_2$. Using this system, biologically active TGFβ$_2$ in media, plasma, serum or urine can be quantitated in the range of 15.6–1,000pg/ml.

Features:

- **High Value:** Optimized reagents and protocol provided.
- **Specificity:** Specific detection of TGFβ$_2$; less than 5% cross-reactivity with other related growth factors (TGFβ$_1$ and TGFβ$_3$).
- **Sensitivity:** Detect picogram levels of factor per milliliter of sample.
- **Flexibility:** Can configure plates as desired.

Protocol	Part#
Technical Bulletin	TB224

Storage Conditions: Store the entire system in its original package protected from light at –20°C.

TGFβ Sample 10X Buffer

Product	Size	Cat.#
TGFβ Sample 10X Buffer	20 ml	G1291

Description: The TGFβ Sample 10X Buffer is an optimized proprietary buffer designed for use with the TGFβ$_1$ and TGFβ$_2$ E$_{max}$® ImmunoAssay Systems to reduce high background, a common problem with traditional buffers used in TGFβ ELISAs.

Storage Conditions: Store at 4°C.

Anti-pS^{473} Akt pAb

Product	Size	Cat.#
Anti-pS^{473} Akt pAb	40 µl	G7441

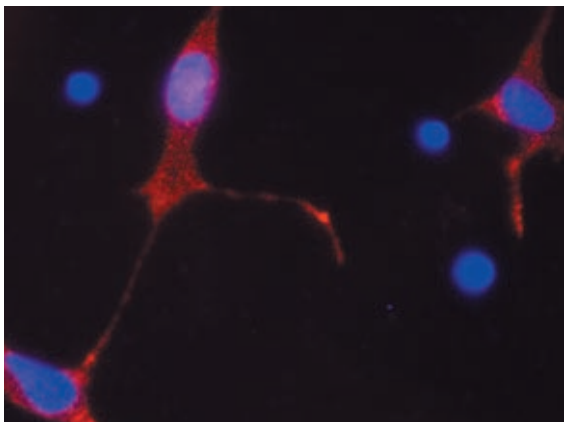
Description: Anti-pS^{473} Akt pAb is an affinity-purified polyclonal rabbit antibody. The antibody is purified using a phosphorylated peptide that corresponds to the phospho-S^{473} form of Akt-1 and is useful for both Western blotting and immunocytochemistry.

Features:

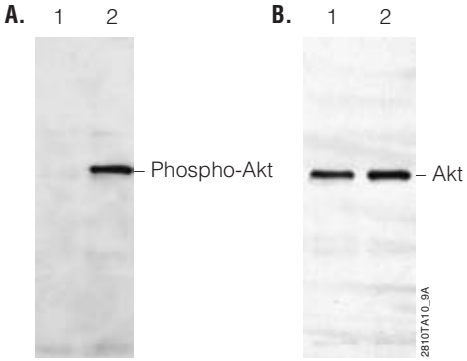
- **Specificity:** The antibody is selective for the Ser^{473} phosphorylated isoforms of Akt and does not show cross-reactivity with nonphosphorylated Akt.
- **Immunogen:** Peptide from the singly phosphorylated Ser^{473} region from the C-terminus of Akt-1 protein.
- **Antibody Form:** Affinity-purified rabbit IgG, supplied in PBS with 50µg/ml gentamicin.
- **Value:** Will generate 100ml of blotting solution, sufficient for 10 Western blots of 10ml each.

Protocol	Part#
Promega Product Information	9PIG744

Storage Conditions: Store at 4°C for daily/weekly use or dispense into aliquots and store at –20°C for long-term storage.



Immunocytochemical staining of Akt in embryonic rat brain cells. Embryonic (day 17) rat brain cells were collected and treated with 20ng/ml each of EGF and FGF. Anti-pS^{473} Akt pAb was used at a 1:50 dilution. Positive cells were visualized using a donkey anti-rabbit, Cy®3-conjugated secondary antibody. Nuclei were stained using DAPI. Protocols developed and performed at Promega.



Detection of phosphorylated Akt by Western blot analysis with Anti-pS^{473} Akt pAb. Panel A. NIH/3T3 total cell extract (10µg per lane) was resolved by polyacrylamide gel electrophoresis and blotted onto nitrocellulose. Lane 1, untreated cells; lane 2, cells pretreated with PDGF (Invitrogen) at 50ng/ml for 20 minutes. Anti-pS^{473} Akt pAb (Cat.# G7441) was used at a 1:2,500 dilution. The blot was probed with Donkey Anti-Rabbit IgG (H+L), HRP, Anti-ACTIVE® Qualified pAb (Cat.# V7951) at 1:10,000 dilution followed by chemiluminescent detection. **Panel B.** A pan-Akt pAb (New England Biolabs) reveals total Akt in both stimulated and unstimulated NIH/3T3 cell extracts. Secondary antibody and detection methods were the same as those used in Panel A.

Anti-ACTIVE® CaM KII pAb, Rabbit, (pT^{286})

Product	Size	Cat.#
Anti-ACTIVE® CaM KII pAb, Rabbit, (pT^{286})	40 µl	V1111

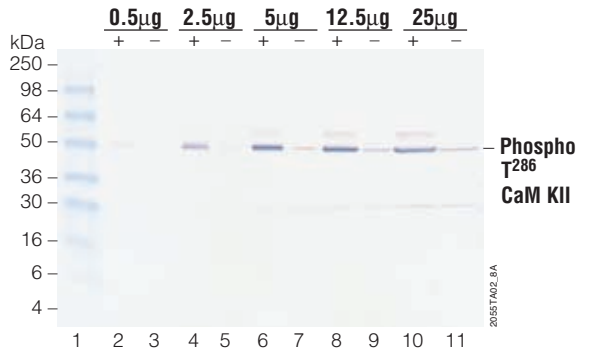
Description: This polyclonal antibody (pAb) is specific for the multifunctional calcium/calmodulin-dependent protein kinase CaM kinase II (CaM KII) that is phosphorylated on threonine 286 (pT^{286}). The Anti-ACTIVE® CaM KII pAb was raised against the phosphothreonine-containing peptide.

Features:

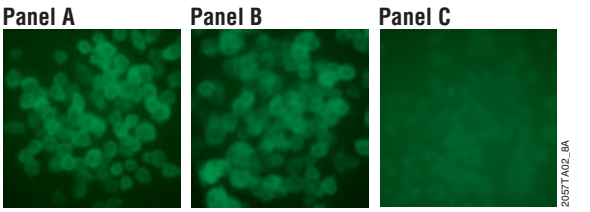
- **Specificity:** Preferentially detects CaM KII autophosphorylated on threonine 286.
- **Immunogen:** Threonine-phosphorylated peptide corresponding to the phosphorylated Thr^{286} region of the mammalian calcium/calmodulin-dependent protein kinase.
- **Antibody Form:** Affinity-purified rabbit IgG; supplied in 10mM sodium phosphate (pH 7.4), 20mM NaCl.
- **Value:** When used at the recommended dilution of 1:5,000, it generates up to 200ml of blotting solution sufficient for 20 Western blots of 10ml each.

Protocol	Part#
Technical Bulletin	TB264

Storage Conditions: Store at –20°C.



Detection of CaM KII by Anti-ACTIVE® CaM KII pAb in Western analysis of rat brain homogenate. Lanes 2, 4, 6, 8 and 10 contain autophosphorylated (+) brain cytosolic protein in the amounts shown; lanes 3, 5, 7, 9 and 11 contain nonphosphorylated (–) brain cytosolic protein in the amounts shown. The presence of the autophosphorylated CaM KII was detected using the Anti-ACTIVE® CaM KII pAb diluted 1:5,000.



Immunocytochemical detection of autophosphorylated CaM KII in PC12 cells with Anti-ACTIVE® CaM KII pAb. PC12 cells were adhered to slides coated with collagen, fixed in 10% paraformaldehyde for 30 minutes, rinsed in PBS and permeabilized in methanol for 10 minutes at –20°C. The cells were then blocked in 1% BSA in PBS for 45 minutes followed by 2% horse serum in PBS for 60 minutes. Cells were incubated overnight at 4°C with **(Panel A)** Ab alone, **(Panel B)** Ab preincubated with a nonphosphorylated CaM KII peptide fragment (1µg/ml) or **(Panel C)** Ab preincubated with a phosphorylated CaM KII peptide fragment (1µg/ml). The Anti-ACTIVE® CaM KII pAb was used at 1:500 dilution and preincubated with peptide for 8 hours at 4°C. After incubation with the Anti-ACTIVE® CaM KII pAb or Ab/peptide mixture, the cells were rinsed in PBS and incubated with a donkey anti-rabbit FITC-conjugated secondary Ab (1:500) for 60 minutes at room temperature. Staining was visualized with a Zeiss® fluorescence microscope. The results demonstrate that preincubation of the Anti-ACTIVE® CaM KII pAb with phosphorylated CaM KII peptide completely abolishes immunostaining **(Panel C)**, but preincubation with nonphosphorylated CaM KII peptide has no effect on immunostaining **(Panel A versus Panel B).** Protocols developed and performed at Promega.

Anti-ACTIVE® JNK pAb, Rabbit, (pTPpY)

Product	Size	Cat.#
Anti-ACTIVE® JNK pAb, Rabbit, (pTPpY)	40 µl	V7931
	120 µl	V7932

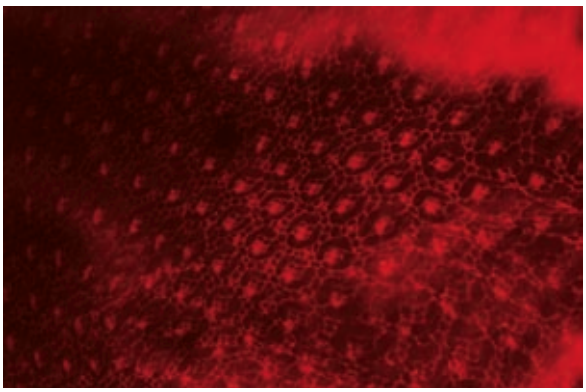
Description: Anti-ACTIVE® JNK pAb is a polyclonal antibody from rabbit serum. The antibody is affinity purified using a dually phosphorylated peptide that corresponds to the active form of the JNK enzymes.

Features:

- **Specificity:** Preferentially detects the dually phosphorylated, active form of the stress-activated protein kinase (SAPK), also known as c-Jun N-terminal kinase, JNK.
- **Immunogen:** Dually phosphorylated Thr/Pro/Tyr region (pTPpY) derived from the catalytic core of the active form of JNK kinase, which corresponds to Thr^{183} and Tyr^{185} of the mammalian JNK2 enzyme.
- **Antibody Form:** Affinity-purified rabbit IgG; supplied in 10mM sodium phosphate (pH 7.4), 20mM NaCl.
- **Value:** Anti-ACTIVE® JNK pAb is available in two convenient sizes. Cat.# V7931 will generate up to 200ml of blotting solution, sufficient for 20 Western blots of 10ml each. The larger size, Cat.# V7932, will generate up to 600ml of blotting solution, sufficient for 60 Western blots of 10ml each.

Protocol	Part#
Technical Bulletin	TB262

Storage Conditions: Store at –20°C.



1825TA07_7A

Immunocytochemical detection of active JNK enzyme in *Drosophila* pupal retina using the Anti-ACTIVE® JNK pAb. *Drosophila* pupal retina at 25% of pupal development were fixed in 3% paraformaldehyde in PBS. The Anti-ACTIVE® JNK pAb was diluted 1:100 in PBS containing 10% fetal bovine serum and 0.2% Triton® X-100. Samples were incubated with the primary antibody overnight at 4°C, washed 3 times (10 minutes each) with 0.2% Triton® X-100 and then incubated with a goat anti-rabbit Cy®3 conjugate for 2 hours at 4°C. Whole mounts were visualized with a Zeiss® Axioskop fluorescent microscope. The results illustrate the presence of dually phosphorylated, active forms of JNK in discrete structures of the fly retinal ommatidia including intense staining of the inner cone cells as well as the mechanosensory bristles and surrounding pigment cells. The pattern of staining (which was distinct from results obtained with an antibody for active p38) and the absence of staining in control experiments (data not shown) support the high specificity of the Anti-ACTIVE® JNK pAb. Image kindly provided by David T. Miller and Ross Cagan, Department of Molecular Biology and Pharmacology, Washington University School of Medicine, St. Louis, Missouri.

Anti-ACTIVE® MAPK pAb, Rabbit, (pTEpY)

Product	Size	Cat.#
Anti-ACTIVE® MAPK pAb, Rabbit, (pTEpY)	40 µl	V8031

Description: Anti-ACTIVE® MAPK pAb is a polyclonal rabbit antibody. The antibody is affinity purified using a dually phosphorylated peptide that corresponds to the active form of the mitogen-activated protein (MAP) kinase enzymes.

Features:

- **Specificity:** Preferentially detects the dually phosphorylated, active form of the mitogen-activated protein kinase (MAPK) enzymes (ERK1 and ERK2).
- **Immunogen:** Dually phosphorylated Thr/Glu/Tyr region (pTEpY) derived from the catalytic core of the active form of the mitogen-activated protein kinase (MAPK) enzymes, ERK1 and ERK2, which corresponds to Thr^{183} and Tyr^{185} of the mammalian ERK2 enzyme.
- **Antibody Form:** Affinity-purified rabbit IgG; supplied in PBS (pH 7.4).
- **Value:** When used at the recommended 1:5,000 dilution, this product will generate 200ml of blotting solution, sufficient for 20 Western blots of 10ml each.

Protocol	Part#
Technical Bulletin	TB262

Storage Conditions: Store at –20°C.

Anti-ACTIVE® MAPK Family Sampler

Product	Size	Cat.#
Anti-ACTIVE® MAPK Family Sampler	1 each	V3281

Description: The Anti-ACTIVE® MAPK Family Sampler contains polyclonal antibodies to three members of the MAPK superfamily of protein kinases: MAPK (also known as ERK1 and 2), c-Jun N-terminal kinase or JNK, and p38 kinase. Also included in the sampler is a Donkey Anti-Rabbit IgG-Alkaline Phosphatase secondary conjugate. When used at the recommended dilutions, the sampler includes enough antibody to generate 40ml of blotting solution for each Anti-ACTIVE® antibody.

Protocol	Part#
Technical Bulletin	TB262

Anti-ACTIVE® p38 pAb, Rabbit, (pTGpY)

Product	Size	Cat.#
Anti-ACTIVE® p38 pAb, Rabbit, (pTGpY)	100 µl	V1211

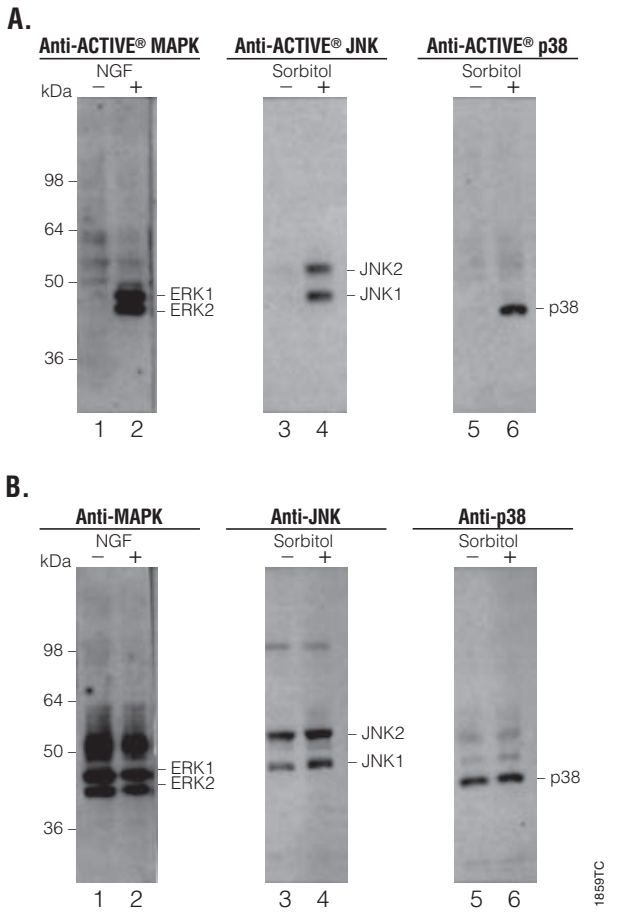
Description: Anti-ACTIVE® p38 pAb is a polyclonal rabbit antibody. The antibody is affinity-purified using a dually phosphorylated peptide that corresponds to the active form of the p38 enzymes.

Features:

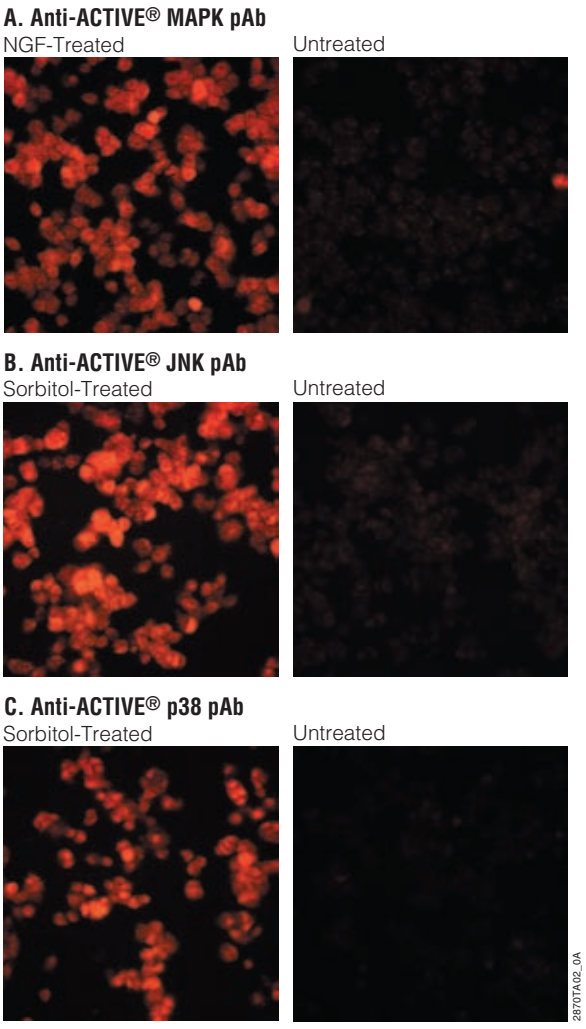
- **Specificity:** Preferentially detects the dually phosphorylated, active form of p38 kinase.
- **Immunogen:** Dually phosphorylated Thr/Gly/Tyr region (pTGpY) derived from the catalytic core of the active form of p38 kinase, which corresponds to Thr^{180} and Tyr^{182} of the mammalian p38 enzyme.
- **Antibody Form:** Affinity-purified rabbit IgG; supplied in PBS (pH 7.4).
- **Value:** When used at the recommended 1:2,000 dilution, this product will generate up to 200ml of blotting solution, sufficient for 20 Western blots of 10ml each.

Protocol	Part#
Technical Bulletin	TB262

Storage Conditions: Store at –20°C.



Detection of MAPK, JNK and p38 in PC12 cell extracts. Panel A. Western blot analysis using Anti-ACTIVE® MAPK, Anti-ACTIVE® JNK and Anti-ACTIVE® p38 polyclonal antibodies to detect activated MAPK, JNK and p38. **Panel B.** Western blot analysis using anti-MAPK, anti-JNK and anti-p38 antibodies to detect activated and nonactivated MAPK, JNK and p38 in untreated or NGF- or sorbitol-treated PC12 cells.



Detection of activated MAPK, JNK and p38 in PC12 cells by immunocytochemistry. PC12 cells were grown to 80% confluence in RPMI 1640 medium supplemented with 25mM HEPES, 300mg/l l-glutamine, 10% horse serum, 5% fetal bovine serum and 0.5mM EGTA. Cells were either untreated or treated with 200ng/ml NGF or 1M sorbitol as indicated. ICC was performed as described in Promega Technical Bulletin #TB262. Anti-ACTIVE® antibodies were used at the following dilutions: **Panel A.** MAPK, 1:500; **Panel B.** JNK, 1:1,000; **Panel C.** p38, 1:500. Protocols developed and performed at Promega.

Anti-pT^{183} MAPK pAb, Rabbit

Product	Size	Cat.#
Anti-pT^{183} MAPK pAb, Rabbit	50 µl	V8081

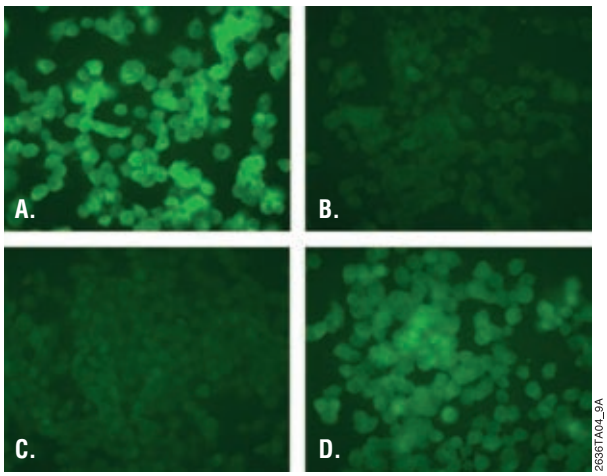
Description: Anti-pT^{183} MAPK pAb is a polyclonal antibody purified from rabbit serum. The antibody is affinity-purified using a monophosphorylated peptide (mono pT peptide) corresponding to a monophospho-threonine form of MAP kinase enzymes 1 and 2.

Features:

- **Specificity:** Detects the monophosphorylated threonine residue 183 of ERK1 and ERK2.
- **Immunogen:** Peptide sequence corresponding to the monophosphorylated form of ERK1 and ERK2 enzymes phosphorylated on the threonine 183 residue.
- **Antibody Form:** Affinity-purified rabbit IgG; supplied in PBS (pH 7.4).
- **Value:** When used at the recommended 1:4,000 dilution, generates up to 200ml of blotting solution, sufficient for 20 Western blots of 10ml each.

Protocol	Part#
Promega Product Information	9PIV808

Storage Conditions: Store at –20°C.



Immunostaining of PC12 cells labeled with Anti-pT^{183} MAPK pAb. **Panel A.** PC12 cells stimulated with 0.2µg/ml NGF for 5 minutes. **Panel B.** Unstimulated PC12 cells. **Panel C.** NGF-stimulated PC12 cells plus antibody preincubated with 1µg/ml pT^{183} peptide for 4 hours at 4°C. **Panel D.** NGF-stimulated PC12 cells plus antibody preincubated with 1µg/ml pY^{185} peptide for 4 hours at 4°C. Protocols developed and performed at Promega.

Anti-ERK 1/2 pAb, Rabbit

Product	Size	Cat.#
Anti-ERK 1/2 pAb, Rabbit	40 µl	V1141

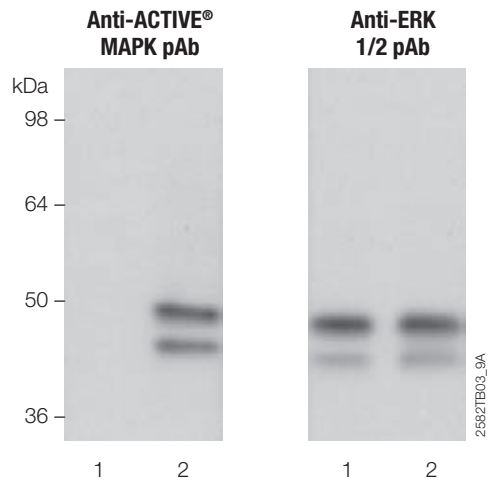
Description: Anti-ERK 1/2 pAb is a polyclonal antibody purified from rabbit serum. The antibody is affinity-purified using a peptide sequence in human/rat ERK1.

Features:

- **Specificity:** Detects ERK1 and ERK2 in the nonphosphorylated, monophosphorylated and dually phosphorylated forms.
- **Immunogen:** Sequence representing a conserved region in human and rat ERK1 located outside of the catalytic core of the enzyme.
- **Antibody Form:** Affinity-purified rabbit IgG; supplied in PBS (pH 7.4).
- **Value:** When used at the recommended 1:5,000 dilution, this product will generate up to 200ml of blotting solution, sufficient for 20 Western blots of 10ml each.

Protocol	Part#
Promega Product Information	9PIV114

Storage Conditions: Store at –20°C.



Detection of the specifically phosphorylated form of MAPK in NGF-treated PC12 cell extracts. Anti-ACTIVE® MAPK pAb (Cat.# V8031) and Anti-ERK 1/2 ("pan ERK 1/2") pAb (Cat.# V1141) detection of ERK 1/2 in untreated (lanes 1) or NGF-treated (lanes 2) PC12 cell extracts (2µg).

Anti-Human BDNF pAb

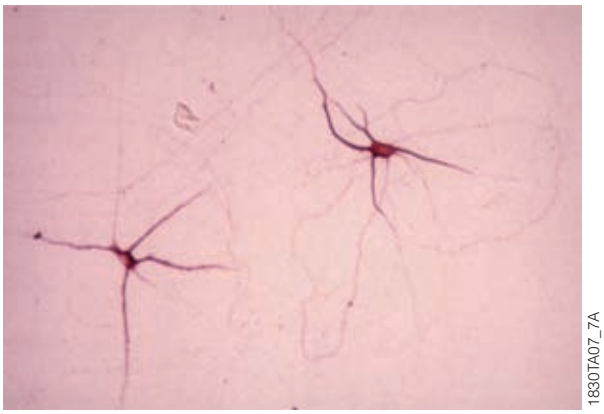
Product	Size	Cat.#
Anti-Human BDNF pAb	200 µg	G1641

Description: BDNF, a 27kDa homodimer originally derived from human brain, shares high sequence homology with NGF, NT-3 and NT-4/5 and influences many neuron types in the CNS. Anti-Human BDNF pAb is generated in chickens and purified using the EGGstract® IgY Purification System. IgY, the 180kDa chicken IgG homolog, can be produced in chickens against certain biological antigens that fail to elicit a humoral immune response in rabbits or other mammals due to species relatedness. This antibody is highly specific for BDNF.

Features:

- **Immunogen:** Human recombinant BDNF.
- **Antibody Form:** Chicken IgY, provided at 0.5mg/ml in 0.1M NaCl, 0.01M K_2HPO_4 and 50µg/ml gentamicin.
- **Specificity:** Cross-reactive between mammalian species; does not cross-react with other neurotrophic factors.

Storage Conditions: Store at 4°C.



1830TA07_7A

Localization of BDNF in primary cultures of hippocampal neurons. The Anti-Human BDNF pAb was used at a 1:200 dilution. Primary antibody was detected using HRP-conjugated goat anti-chicken IgY secondary antibody. Photomicrograph kindly provided by Dr. Laurie Goodman, Lynx Therapeutics, Hayward, CA. Reprinted by permission of Academic Press, Goodman, L. *et al.* (1996) *Mol. Cell Neurosci.* **7**, 222.

Anti-ACTIVE® Caspase-3 pAb

Product	Size	Cat.#
Anti-ACTIVE® Caspase-3 pAb	50 µl	G7481

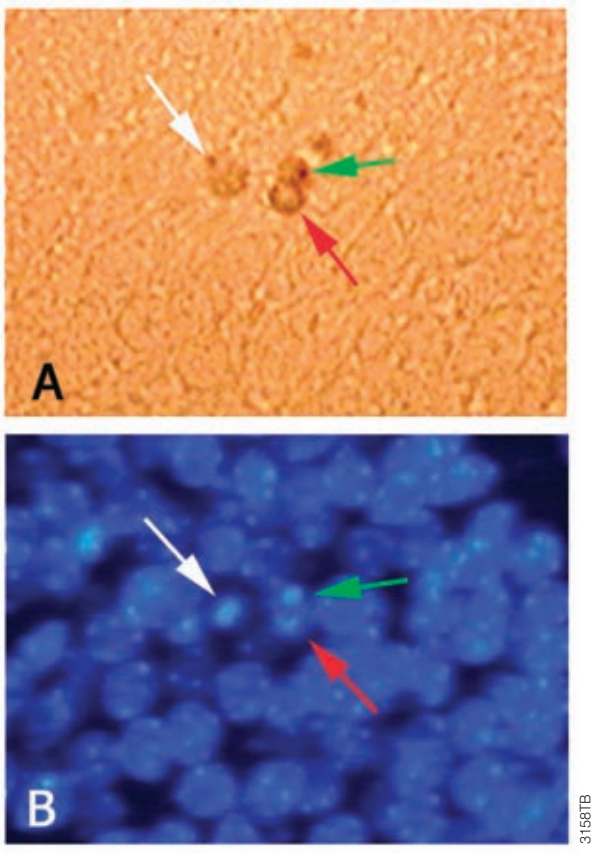
Description: Anti-ACTIVE® Caspase-3 pAb is intended for use as a marker of apoptosis; it specifically stains apoptotic cells without staining nonapoptotic cells. Includes sufficient antibody to perform 125 immunocytochemical assays (100µl/assay) at a 1:250 dilution.

Features:

- **Immunogen:** Peptide derived from the p17 fragment of caspase-3 and having sequence homology in human, mouse, rat and hamster.
- **Antibody Form:** Affinity-purified rabbit IgG; supplied in Dulbecco's PBS.
- **Specificity:** Specifically recognizes the cleaved active form of caspase-3 in human, rat and mouse.

Protocol	Part#
Promega Product Information	9PIG748

Storage Conditions: Store at –20°C.



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Demonstration of Anti-ACTIVE® Caspase-3 pAb positive cells in postnatal day 0 (P0) mouse brain paraffin-embedded sections. **Panel A.** Three Anti-ACTIVE® Caspase-3 pAb-positive cells (colored arrows). **Panel B.** Corresponding DAPI-stained nuclei. Note the correspondence of Anti-ACTIVE® Caspase-3 pAb label with the typical apoptotic, condensed nuclear morphology in Panel B. Protocols developed and performed at Promega.

Anti-Rat CNTF pAb

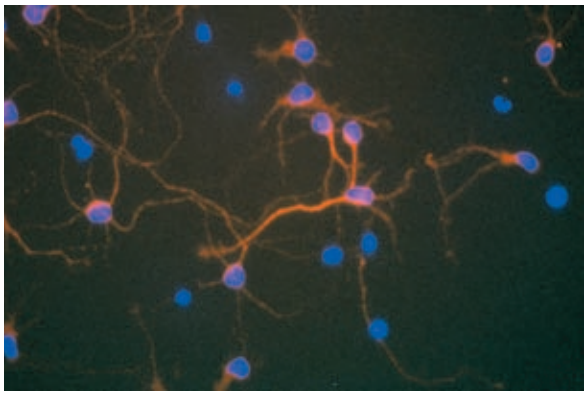
Product	Size	Cat.#
Anti-Rat CNTF pAb	200 µg	G1631

Description: Ciliary Neurotrophic Factor (CNTF) is a 23kDa protein shown to exert effects on a wide number of peripheral and central nervous system cell types. CNTF is structurally unrelated to nerve growth factor, neurotrophin-3 and neurotrophin-4, and uses a signaling pathway distinct from the Trk pathways utilized by these factors. Anti-Rat CNTF pAb provides a useful tool for understanding CNTF's effects by Western blotting, immunostaining and ELISA applications.

Features:

- **Immunogen:** Rat recombinant CNTF.
- **Antibody Form:** Chicken IgY, provided at 0.5mg/ml as frozen liquid in 0.1M NaCl, 0.01M K_2HPO_4 and 50µg/ml gentamicin.
- **Specificity:** Cross-reactive between mammalian species; does not cross-react with other neurotrophic factors.

Storage Conditions: Store at –20°C.



Double-fluorescence staining of a human fetal CNS cell preparation. Red cells are immunostained with chicken Anti-Rat CNTF pAb and visualized with donkey anti-chicken Cy®3 conjugate. Nuclei (blue) are stained with DAPI. Protocols developed and performed at Promega.

Anti-β-Galactosidase mAb

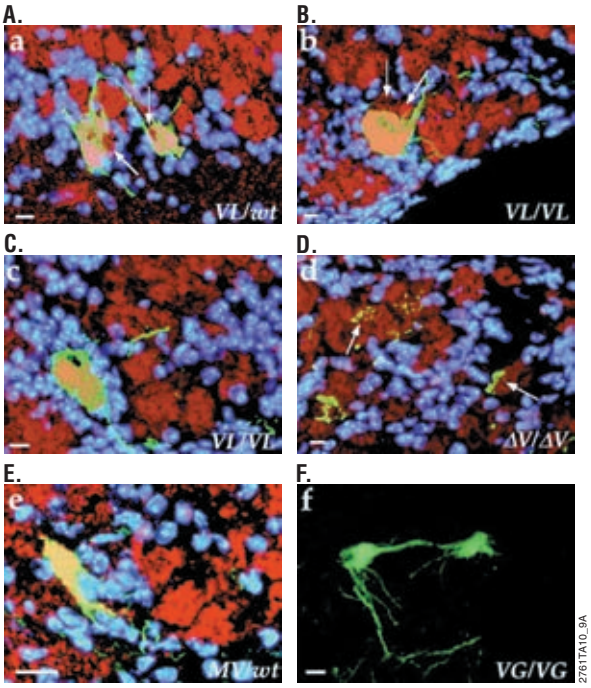
Product	Size Conc.	Cat.#
Anti-β-Galactosidase, Purified Monoclonal Antibody	100 µg 2.0–2.5 mg/ml	Z3781
	2 mg 2.0–2.5 mg/ml	Z3783
For Laboratory Use.		

Description: This antibody [subclass $IgG_{2a}(\kappa)$] was purified from ascites of a mouse hybridoma and recognizes *E. coli* β-galactosidase.

Features:

- **Immunogen:** β-galactosidase.
- **Antibody Form:** 2.0–2.5mg/ml in 10mM Tris-HCl (pH 8.0), 150mM NaCl, 0.02% sodium azide.
- **Specificity:** *E. coli* β-galactosidase near the C-terminal end.

Storage Conditions: Store undiluted at –20°C.



Histological analysis of axonal termination in the accessory olfactory bulb (AOB). Sagittal sections through the AOB stained with the Anti-β-Galactosidase mAb (Cat.# Z3781; green) and antibodies against synaptophysin (DAKO, red). DAPI staining is shown in blue. **Panel A.** Heterozygous VL mouse. **Panels B and C.** Homozygous VL mouse. **Panel D.** Homozygous ΔV mouse. **Panel E.** Heterozygous MV mouse. **Panel F.** Homozygous VG mouse. Details on gene targeting, mutations and immunostaining may be found in Rodriguez, J., Feinstein, P. and Mombaerts, P. (1999) *Cell* **97**, 199. Images kindly provided by Dr. Peter Mombaerts, The Rockefeller University, New York. Reprinted by permission of Cell Press.

Anti-Human GDNF pAb

Product	Size	Cat.#
Anti-Human GDNF pAb	200 µg	G2791

Description: Human glial cell-line-derived neurotrophic factor (GDNF), a 30kDa homodimer, has been shown to be a potent survival factor for a variety of neurons. The receptor complex for GDNF has been elucidated, though members of the multicomponent receptor family continue to grow. With applications in Western blotting, ELISA and immunostaining, the Anti-Human GDNF pAb is a useful tool to continue the investigation of GDNF's role in multiple facets of neurological systems.

Features:

- **Immunogen:** Human recombinant GDNF.
- **Antibody Form:** Chicken IgY; 0.5mg/ml in 0.1M NaCl, 0.01M K_2HPO_4, 50µg/ml gentamicin.
- **Specificity:** Cross-reactive between mammalian species; does not cross-react with TGFα, TGFβ_1, NGF or BDNF at up to 10µg/ml.

Storage Conditions: Store at 4°C.

Anti-GFAP pAb

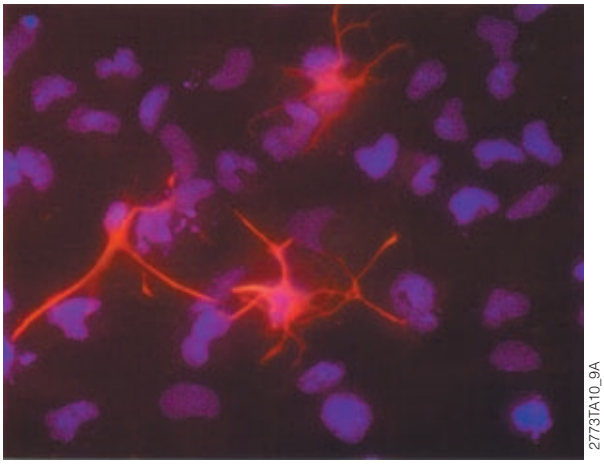
Product	Size	Cat.#
Anti-GFAP pAb	100 µg	G5601

Description: Anti-GFAP pAb is a polyclonal antibody against glial fibrillary acidic protein (GFAP), a specific marker of astrocytes in the central nervous system, and is qualified for immunostaining applications.

Features:

- **Immunogen:** Purified glial fibrillary acidic protein from bovine spinal cord.
- **Antibody Form:** Purified rabbit IgG; supplied at 1mg/ml in PBS containing 50µg/ml gentamicin.
- **Specificity:** Human, bovine and rat GFAP; not recommended for mouse.

Storage Conditions: Store at 4°C.



2773TA10_9A

Anti-GFAP-labeled astrocytes in mixed rat neural progenitor cultures. Cells were labeled with DAPI (blue) and Anti-GFAP pAb with a Cy®3-conjugated secondary antibody (red). Protocols developed and performed at Promega.

Anti-Luciferase pAb

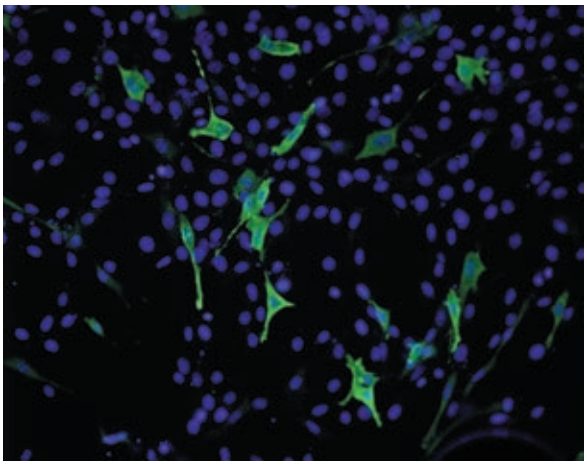
Product	Size	Cat.#
Anti-Luciferase pAb	200 µg	G7451

Description: Anti-Luciferase pAb is a goat polyclonal antibody designed for use in immunocytochemistry and Western blot applications. Anti-Luciferase pAb can detect luciferase enzyme expression in situ.

Features:

- **Immunogen:** Recombinant luciferase from North American firefly (*Photinus pyralis*).
- **Antibody Form:** Goat polyclonal IgG at 1mg/ml in PBS containing 50µg/ml gentamicin.
- **Specificity:** Anti-Luciferase pAb is specific for firefly luciferase (*Photinus pyralis*) and does not cross-react with sea pansy (*Renilla reniformis*) luciferase.

Storage Conditions: Store at 4°C.



2760TA10_9A

NIH/3T3 cells transiently transfected with a luciferase gene. Luciferase-expressing cells were detected using the Anti-Luciferase pAb (Cat.# G7451). Protocols developed and performed at Promega.

Anti-NGF mAb

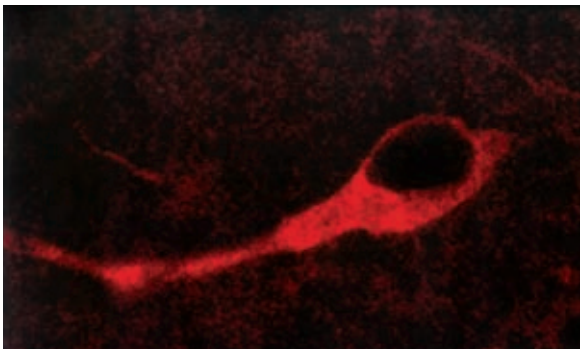
Product	Size	Cat.#
Anti-NGF mAb	20 µg	G1132
	100 µg	G1131

Description: Nerve growth factor (NGF) is a member of the neurotrophin family of growth factors. NGF is expressed in sympathetic and sensory-innervated peripheral tissues and mediates phosphorylation of specific intracellular proteins. At the cellular level, NGF expression has been demonstrated in lymphocytes, smooth muscle cells, epithelial cells, astrocytes, fibroblasts and Schwann cells. Anti-NGF mAb was designed as a specific marker of NGF in Western blotting, ELISA and immunostaining applications.

Features:

- **Immunogen:** Purified murine NGF, 2.5S.
- **Antibody Form:** Rat IgG (clone 1G3) provided at 1mg/ml as frozen liquid in PBS containing no preservatives.
- **Specificity:** Reacts with human NGF, 2.5S mNGF and to a lesser extent with 7S mNGF. Cross-reacts between mammalian species.
- **Activity:** The Anti-NGF mAb exhibits a half-maximal titer of less than or equal to 250ng/ml in an ELISA protocol using 100ng of 2.5S mNGF (Cat.# G5141).

Storage Conditions: Store at –20°C.



1283TA12_5B

Rat basal forebrain cholinergic neuron stained with Anti-NGF mAb following intraventricular injection of 30µg of NGF and factor uptake. Photomicrograph kindly provided by Dr. Charles Howe, University of California, San Francisco.

Anti-Human NT-3 pAb

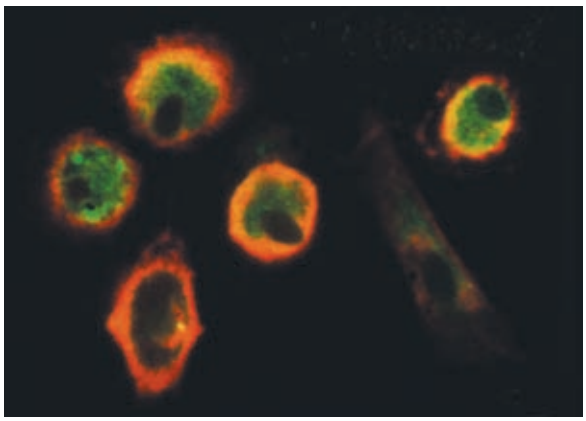
Product	Size	Cat.#
Anti-Human NT-3 pAb	200 µg	G1651

Description: Neurotrophin-3, a 27kDa homodimer that shares high sequence homology with NGF, BDNF, NT-4 and NT-5, influences many neuron types in the central and peripheral nervous system. NT-3 is also highly conserved across species. Anti-Human NT-3 pAb is generated in chickens and purified using the EGGstract® IgY Purification System. IgY, the 180kDa chicken IgG homolog, can be produced in chickens against certain biological antigens that fail to elicit a humoral immune response in rabbits or other mammals due to species relatedness. This antibody is highly specific for NT-3 in a variety of mammalian species.

Features:

- **Immunogen:** Human recombinant NT-3.
- **Antibody Form:** Chicken IgY is provided at 0.5mg/ml in 0.1M NaCl, 0.01M K_2HPO_4 and 50µg/ml gentamicin.
- **Specificity:** Cross-reactive with human and mouse NT-3 and is presumed to cross-react with rat and Rhesus monkey NT-3 based on factor sequence identity across species; does not cross-react with BDNF or NGF and has limited cross-reactivity to NT-4.

Storage Conditions: Store at 4°C.



1411TA03_6A

Immunofluorescent detection of NT-3 in monocyte-derived macrophages and purified human fetal microglia. Chicken Anti-Human NT-3 pAb in red and RCA-1 (macrophage marker) in green. Image kindly provided by Drs. Pam Sarnacki, Wanda Wang and Chris Achim, University of Pittsburgh.

Anti-PARP p85 Fragment pAb

Product	Size	Cat.#
Anti-PARP p85 Fragment pAb	50 µl	G7341

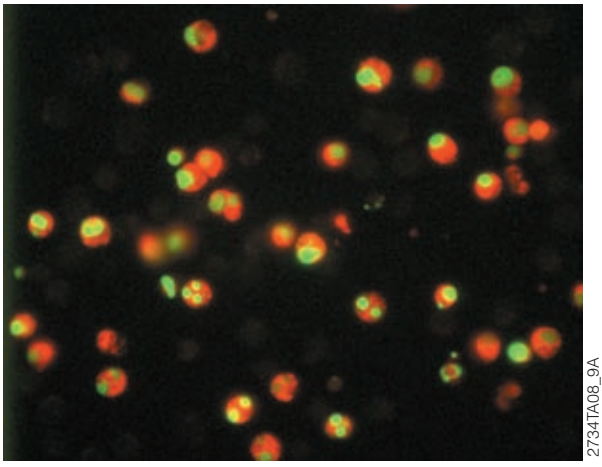
Description: Poly (ADP-ribose) polymerase (PARP), a nuclear enzyme involved in DNA repair, is a well known substrate for caspase-3 cleavage during apoptosis. Anti-PARP p85 Fragment pAb is a rabbit polyclonal antibody specific for the p85 fragment of PARP that results from caspase cleavage of the 116kDa intact molecule and thus provides an in situ marker for apoptosis. The antibody is affinity-purified using a peptide that corresponds to a region of the p85 fragment of PARP. The PARP immunogen is a synthetic peptide, gly-val-asp-glu-val-ala-lys (GVDEVAK), representing the N-terminus of the large C-terminal fragment of human PARP that results from caspase-3 cleavage. Each batch of antibody is quality assurance tested for use in immunostaining applications and contains sufficient antibody for 50 immunocytochemical reactions at the suggested working dilution of 1:100.

Features:

- **Immunogen:** N-terminal peptide from p85 fragment.
- **Antibody Form:** Affinity-purified rabbit polyclonal antibody provided in Dulbecco's PBS.
- **Specificity:** Specifically detects PARP p85 fragment in human, rat and bovine cells and tissues. Does not recognize the 116kDa intact PARP protein.

Protocol	Part#
Technical Bulletin	TB273

Storage Conditions: Store at –20°C.



2734TA08_9A

Anti-PARP p85 Fragment pAb and TUNEL double-labeling of apoptotic Jurkat cells. Cells were labeled with the Anti-PARP p85 Fragment pAb (red) and the DeadEnd™ Fluorometric TUNEL System (Cat.# G3250; green). The colocalization of cleaved PARP in cells containing TUNEL-positive nuclei demonstrates that the Anti-PARP p85 Fragment pAb specifically labels apoptotic cells. Protocols developed and performed at Promega.

Anti-Human p75 pAb

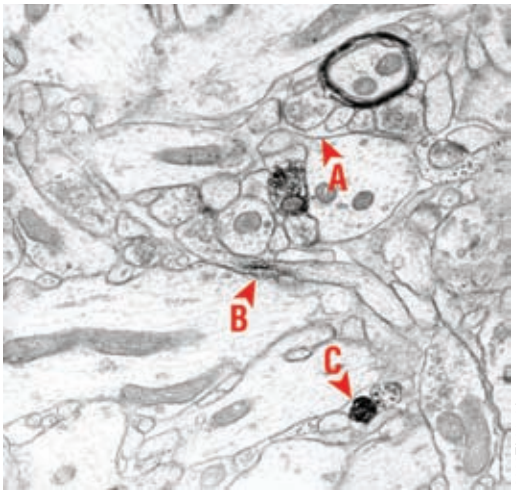
Product	Size	Cat.#
Anti-Human p75 pAb	200 µg	G3231

Description: The p75 neurotrophin receptor ($p75^{NTR}$), also known as low-affinity NGF receptor (LNGFR) and $p75^{LNGFR}$, binds nerve growth factor, brain-derived neurotrophic factor, neurotrophin-3 and neurotrophin-4 with varying specificities. $p75^{NTR}$ plays an important role in neurotrophic factor signaling including neuronal apoptosis. Anti-Human p75 pAb provides a valuable tool for understanding the role of $p75^{NTR}$ in neuronal death.

Features:

- **Immunogen:** Cytoplasmic domain of the human p75 neurotrophin receptor.
- **Antibody Form:** Purified rabbit IgG; 1mg/ml in PBS containing 50µg/ml gentamicin.
- **Specificity:** Human, rat, mouse and chicken p75.

Storage Conditions: Store at 4°C.



2029TA01_8A

Electron micrograph demonstrating immunostaining with Anti-Human p75 pAb in the inner molecular layer of the rat dentate gyrus. An axon terminal containing p75 immunoreactivity (A➤) is seen forming a synapse with a large unlabeled dendrite. Also labeled are a lengthwise axonal profile (B➤) and a small axonal cross section (C➤). Pre-embedding (Epon) immunohistochemistry was visualized with VECTASTAIN® ABC Reagent. Myelin sheath appears black due to OsO_4 fixation. Image kindly provided by Drs. Karen Dougherty and Teresa Milner, Cornell University Medical College.

Anti-TGFβ$_1$ pAb

Product	Size	Cat.#
Anti-TGFβ$_1$ pAb	100 µg	G1221

Description: Transforming growth factor β$_1$ (TGFβ$_1$) is a 25kDa homodimer composed of two 12.5kDa subunits held together by disulfide bonds. TGFβ$_1$ is a protein of immense interest to a number of

fields and has been associated with intracellular matrix deposition and tissue repair/damage, cell cycle control and apoptosis. The Anti-TGFβ$_1$ pAb is directed against biologically active
human TGFβ$_1$, providing a useful tool to analyze TGFβ$_1$ in Western blot analysis or immunostaining applications.

Features:

- **Immunogen:** Biologically active human TGFβ$_1$.
- **Antibody Form:** Rabbit IgG provided at 1mg/ml in PBS containing 0.02mg/ml gentamicin as a preservative.
- **Specificity:** Reacts with biologically active TGFβ$_1$ with no cross-reactivity to TGFβ$_2$ and TGFβ$_3$.

Storage Conditions: Store at –20°C.

Anti-TrkB In pAb

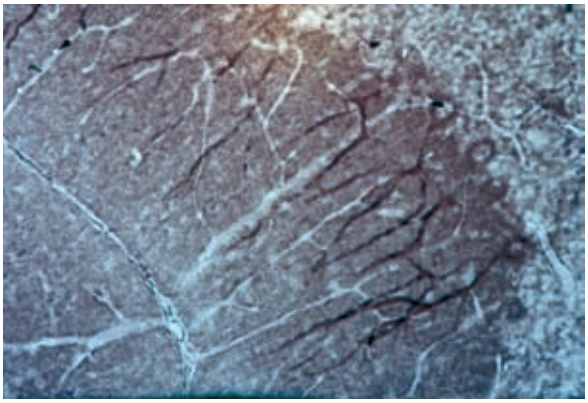
Product	Size	Cat.#
Anti-TrkB In pAb	100 µg	G1561

Description: TrkB is the high-affinity receptor for brain-derived neurotrophic factor (BDNF) and neurotrophin-4 (NT-4). Two forms of the TrkB receptor exist: the full-length, active 145kDa protein and a truncated, nonsignaling 95kDa protein. The truncated isoform lacks the cytoplasmic tyrosine kinase catalytic region and therefore does not signal in response to BDNF or NT-4. The Anti-TrkB In pAb is a polyclonal antibody raised in chickens against a cytoplasmic domain of the carboxy terminus of mouse TrkB.

Features:

- **Immunogen:** A fusion protein containing residues 482–501 (HISNGSNTPSSSEGGPDAVI) derived from a portion of the intracellular domain of mouse TrkB generated in *E. coli* using a proprietary synthetic carrier protein technology.
- **Antibody Form:** Chicken IgY provided at 0.5mg/ml in 0.1M NaCl, 0.01M K_2PO_4 (pH 7.4).
- **Specificity:** Detects full-length TrkB, the high-affinity receptor for BDNF and NT-4, in human, rat and primate brain.

Storage Conditions: Store at –20°C.



Immunohistochemical staining of rat cerebellum Purkinje cells with chicken Anti-TrkB In pAb. Visualized with a donkey anti-chicken IgY, biotin conjugate, followed by VECTASTAIN® Elite AB reagent, DAB reaction and OsO_4 enhancement. Protocols developed and performed at Promega.

Anti-βIII Tubulin mAb

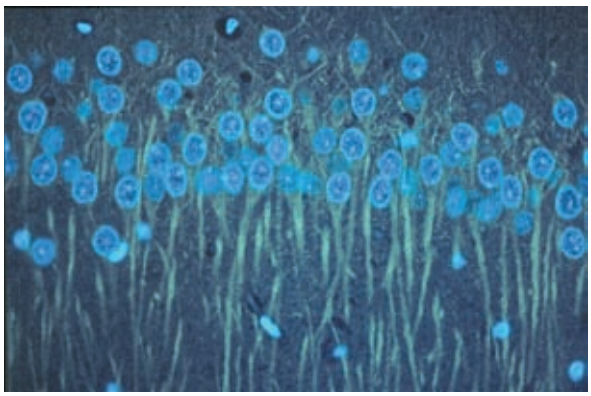
Product	Size	Cat.#
Anti-βIII Tubulin mAb	100 µg	G7121

Description: Anti-βIII Tubulin mAb is a protein G-purified IgG$_1$ monoclonal antibody (from clone 5G8) raised in mice against a peptide (EAQGPK) corresponding to the C-terminus of βIII tubulin. It is directed against βIII tubulin, a specific marker for neurons. The major use of this antibody is for labeling neurons in tissue sections and cell culture. The antibody has been tested to perform in frozen and paraffin-embedded sections of rat brain, cerebellum and spinal cord, human and rat fetal CNS progenitor cell cultures and adult human paraffin-embedded brain.

Features:

- **Immunogen:** Peptide corresponding to the C-terminus (EAQGPK) of βIII tubulin.
- **Antibody Form:** Mouse monoclonal IgG$_1$ (clone 5G8), 1mg/ml in PBS containing no preservatives.
- **Specificity:** Cross-reacts with most mammalian species. Does not label nonneuronal cells (e.g., astrocytes).

Storage Conditions: Store at 4°C.



Immunostaining for βIII tubulin in rat cerebellum using Anti-βIII Tubulin mAb. Paraffin-embedded rat brain section double-immunofluorescence labeled with the primary antibody and detected using an anti-mouse Cy®3-conjugated secondary antibody (yellow). Nuclei were stained with DAPI (blue). Protocols developed and performed at Promega.

Anti-VAChT pAb

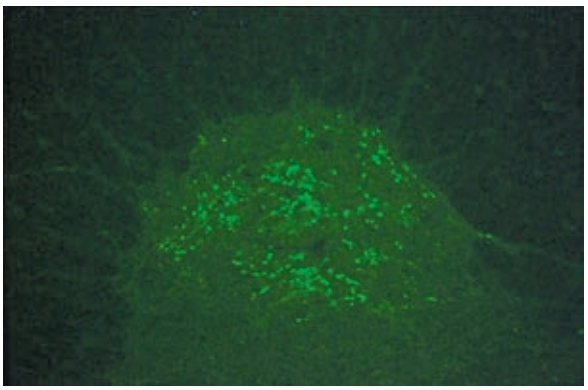
Product	Size	Cat.#
Anti-VAChT pAb	100 µg	G4481

Description: The purified Anti-VAChT (Vesicular Acetylcholine Transporter) pAb is raised in goats against a peptide (CSPPGPFDGCEDDYNYYSRS) corresponding to amino acids 511–530 of the carboxy terminus of the cloned rat VAChT. It is a novel tool to identify functional cholinergic neurons in the central and peripheral nervous system where the antibody stains fibers and neuronal cell bodies. This antibody has application for the study of the pathophysiology of neurodegenerative diseases involving the cholinergic system and for mapping cholinergic neurons in the nervous system.

Features:

- **Immunogen:** Carboxy-terminal peptide sequence 511–530 corresponding to cloned rat VAChT protein.
- **Antibody Form:** Purified goat polyclonal IgG; 0.5mg/ml in PBS containing no preservatives.
- **Specificity:** Cross-reacts with VAChT in rat and mouse, but not in human, guinea pig, rabbit or cat.

Storage Conditions: Store at –20°C.



2018TB

Immunofluorescence staining for VAChT protein in ventral gray matter of rat spinal cord. VAChT was detected using Anti-VAChT pAb at 1µg/ml followed by anti-goat Cy®3 conjugate. Specificity was demonstrated by preabsorption of Anti-VAChT pAb (1µg/ml) with the peptide immunogen (5µg/ml) prior to incubation with the primary antibody and anti-goat Cy®3 conjugate (data not shown). Protocols developed and performed at Promega.

Donkey Anti-Rabbit IgG (H+L) HRP, Anti-ACTIVE® Qualified

Product	Size	Cat.#
Donkey Anti-Rabbit IgG (H+L), HRP	60 µl	V7951
For Laboratory Use.		

Description: Donkey Anti-Rabbit Ig, (H+L), HRP is a horseradish peroxidase-conjugated secondary antibody specifically tested for use with the Anti-ACTIVE® antibodies. It is qualified for use in Western blot analysis using chemiluminescent and colorimetric detection methods. This antibody conjugate exhibits minimal cross-reactivity to goat, mouse and sheep IgG, bovine serum albumin (BSA) and proteins in mammalian cell extracts. This secondary antibody conjugate provides low background and highly specific signals when used at the recommended dilution with Anti-ACTIVE® MAPK, Anti-ACTIVE® JNK and Anti-ACTIVE® p38 pAbs. The conjugate is provided in phosphate-buffered saline containing BSA as a stabilizer and gentamicin as a preservative.

Features:

- **Sensitivity:** When conjugate is used in a Western blot at a 1:10,000 dilution along with Anti-ACTIVE® MAPK pAb (Cat.# V8031), it can detect active MAP kinase in 5µg of activated (nerve growth factor [NGF]-treated) PC12 cell extract using colorimetric detection.
- **Specificity:** Preferentially detects rabbit IgG with minimum reactivity with immunoglobulins from other species (including goat, sheep and mouse) or with bovine serum albumin and mammalian cell extract proteins.
- **Value:** 60µl per vial, sufficient to generate 300–600ml of Western blotting solution when used at the recommended dilution of 1:5,000 to 1:10,000.
- **Immunogen:** Intact rabbit IgG (H+L chains).
- **Antibody Form:** Donkey IgG, affinity-purified polyclonal antibody conjugated to horseradish peroxidase (HRP).

Protocol	Part#
Technical Bulletin	TB262

Storage Conditions: Store at –20°C.

Donkey Anti-Rabbit IgG (H+L) AP, Anti-ACTIVE® Qualified

Product	Size	Cat.#
Donkey Anti-Rabbit IgG (H+L), AP	60 µl	V7971
For Laboratory Use.		

Description: Donkey Anti-Rabbit IgG, (H+L), AP is alkaline phosphatase-conjugated secondary antibody specifically tested for use with the Anti-ACTIVE® antibodies. It is qualified for use in Western blot analysis using chemiluminescent and colorimetric detection methods. This antibody conjugate exhibits minimal cross-reactivity to goat, mouse and sheep IgG, bovine serum albumin (BSA) and proteins in mammalian cell extracts. This secondary antibody conjugate provides low background and highly specific signals when used at the recommended dilution with Anti-ACTIVE® MAPK, Anti-ACTIVE® JNK and Anti-ACTIVE® p38 pAbs. The conjugate is provided in 10mM sodium phosphate (pH 7.6), 0.25M NaCl containing 15mg/ml BSA as a stabilizer and 0.01% thimerosal and 0.02% gentamicin as preservatives.

Features:

- **Sensitivity:** When conjugate is used in a Western blot at a 1:10,000 dilution along with Anti-ACTIVE® MAPK pAb (Cat.# V8031), it can detect active MAP kinase in 5µg of activated (Nerve Growth Factor [NGF]-treated) PC12 cell extract using colorimetric detection.
- **Specificity:** Preferentially detects rabbit IgG with minimum reactivity with immunoglobulins from other species (including goat, sheep and mouse) or with bovine serum albumin and mammalian cell extract proteins.
- **Value:** Provided at 60µl per vial, sufficient to generate 300–600ml of Western blotting solution when used at the recommended dilution of 1:5,000 to 1:10,000.
- **Immunogen:** Intact rabbit IgG (H+L chains).
- **Antibody Form:** Donkey IgG, affinity-purified polyclonal antibody conjugated to alkaline phosphatase (AP).

Protocol	Part#
Technical Bulletin	TB262

Storage Conditions: Store at –20°C. When stored and handled properly, this product is stable for at least 6 months from date of purchase.

Alkaline Phosphatase-Conjugated Antibodies

Product	Size	Cat.#
Anti-Mouse IgG (H+L), AP Conjugate	100 µl	S3721
Anti-Rabbit IgG (Fc), AP Conjugate	100 µl	S3731
Anti-Human IgG (H+L), AP Conjugate	100 µl	S3821
Anti-Rat IgG (H+L), AP Conjugate	100 µl	S3831
Donkey Anti-Goat IgG, AP	60 µl	V1151

For Laboratory Use.

Description: The Alkaline Phosphatase-Conjugated Antibodies are used in the detection of proteins in Western blotting and ELISAs. The alkaline phosphatase (AP) catalyzes colorimetric reactions using Western Blue® Substrate (standard BCIP/NBT) or AttoPhos® Reagent. It can also drive the chemiluminescent detection reactions involving substrates such as 3-(2′-spiroadamantane)-4-methyl-4-(3′′ phosphoryloxyphenyl-1, 2-dioxetane (AMPPD®).

Goat Anti-Mouse IgG (H+L), Alkaline Phosphatase Conjugate is an affinity-purified goat anti-mouse antibody. It reacts with mouse IgG (all subclasses) as well as with light chains on other mouse immunoglobulins. Antibodies may cross-react with immunoglobulins from other species.

Goat Anti-Rabbit IgG (Fc), Alkaline Phosphatase Conjugate is an affinity-purified goat anti-rabbit antibody. It reacts with the heavy chains of rabbit IgG but not with light chains. Antibodies may cross-react with immunoglobulins from other species.

Goat Anti-Human IgG (H+L), Alkaline Phosphatase Conjugate is an affinity-purified goat anti-human antibody. This product has been isolated from antisera by immunoaffinity chromatography using immobilized antigens. It reacts with all subclasses of human IgG as well as with light chains on other human immunoglobulins. Antibodies may cross-react with immunoglobulins from other species. This product displays minimal cross-reactivity with horse and bovine serum proteins.

Goat Anti-Rat IgG (H+L), Alkaline Phosphatase Conjugate is an affinity-purified goat anti-rat antibody. This product has been isolated from antisera by immunoaffinity chromatography using immobilized antigens. It reacts with heavy and light chains of rat IgG. Antibodies may cross-react with immunoglobulins from other species.

Donkey Anti-Goat IgG, Alkaline Phosphatase Conjugate is a secondary antibody developed in donkeys against goat IgG; it has been affinity-purified and conjugated to alkaline phosphatase.

Horseradish Peroxidase-Conjugated Antibodies

Product	Size	Cat.#
Anti-Rabbit IgG (H+L), HRP Conjugate	300 µl	W4011
Anti-Mouse IgG (H+L), HRP Conjugate	300 µl	W4021
Anti-Human IgG (H+L), HRP Conjugate	300 µl	W4031
Anti-Chicken IgY, HRP Conjugate	300 µl	G1351
Donkey Anti-Goat IgG, HRP	60 µl	V8051

Cat.# W4011, W4021, W4031, V8051 For Laboratory Use.

Description: These high-quality antibodies are raised in goat, rabbit or donkey, affinity-purified and conjugated to horseradish peroxidase. The human, mouse and rabbit antibodies react with all IgG subclasses. All antibodies may cross-react with immunoglobulins of other species.

Goat Anti-Rabbit IgG (H+L), HRP Conjugate is an affinity-purified goat anti-rabbit antibody. This antibody has been isolated from antisera by immunoaffinity chromatography using immobilized antigens.

This anti-rabbit antibody reacts with heavy and light chains of rabbit IgG. A working dilution of 1:2,500 is suggested for Western blots and ELISAs.

Goat Anti-Mouse IgG (H+L), HRP Conjugate is an affinity-purified goat anti-mouse antibody. This antibody has been isolated from antisera by immunoaffinity chromatography using immobilized antigens.

This anti-mouse antibody reacts with all subclasses of mouse IgG as well as with light chains on other mouse immunoglobulins. A working dilution of 1:2,500 is suggested for Western blots and ELISAs.

Goat Anti-Human IgG (H+L), HRP Conjugate is an affinity-purified goat anti-human antibody. This product has been isolated from antisera by immunoaffinity chromatography using immobilized antigens.

This anti-human antibody reacts with all subclasses of human IgG as well as with light chains on other human immunoglobulins. A working dilution of 1:2,500 is suggested for Western blots and ELISAs.

Rabbit Anti-Chicken IgY, HRP Conjugate is a secondary antibody developed in rabbits against chicken IgY; it has been affinity-purified and conjugated to horseradish peroxidase. Anti-Chicken IgY, HRP Conjugate recognizes both the heavy and light chains of IgY and has been validated for use in Western blots, dot blots and ELISAs.

Donkey Anti-Goat IgG, HRP Conjugate is a secondary antibody developed in donkeys against goat IgG; it has been affinity-purified and conjugated to horseradish peroxidase. When used in a Western blot at a 1:10,000 dilution with colorimetric detection, Donkey Anti-Goat IgG, HRP Conjugate shows reactivity to goat and sheep IgG but minimal cross-reactivity to rabbit and mouse IgG and extracts of PC12 cells. It detects 100ng of goat IgG with minimal background signal.

Anti-Chicken IgY, Biotin Conjugate

Product	Size	Cat.#
Anti-Chicken IgY, Biotin Conjugate	500 µg	G2891

For Laboratory Use.

Description: Anti-Chicken IgY, Biotin Conjugate is developed in rabbits against chicken IgY; it is affinity-purified and conjugated to biotin. The antibody conjugate recognizes both the heavy and light chains of IgY.

Chicken IgY, Control Immunoglobulin

Product	Size	Cat.#
Chicken IgY, Control Immunoglobulin	1 mg	G1161

For Laboratory Use.

Description: Chicken IgY, Control Immunoglobulin is specific to chickens and is the counterpart to IgG from mammals. Chicken IgY may be used as a control antigen when performing Westerns, ELISAs and in vivo and in vitro immunohistochemical studies using anti-chicken IgY antibodies. The antibody is prepared by sequential precipitation, is chromatography-purified and is provided at 1mg/ml in PBS with no preservatives. The purity is >75%.

ECL Western Blotting Substrate

Product	Size	Cat.#
ECL Western Blotting Substrate	250 ml	W1001
	500 ml	W1015

Description: The ECL Western Blotting Substrate is a highly sensitive non-radioactive, enhanced luminol-based chemiluminescent substrate for the detection of horseradish peroxidase (HRP) on immunoblots. The ECL Western Blotting Substrate detects picogram amounts of antigen, and with the use of photographic or other imaging methods, visualizes the presence of HRP.

Features:

- **High Sensitivity:** Detect picogram levels of protein.
- **Save Time:** No optimization required; you can switch from other entry-level ECL substrates.

Protocol	Part#
Technical Manual	TM317

Storage Conditions: Store at 2–8°C.

TMB One Solution

Product	Size	Cat.#
TMB One Solution	100 ml	G7431

For Laboratory Use.

Description: TMB One Solution is a chromagen substrate, 3,3´,5,5´-tetramethylbenzidine (TMB) provided in a mildly acidic, nonhazardous buffer for horseradish peroxidase detection in an ELISA format. The substrate develops a blue reaction product when oxidized by peroxidase and a yellow reaction product in an endpoint multiwell assay after the addition of an acid solution provided by the end user.

Features:

- **Convenient:** Single solution provided ready-to-use; just add, incubate, stop and read. This homogeneous reagent improves assay variation.
- **Stable:** Stable for 12 months at 4°C, providing extended shelf life; the assay end product is stable for at least one hour after stopping the assay.
- **Safe:** Provided in a slightly acidic, nonhazardous proprietary buffer without aprotic solvents; noncaustic to plastics used in automated systems.
- **Sensitive:** Low background provides greater assay sensitivity.

Storage Conditions: Store at 4°C protected from light.

AttoPhos® AP Fluorescent Substrate System

Product	Size	Cat.#
AttoPhos® AP Fluorescent Substrate System	3 × 36 mg	S1000
AttoPhos® AP Fluorescent Substrate System Trial Size	1 × 36 mg	S1001
Available Separately	**Size**	**Cat.#**
AttoPhos® Substrate	36 mg	S1011
	100 mg	S1012
	1 g	S1013
AttoPhos® Buffer	60 ml	S1021
	240 ml	S1022

Description: AttoPhos® AP Fluorescent Substrate System contains a highly sensitive fluorescent alkaline phosphatase (AP) substrate.

Features:

- **Sensitivity:** Low fluorescence signal until enzymatically acted upon, yielding detection of AP to 0.1 attomole.
- **Low Background:** Low fluorescence from interfering biological molecules.
- **Linearity:** Linear kinetics over five orders of magnitude of AP concentration.
- **Additional Features:** Excitation at 435nm, emission at 555nm and large Stokes' shift (≈120nm).

Protocol	Part#
Technical Bulletin	TB280

Storage Conditions: Store at 4°C.

Blocking Agents

Product	Size	Cat.#
Blot-Qualified BSA	10 g	W3841
Tween® 20	2.5 ml	W3831
E. coli Extract for Background Reduction	2 ml	S3761

For Laboratory Use.

Description: This BSA (bovine serum albumin) has been tested and qualified for optimum performance in immunoblotting applications with alkaline phosphatase antibody conjugates. It is shown to be alkaline phosphatase-free.

Tween® 20 is a nonionic detergent used as a buffer component for immunoscreening in the ProtoBlot® Systems. In addition to blocking agents such as BSA, which saturate excess sites of antibody binding on membranes, this detergent actsin solution to dissociate nonspecific interactions with an antibody probe.

E. coli extract is used for background reduction for Western blotting in cases where primary antisera contain cross-reacting nonspecific antibodies.

ProtoBlot® II AP Systems with Stabilized Substrate and *Western Express*® Fast Blotting Protocol

Product	Size	Cat.#
ProtoBlot® II AP System with Stabilized Substrate, Human	1 each	W3940
ProtoBlot® II AP System with Stabilized Substrate, Mouse	1 each	W3950
ProtoBlot® II AP System with Stabilized Substrate, Rabbit	1 each	W3960

For Laboratory Use.

Description: The ProtoBlot® II AP Systems with Stabilized Substrate are designed for the rapid and sensitive detection of proteins or other macromolecular antigens immobilized on nitrocellulose or PVDF membranes. Proteins can be transferred from gels after electrophoresis (Western blots) or bound directly from solution ("dot" blots). The *Western Express*® Fast Blotting Protocol is included with the system and can reduce dramatically the time required to perform immunodetection. All ProtoBlot® II AP Systems contain BSA as a stabilizer and 0.05% sodium azide as a preservative.

Features:

- **Fast:** Easy-to-use *Western Express*® Protocol allows the detection of dot blots in 30–45 minutes and the detection of Western blots in 1–2 hours.
- **Convenient:** The system contains Western Blue® Stabilized Substrate for AP, which is a ready-to-use solution of BCIP/NBT. No reagent preparation is required for the substrate.

For many applications, AP conjugates are superior to HRP conjugates because they:

- offer greater sensitivity (tenfold) of detection.
- are not inhibited by azide.
- use a substrate that is not prone to fading during long-term storage.
- have protocols provided for both PVDF and nitrocellulose membranes.

Protocol	Part#
Technical Manual	TM026

Storage Conditions: Store antibody conjugates at 4°C (undiluted). Store Western Blue® Substrate at room temperature.

EGGstract® IgY Purification System

Product	Size	Cat.#
EGGstract® IgY Purification System	6 isolations	G2610
	25 isolations	G1531

Description: The EGGstract® IgY Purification System provides an easy and rapid method for the isolation of immunoglobulins (IgY antibodies) from egg yolks. Because a single egg contains as much antibody as an average bleed from a rabbit, this represents an appealing alternative to the generation of pAb in rabbits. Additionally, IgY can be produced in chickens against certain biological antigens that fail to elicit a humoral response in rabbits or other mammals due to species relatedness.

The procedure is very simple and requires less than one hour to complete. To use the system in purification of IgY antibodies from eggs, simply separate the yolks from immunized chicken eggs and dilute the yolks into Precipitation Solution A. After mixing, remove the lipids by centrifugation, and precipitate the IgY using Precipitation Solution B. The solution is centrifuged, and the pellet obtained from solution contains the IgY antibody.

Features:

- **Easy:** No cumbersome chromatography or dialysis.
- **Fast:** IgY purified in 60 minutes.
- **Highly Pure:** Approximately 75% pure. Approximately 90% pure with an additional 15-minute precipitation.
- **High Yield:** 2–5mg IgY per 1ml of egg yolk.

Protocol	Part#
Technical Bulletin	TB188

Storage Conditions: Store at room temperature.

Industrial and Environmental Monitoring

Microbial Detection and Quantitation 184

Protein Degradation Detection 184

ENLITEN® ATP Assay System

Product	Size	Cat.#
ENLITEN® ATP Assay System	100 assays	FF2000

Description: The ENLITEN® ATP Assay System can be used to measure ATP levels for the indirect detection of biocontamination on food processing surfaces, in cosmetics and beverages or to assay for enzymes that degrade ATP and to quantitate ATP in biological fluids.

Features:

- **Less Variation:** Stable light output.
- **User Friendly:** Easy-to-prepare reagents.
- **Performance:** Fast and convenient assay method.
- **Sensitive:** Detects as little as 10^{-15} moles of ATP.

Protocol	Part#
Technical Bulletin	TB267

Storage Conditions: Store at –20°C unopened. See product insert for individual component storage conditions before and after opening.

ENLITEN® Total ATP Rapid Biocontamination Detection Kit

Product	Size	Cat.#
ENLITEN® Total ATP Rapid Biocontamination Detection Kit	100 assays	FF3710

Description: The ENLITEN® Total ATP Rapid Biocontamination Detection Kit is intended to test surfaces for biocontamination by detecting ATP that may result from food residue, microbes or other biological sources. Using bioluminescence technology, the kit can quickly measure the presence of ATP as an indicator of cleaning efficacy. This kit can be used with most commercial luminometers.

Features:

- **Sensitive:** Measures total ATP.
- **User Friendly:** Easy-to-perform procedure.
- **Versatile:** Compatible with most commercial luminometers.
- **Reproducible:** Recombinant Luciferase provides better reproducibility.

Protocol	Part#
Technical Bulletin	TB265

Storage Conditions: Store at –20°C unopened.

ENLITEN® rLuciferase/Luciferin Reagent

Product	Size	Cat.#
ENLITEN® rLuciferase/Luciferin Reagent	100 assays	FF2021

Description: The ENLITEN® rLuciferase/Luciferin Reagent is intended for the rapid and quantitative detection of ATP in liquid samples. The reagent is designed to measure 10^{-11} to 10^{-15} moles of ATP. Some of the applications may include the indirect measurement of bacteria, yeasts and fungi on surfaces or in products, assaying enzymes that degrade ATP or quantitation of ATP in biological fluids.

Features:

- **Less Variation:** Stable light output.
- **User Friendly:** Easy-to-prepare reagents.
- **Performance:** Fast and convenient assay method.
- **Sensitive:** Detects as little as 10^{-15} moles of ATP.

Protocol	Part#
Technical Bulletin	TB268

Storage Conditions: Store at –20°C.

ISOQUANT® Isoaspartate Detection Kit

Product	Size	Cat.#
ISOQUANT® Isoaspartate Detection Kit	100 assays	MA1010

Description: The ISOQUANT® Isoaspartate Detection Kit is intended for quantitative detection of isoaspartic acid residues in proteins and peptides, which can result from the gradual, nonenzymatic deamidation of asparagine or rearrangement of aspartic acid residues during storage or handling. Because the kit does not depend on the monitoring of charge differences for detection, charge heterogeneity does not interfere with the assay. The ISOQUANT® Kit can be used on peptides or proteins such as monoclonal antibodies.

Features:

- **Great Efficiency:** Simple procedure with a test time of less than one hour. Automation possible with HPLC autosampler capability.
- **Economical:** HPLC detection eliminates cost and inconvenience of radioactive materials handling.
- **Analytical:** Quantitative results available.
- **Versatile:** Perform individual samples or batches. Small sample size makes the assay suitable for research, analytical methods, formulations and process development work.
- **Robust:** Not affected by common buffer components.
- **HPLC Detection Method:** Fits with existing equipment and expertise.
- **Sensitive:** Detects isoaspartate resulting from aspartic acid rearrangement as well as deamidation of asparagine.

Protocol	Part#
Technical Bulletin	TBI001

Storage Conditions: Store at –20°C.

Promega

11 In Vitro Transcription and Transcription Regulation

RNA Production Systems	**186**
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RiboMAX™ Large Scale RNA Production Systems

Product	Size	Cat.#
RiboMAX™ Large Scale RNA Production System—SP6	1 system	P1280
RiboMAX™ Large Scale RNA Production System—T7	1 system	P1300

For Laboratory Use.

Description: The RiboMAX™ Large Scale RNA Production Systems consistently produce 2–5mg/ml of RNA in a 1ml reaction, about 10- to 20-fold more RNA than is produced with the standard Riboprobe® System transcription reaction. The RiboMAX™ System reactions differ from those of the Riboprobe® Systems in three primary ways: a HEPES (pH 7.5) buffer is used rather than a Tris-HCl (pH 7.9) buffer; rNTP and magnesium concentrations are elevated at levels appropriate for either SP6 or T7 RNA polymerase; and inorganic pyrophosphatase is included in the reaction.

RNAs synthesized with the RiboMAX™ System perform better for in vitro translation in rabbit reticulocyte translation systems than RNA synthesized by standard methods. The reduction of components inhibitory to translation may be advantageous for other applications requiring biologically active RNA. Because the RiboMAX™ Systems produce large quantities of RNA, these systems are not recommended for the generation of high-specific-activity RNA probes.

Features:

- **Flexible:** Systems are available for use with SP6 and T7 RNA polymerases.
- **Scalable:** Reactions can be scaled up or down to suit varying RNA production requirements.
- **High-Quality:** Synthesis of enhanced, translation-grade RNA.

Protocol	Part#
Technical Bulletin	TB166

Storage Conditions: Store at –20°C.

T7 RiboMAX™ Express Large Scale RNA Production System

Product	Size	Cat.#
T7 RiboMAX™ Express Large Scale RNA Production System	1 system	P1320

For Laboratory Use.

Description: The T7 RiboMAX™ Express Large Scale RNA Production System is an in vitro transcription system designed for the consistent production of milligram amounts of RNA in a short amount of time. Due to optimization of the enzyme mix and transcription buffer, yields of 5–8.5mg/ml are generated in 30 minutes, compared to 2–4 hours with other commercially available systems. To minimize pipetting steps and errors, the 2X transcription buffer includes all four rNTPs. In addition, the system includes RQ1 RNase-Free DNase for the removal of plasmid template after transcription.

Due to the combined 2X buffer and rNTPs, the T7 RiboMAX™ Express System is not recommended for the synthesis of RNA for applications that require capped RNA. For synthesis of capped RNA, please order the standard RiboMAX™ Large Scale RNA Production System—T7 (Cat.# P1300).

Features:

- **Fast:** The T7 RiboMAX™ Express System produces milligram amounts of RNA in as little as 30 minutes rather than 2–4 hours as with other commercially available systems.
- **Convenient:** The four rNTPs and 2X transcription buffer have been combined, thus minimizing pipetting errors and setup time.
- **Flexible:** Efficiently transcribes DNA templates of varying sizes. Works with transcripts as short as 21bp.

Protocol	Part#
Technical Bulletin	TB298

Storage Conditions: Store at –20°C.

Riboprobe® Systems

Product	Size	Cat.#
Riboprobe® System—SP6	1 system	P1420
Riboprobe® System—T3	1 system	P1430
Riboprobe® System—T7	1 system	P1440

Cat.# P1430, P1440 For Laboratory Use.

Description: The Riboprobe® Systems are designed for in vitro preparation of high-specific-activity single-stranded RNA probes or microgram quantities of defined RNA transcripts from cloned DNA inserts. These systems contain all components necessary for in vitro transcription from a DNA template (excluding the radioisotope) and also contain RQ1 RNase-Free DNase (Cat.# M6101) for template removal following transcription.

Features:

- **Specific:** SP6, T7 and T3 RNA Polymerases are extremely promoter-specific, allowing production of virtually homogeneous RNA using plasmid DNA as a template.
- **Choice of Enzyme:** Systems available with SP6 RNA Polymerase, T7 RNA Polymerase or T3 RNA Polymerase.
- **Convenient:** Includes positive control template for use with SP6, T7 or T3 RNA Polymerase, DNase I for removal of DNA template and Recombinant RNasin® Ribonuclease Inhibitor.

Protocol	Part#
Technical Manual	TM016

Storage Conditions: Store at –20°C.

Riboprobe® Combination Systems

Product	Size	Cat.#
Riboprobe® Combination System—T3/T7 RNA Polymerase	1 system	P1450
Riboprobe® Combination System—SP6/ T7 RNA Polymerase	1 system	P1460

For Laboratory Use.

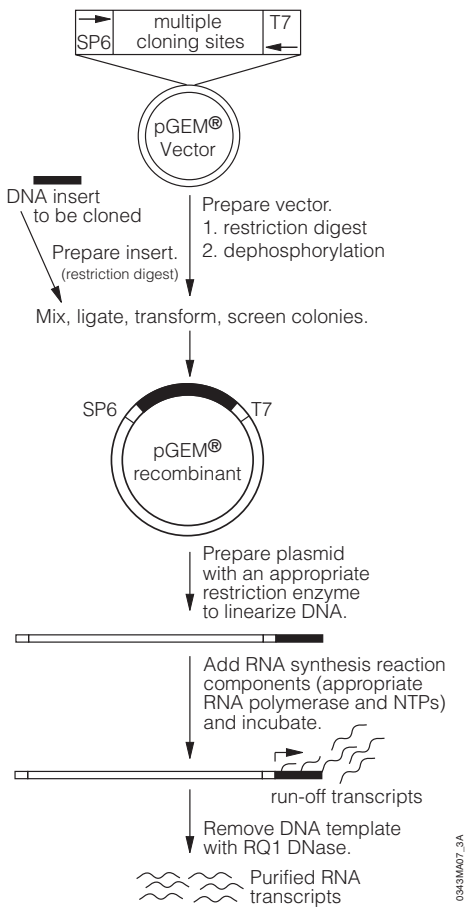
Description: The Riboprobe® Combination Systems are designed for in vitro preparation of high-specific-activity single-stranded RNA probes or microgram quantities of defined RNA transcripts from cloned DNA inserts. The Riboprobe® Combination Systems include the RNA polymerases, all of the required reagents (excluding radioisotope) for performing transcription reactions in vitro and RQ1 RNase-Free DNase (Cat.# M6101) for removal of the template following transcription.

Features:

- **Flexible:** Allows synthesis of RNA corresponding to either the coding or noncoding strand of cloned DNA from a single plasmid construct.
- **Specific:** SP6, T7 and T3 RNA Polymerases are extremely promoter-specific, allowing production of virtually homogeneous RNA using plasmid DNA as a template.
- **Convenient:** Includes positive control template for use with T7, T3 or SP6 RNA polymerase, DNase I for removal of DNA template and Recombinant RNasin® Ribonuclease Inhibitor.

Protocol	Part#
Technical Manual	TM016

Storage Conditions: Store at –20°C.



Schematic diagram of the Riboprobe® Combination Systems.

Riboprobe® System Components and Buffers

Product	Size	Conc.	Cat.#
Riboprobe® System Buffers	1 system	–	P1121
rATP, rCTP, rGTP, rUTP, each at 10mM in separate tubes	0.5 ml	10 mM	P1221
Available Separately	**Size**	**Conc.**	**Cat.#**
RQ1 RNase-Free DNase	1,000 u	1 u/µl	M6101
rATP, 10mM	0.5 ml	10 mM	P1132
rCTP, 10mM	0.5 ml	10 mM	P1142
rGTP, 10mM	0.5 ml	10 mM	P1152
rUTP, 10mM	0.5 ml	10 mM	P1162
DTT, Molecular Grade	100 µl	100 mM	P1171
Transcription Optimized 5X Buffer	200 µl	–	P1181
Nuclease-Free Water	50ml (2 × 25 ml)		P1193

For Laboratory Use.

Description: Riboprobe® System Buffers are components of the single and combination Riboprobe® Systems. The buffers are also available as standalone products.

RQ1 RNase-Free DNase is used to remove template DNA from RNA preparations and is qualified for use in applications where maintaining the integrity of RNA is critical. Product is quality tested to ensure the absence of detectable RNase activity. 10X Reaction Buffer and 10X Stop Buffer included.

rATP, rCTP, rGTP and rUTP are provided in individual tubes, qualified for use with the Riboprobe® Systems. The rNTPs are supplied in nuclease-free water. Purity has been verified by HPLC analysis.

Features:

- **Pretested:** Reagents are tested with other Riboprobe® System components. rNTPs are tested for functionality with in vitro transcription reactions.
- **Transcription Qualified:** Reagents are qualified for use for in vitro transcription reactions with SP6, T7 or T3 RNA Polymerase.

Protocol	Part#
Technical Manual	TM016

Storage Conditions: Store at –20°C.

Ribo m^{7}G Cap Analog

Product	Size	Conc.	Cat.#
Ribo m^{7}G Cap Analog	10 A_{254} units	40 mM	P1711
	25 A_{254} units	40 mM	P1712

Description: The Ribo m^{7}G Cap Analog is a modified ribonucleotide with the structure (m^{7}G(5′)ppp(5′)G). This methylated ribonucleotide can be incorporated onto the 5′-end of transcripts synthesized in vitro and simulates the 7-methyl guanosine 5′-cap structure found on most eukaryotic mRNA molecules.

Features:

- **Improved Translation:** Enhances translation efficiency in many reticulocyte-based reactions.
- **Effective:** Protects RNA from intracellular digestion.
- **Flexible:** Can be used in either the Riboprobe® Systems or RiboMAX™ Large Scale RNA Production Systems.

Protocol	Part#
Promega Product Information	9PIP171

Storage Conditions: Store at –20°C.

pGEM® Express Positive Control Template

Product	Size	Cat.#
pGEM® Express Positive Control Template	10µg (2 × 5 µg)	P2561

Description: The pGEM® Express Positive Control Template is created by linearizing a vector with the restriction enzyme ScaI. The Positive Control Template may be used to monitor in vitro transcription reactions when using the Riboprobe® Systems.

Features:

- **Multi-Sized RNAs:** SP6 RNA polymerase produces transcripts of 1,787 and 2,566 bases; T7 RNA polymerase produces transcripts of 1,065 and 2,346 bases; T3 RNA Polymerase produces transcripts of 250 and 1,525 bases.
- **Flexible:** Template can be used with SP6, T7 or T3 RNA polymerases.

Protocol	Part#
Promega Product Information	9PIP256

Storage Conditions: Store at –20°C.

AP1, (c-Jun), Human, Recombinant

Product	Size	Cat.#
rhAP1 (c-Jun)	50 fpu	E3061

1fpu = the amount of protein required to yield a complete footprint on SV40 Early Promoter DNA.

Description: c-Jun is the protein encoded by the human proto-oncogene, *c-jun*. Its expression is greatly modulated by extracellular stimuli such as phorbol ester and growth factor treatment. It is a member of the AP1 family. In many cell types, AP1 activity is composed primarily of Jun and Fos heterodimers and secondarily of Jun homodimers as in HeLa cells. Jun homodimers bind AP1 sites in vitro and activate transcription. c-Jun is expressed in *E. coli* from a human cDNA clone and has a molecular weight of 40kDa.

Features:

- **Performance-Tested:** Tested in gel-shift and footprinting applications.

Storage Conditions: Store at –70°C.

NF-κB, Human, Recombinant

Product	Size	Cat.#
rhNF-κB (p50)	50 gsu	E3770

1gsu = the amount of protein required to gel shift the NF-κB oligonucleotide under defined conditions.

Description: NF-κB (p50), Human, Recombinant is expressed in bacteria from a human cDNA derived from the full-length p105. p50, a processed product of its p105 precursor, is the first DNA-binding subunit of the NF-κB transcription factor described. The amino-terminal half of p50 has strong homology to the proto-oncogene, *c-rel*, its viral counterpart, *v-rel*, and the *Drosophila dorsal* gene.

p50 forms a transcriptionally active heterodimer with NF-κB subunit p65. The p50/p65 heterodimer is subject to regulation by physical sequestration in the cytoplasm by I-κB.

Features:

- **Performance-Tested:** Tested in gel shift assays.

Storage Conditions: Store at –70°C.

TFIIB, Human, Recombinant

Product	Size	Cat.#
rhTFIIB	50 gsu	E3790

1gsu = the amount of protein required to gel shift the TFIID oligonucleotide in the presence of rhTFIID under defined conditions.

Description: rhTFIIB is a general transcription factor involved in formation of an active complex in vitro capable of specifically initiating RNA synthesis by RNA polymerase II. An early stage of initiation complex assembly involves the formation of a D-B or D-A-B complex, which consists of TFIID, TFIIB (TFIIA) and the promoter DNA. The stability of the D-B and D-A-B complexes is thought to be greater than that of TFIID and DNA alone. The full-length human cDNA for TFIIB is expressed in *E. coli* and has a molecular weight of 32kDa. TFIIB alone does not have DNA-binding activity.

Features:

- **Performance-Tested:** Tested by gel shift assay for the formation of the D-B complex. Tested for in vitro transcriptional activity.

Storage Conditions: Store at –70°C.

Transcription Factor Consensus Oligonucleotides

Product	Size	Conc.	Cat.#
AP1 Consensus Oligonucleotide	175 pmol	1.75 pmol/µl	E3201
	35 pmol	1.75 pmol/µl	E3202
AP2 Consensus Oligonucleotide	175 pmol	1.75 pmol/µl	E3211
	35 pmol	1.75 pmol/µl	E3212
CREB Consensus Oligonucleotide	175 pmol	1.75 pmol/µl	E3281
	35 pmol	1.75 pmol/µl	E3282
NF-κB Consensus Oligonucleotide	175 pmol	1.75 pmol/µl	E3291
	35 pmol	1.75 pmol/µl	E3292
OCT1 Consensus Oligonucleotide	175 pmol	1.75 pmol/µl	E3241
	35 pmol	1.75 pmol/µl	E3242
SP1 Consensus Oligonucleotide	175 pmol	1.75 pmol/µl	E3231
	35 pmol	1.75 pmol/µl	E3232
TFIID Consensus Oligonucleotide	175 pmol	1.75 pmol/µl	E3221
	35 pmol	1.75 pmol/µl	E3222

Description: The electrophoretic mobility shift assay (EMSA, gel shift, gel retardation) is a relatively simple and sensitive method to investigate protein:DNA interactions. These oligonucleotides contain consensus DNA-binding sites for individual sequence-specific transcription factors. The double-stranded oligonucleotides are designed with 5′ OH blunt ends, making them easily labeled to high specific activity with T4 polynucleotide kinase.

Protocol	Part#
Technical Bulletin	TB110

Storage Conditions: Store at –20°C.

Characteristics of the Consensus Oligonucleotides and Binding Proteins.

AP1 (c-jun) 5′-CGC TTG ATG AGT CAG CCG GAA-3′
3′-GCG AAC TAC TCA GTC GGC CTT-5′

Forms DNA binding dimers with other members of the AP1 family and with Fos through leucine zipper formation.

AP2 5′-GAT CGA ACT GAC CGC CCG CGG CCC GT-3′
3′-CTA GCT TGA CTG GCG GGC GCC GGG CA-5′

May act independently as both a TPA- and cAMP-inducible element and can be specifically inhibited by large T antigen.

CREB 5′-AGA GAT TGC CTG ACG TCA GAG AGC TAG-3′
3′-TCT CTA ACG GAC TGC AGT CTC TCG ATC-5′

Confers responsiveness to cAMP; it contains a leucine zipper motif for dimerization, and the associated basic domain is homologous to c-Jun DNA binding domains.

NF-κB 5′-AGT TGA GGG GAC TTT CCC AGG C-3′
3′-TCA ACT CCC CTG AAA GGG TCC G-5′

Binds to κ light chain enhancer in B cells and is present in a covert cytoplasmic form in non-B cells.

OCT1 5′-TGT CGA ATG CAA ATC ACT AGA A-3′
3′-ACA GCT TAC GTT TAG TGA TCT T-5′

A member of the OCT family, which is apparently ubiquitous in mammalian cells, the bipartite POU domain includes the POU-box and the homeo domain.

SP1 5′-ATT CGA TCG GGG CGG GGC GAG C-3′
3′-TAA GCT AGC CCC GCC CCG CTC G-5′

O-glycosylated transcription factor with sequence specificity conferred through three zinc fingers in the DNA binding domain.

TFIID 5′-GCA GAG CAT ATA AGG TGA GGT AGG A-3′
3′-CGT CTC GTA TAT TCC ACT CCA TCC T-5′

A general transcription factor that exhibits specific DNA binding to the TATA box. This factor is associated with RNA polymerase I, II and III activities.

9491LA

HeLaScribe® Nuclear Extract in vitro Transcription System

Product	Size	Cat.#
HeLaScribe® Nuclear Extract in vitro Transcription System	40 reactions	E3110

Description: The most well characterized cell-free system for in vitro transcription of eukaryotic genes is derived from HeLa cell nuclei. HeLa nuclear extracts can support accurate transcription initiation by RNA polymerase II and exhibit both basal and regulated patterns of RNA polymerase transcription. The nuclear extract is also a source for a variety of transcription factors, DNA-binding proteins and the enzymatic machinery involved in RNA processing.

The HeLa Nuclear Extract included in the HeLaScribe® Nuclear Extract in vitro Transcription System is prepared by a modification of the method of Dignam *et al.* Extracts prepared by this method have been shown to allow transcription from the human transferrin gene promoter and the adenovirus 2 major late promoter. The system also includes all of the necessary components for in vitro transcription as well as a positive control template (CMV immediate early promoter DNA).

Features:

- **Peformance-Tested:** Tested with cytomegalovirus immediate early gene (CMV) promoter.
- **Convenient:** Available as a complete transcription system or extract alone.
- **Positive Control:** System contains a CMV promoter-positive control template.

Protocol	Part#
Technical Bulletin	TB123

Storage Conditions: Store at –70°C. Avoid multiple freeze-thaw cycles of the extract.

In vitro Transcription Systems Related Products

Product	Size	Cat.#
HeLaScribe® Nuclear Extract in vitro Transcription Grade	40 reactions	E3091
	160 reactions	E3092
Available Separately	**Size**	**Cat.#**
HeLaScribe® Nuclear Extract Positive Control DNA	300 ng	E3621
rCTP, rATP, rUTP, rGTP, 100mM each	4 × 400 µl	E6000
rATP, 100mM	400 µl	E6011
rUTP, 100mM	400 µl	E6021
rGTP, 100mM	400 µl	E6031
rCTP, 100mM	400 µl	E6041

Cat.# E6000, E6011, E6021, E6031, E6041 For Laboratory Use.

Description: HeLaScribe® Nuclear Extract, in vitro Transcription Grade, derived from HeLa cell nuclei, provides a cell-free system for in vitro transcription of eukaryotic genes.

Protocol	Part#
Technical Bulletin	TB123

Storage Conditions: Store HeLaScribe® Nuclear Extracts at –70°C. Store other components at –20°C.

Primer Extension System—AMV Reverse Transcriptase

Product	Size	Cat.#
Primer Extension System—AMV Reverse Transcriptase	40 reactions	E3030

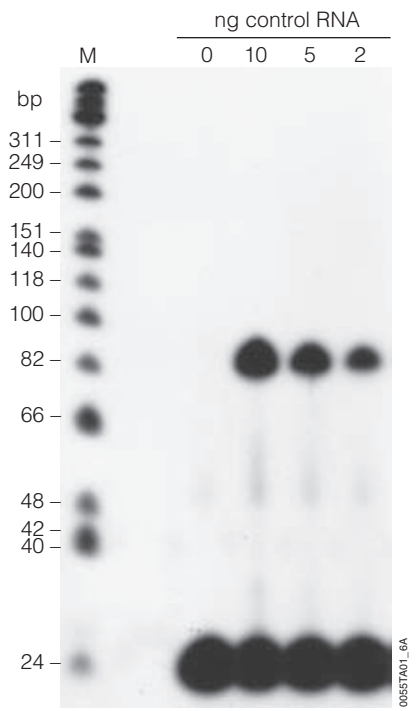
Description: Primer Extension System—AMV Reverse Transcriptase can be used to quantitate specific mRNA transcripts and map the start sites of transcription. An end-labeled oligonucleotide is hybridized to RNA and is used as a primer by reverse transcriptase in the presence of deoxynucleotides. The RNA is thus reverse transcribed into cDNA and is analyzed on a denaturing polyacrylamide gel. The length of the cDNA reflects the number of bases between the labeled nucleotide of the primer and the 5′-end of the RNA; the quantity of cDNA product is related to the amount of targeted RNA.

Features:

- **Convenient:** System includes control RNA and primer as well as size markers ready for phosphorylation with T4 Polynucleotide Kinase.

Protocol	Part#
Technical Bulletin	TB113

Storage Conditions: All components must be stored at –20°C, except for the control RNA, which must be stored at –70°C.



Gel analysis of ^{32}P-labeled ϕX174 DNA/HinfI markers and control RNA primer extension products produced using the Primer Extension System— AMV Reverse Trancriptase (Cat.# E3030).

Gel Shift Assay Systems

Product	Size	Cat.#
Gel Shift Assay Core System	100 reactions	E3050
Gel Shift Assay System	100 reactions	E3300
Available Separately	**Size**	**Cat.#**
HeLaScribe® Nuclear Extract, Gel Shift Assay Grade	3 × 40 µl	E3521
Gel Shift Binding 5X Buffer	5 × 200 µl	E3581

Description: The gel shift or electrophoretic mobility shift assay provides a simple and rapid method for detecting DNA-binding proteins. This method is widely used to study sequence-specific DNA-binding proteins such as transcription factors. The assay is based on the observation that complexes of protein and DNA migrate through a nondenaturing polyacrylamide gel more slowly than free DNA fragments or double-stranded oligonucleotides. The gel shift assay is performed by incubating a purified protein or a complex mixture of proteins (such as nuclear or cell extract preparations) with a ^{32}P end-labeled DNA fragment containing the putative protein binding site. The reaction products are then analyzed on a nondenaturing polyacrylamide gel. The specificity of the DNA-binding protein for the putative binding site is established by competition experiments using unlabeled DNA fragments or oligonucleotides containing a binding site for the protein of interest or other unrelated DNA sequences.

The Core System (Cat.# E3050) includes HeLa Nuclear Extract and SP1 and AP2 Consensus Oligos that can be used as positive controls and serve as a reliable system for obtaining experience with gel shift assays. In addition, the Core System contains T4 Polynucleotide Kinase and Kinase 10X Buffer for labeling oligonucleotides as well as Gel Shift Binding 5X Buffer. Cat.# E3300 contains all of the above plus consensus oligos for AP1, OCT1, CREB, NF-κB, and TFIID.

Features:

- **Positive Controls:** The Gel Shift Assay Core System includes a HeLa Nuclear Extract and consensus oligonucleotides for AP2 and SP1.
- **Versatile:** Oligonucleotides can be 5′ end-labeled and used as protein-specific probes or used as unlabeled oligonucleotides in competition assays.

Protocol	Part#
Technical Bulletin	TB110

Storage Conditions: Store HeLa Nuclear Extract at –70°C. Store other components at –20°C.

Microarrays

Microarray Analysis 192

ChipShot™ Labeling and Clean-Up Systems

Product	Size	Cat.#
ChipShot™ Indirect Labeling and Clean-Up System	25 reactions	Z4000
ChipShot™ Direct Labeling and Clean-Up System	25 reactions	Z4100

Description: The ChipShot™ Labeling and Clean-Up Systems enable researchers to generate consistent, reproducible and sensitive microarray data. The ChipShot™ Systems make it easy for researchers to perform RNA and microarray manipulation, regardless of their experience. The protocols are simple, time-tested and easy-to-follow.

The **ChipShot™ Direct System** provides quality reagents and enzymes for generating fluorescently labeled cDNA via direct incorporation of fluorescently modified nucleotides. Labeled cDNA then can be hybridized to a printed microarray.

The **ChipShot™ Indirect System** provides quality reagents and enzymes for generating fluorescently labeled cDNA using a two-step labeling process that includes synthesis of aminoallyl-modified cDNA, followed by conjugation to the CyDye™ NHS ester. Labeled cDNA then can be hybridized to a printed microarray.

Note: Customers must supply CyDye™ nucleotides or NHS esters.

Features:

ChipShot™ Systems

- **Conserve Sample:** Optimized protocols for 5µg of total RNA or 1.5µg of poly(A)+ RNA per labeling reaction.
- **Rely On Quality Products:** Quality-controlled reagents are always available from Promega.
- **Simplify Your Process:** Easy-to-follow protocols.
- **Increase Productivity:** No hybridization reagent preparation and testing.
- **Count On Technical Support:** Outstanding technical support from Promega for the researcher.

ChipShot™ Direct System

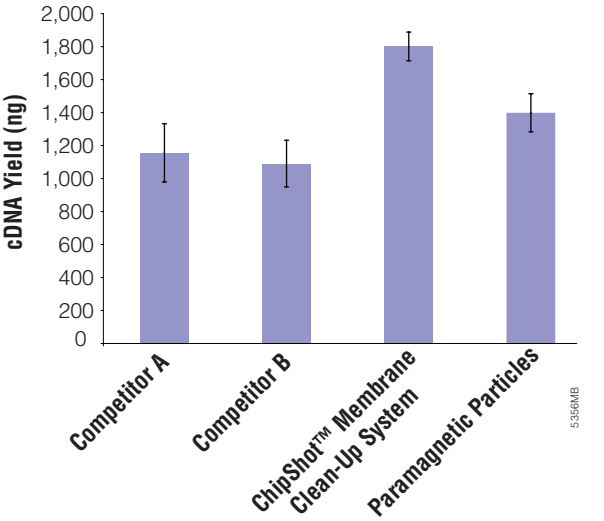
- **Increase Productivity:** Go from RNA to purified, labeled cDNA in approximately 3 hours.
- **Get Reliable Results:** Separate protocols for CyDye™3 and CyDye™5 labeling, so dye swap experiments are not necessary.

ChipShot™ Indirect System

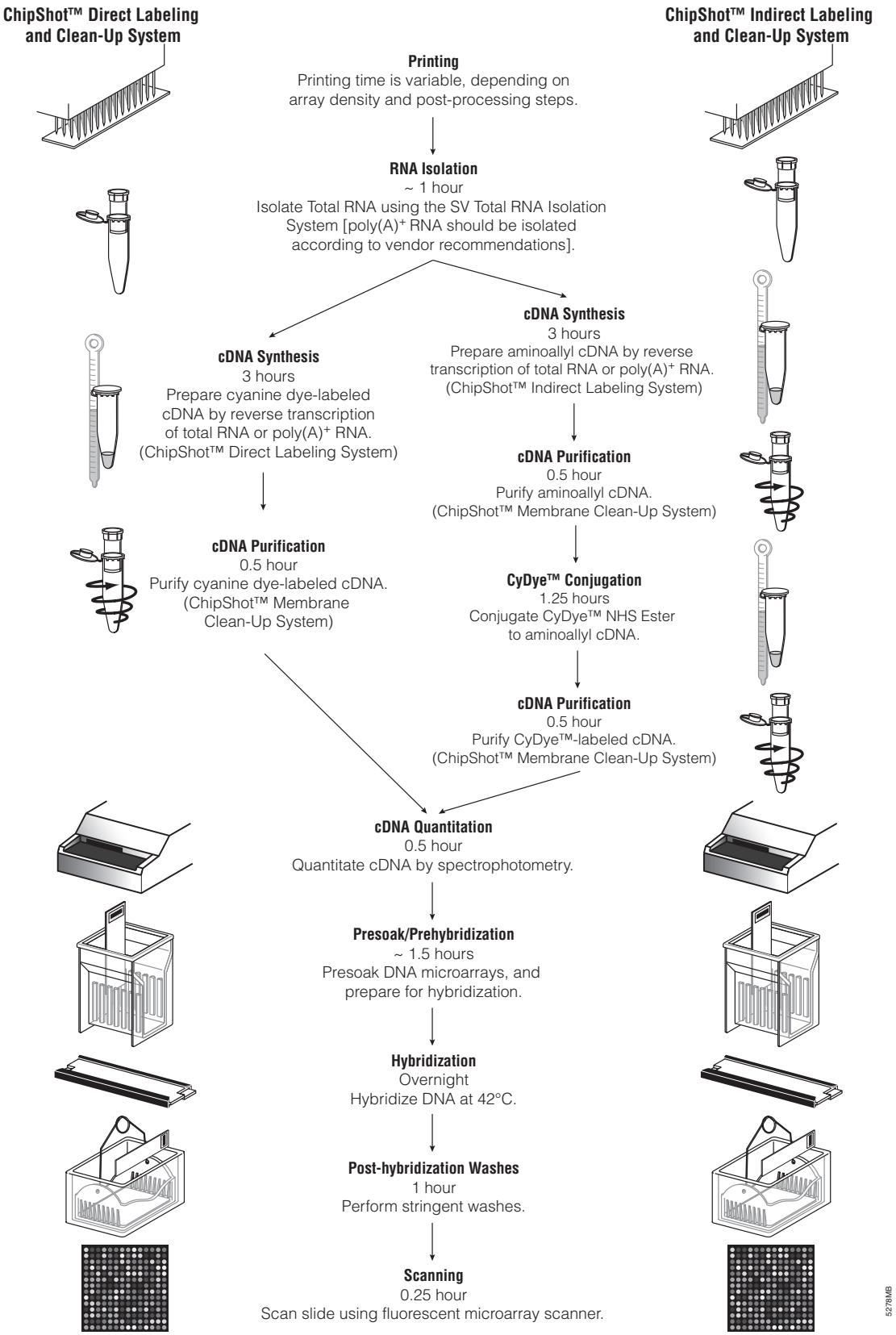
- **Achieve Higher Yields:** Efficient incorporation of aminoallyl dNTP by reverse transcriptase yields more cDNA for labeling.
- **Get Unbiased Dye Incorporation:** Label cDNA with CyDye™3 or CyDye™5 at a comparable rate.

Protocol	Part#
ChipShot™ Direct Systems Technical Manual	TM286
ChipShot™ Indirect Systems Technical Manual	TM287

Storage Conditions: Store the ChipShot™ Direct Labeling System at –20°C, except for the RNase A, which should be stored at room temperature, and the Total RNA Positive Control, which should be stored at –70°C. Store the ChipShot™ Indirect Labeling System at –20°C, except for the RNase Solution, which should be stored at room temperature, and the Total RNA Positive Control, which should be stored at –70°C.



cDNA yield achieved using the ChipShot™ Membrane Clean-Up System compared to competing systems.



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Molecular Diagnostics

Molecular Diagnostics 196

Microsatellite Instability (MSI) Analysis

Product	Size	Cat.#
MSI Analysis System, Version 1.2	100 reactions (50 reaction pairs)	MD1641
Available Separately	**Size**	**Cat.#**
PowerPlex® Matrix Standards, 310	50µl (each dye)	DG4640
PowerPlex® Matrix Standards, 3100/3130	25µl (each dye)	DG4650

Cat.# MD1641 For Research Use Only. Not for use in diagnostic procedures. Cat.# DG4640, DG4650 Not for Medical Diagnostic Use.

Description: The MSI Analysis System, Version 1.2, is a fluorescent multiplex PCR-based method for detection of microsatellite instability (MSI), a form of genomic instability. This instability is due to either insertion or deletion of repeating units during DNA replication and failure of the mismatch repair system (MMR) to correct these errors. MSI analysis typically involves comparison of allelic profiles of microsatellite markers generated by amplification from matching tumor and normal samples. New alleles in the tumor sample not found in the corresponding normal sample indicate the presence of MSI.

The MSI Analysis System, Version 1.2, includes fluorescently labeled primers (marker panel) for co-amplification of seven markers for analysis of the MSI-high (MSI-H) phenotype, including five nearly monomorphic mononucleotide repeat markers (BAT-25, BAT-26, MONO-27, NR-21 and NR-24) and two highly polymorphic pentanucleotide repeat markers (Penta C and Penta D). Amplified fragments are detected using an ABI PRISM® 310, 3100, 3100-*Avant*, 3130 or 3130*xl* Genetic Analyzer after spectral calibration.

This configuration meets the recommendations proposed at the 2002 NCI Workshop for MSI Testing and overcomes the potential misclassification, lack of sensitivity and stutter-artifact problems found in the commonly used five-marker Bethesda Panel.

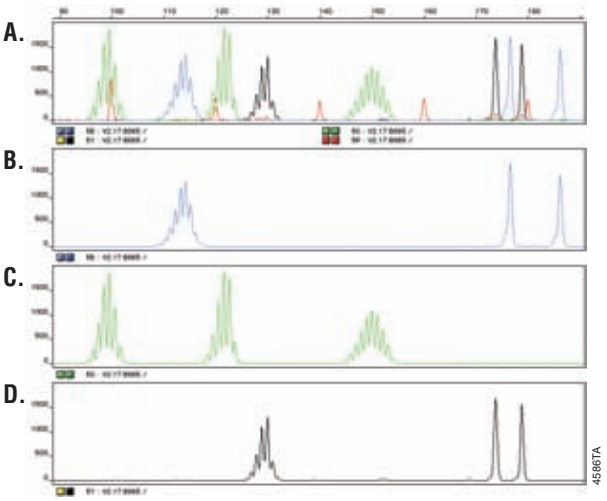
Panels and bins simplify and standardize data analysis by providing automated assignment of genotypes using GeneMapper® 4.0 software.

Features:

- **Understand the Complete MSI Phenotype:** A single multiplex PCR amplifies five informative mononucleotide repeat markers for MSI-H determination.
- **Confidence in Sample Identification:** Co-amplification of highly polymorphic pentanucleotide repeats provides internal sample tracking.
- **Consistent Data Analysis:** Panels and bins for GeneMapper® 4.0 can be downloaded from: **www.promega.com**

Protocol	Part#
MSI Analysis System, Version 12, Technical Manual	TM255
PowerPlex® Matrix Standards, 310, Technical Bulletin	TBD021
PowerPlex® Matrix Standards, 3100/3130, Technical Bulletin	TBD022

Storage Conditions: Store at –20°C.



Analysis of MSI phenotype.

Y Chromosome Deletion Detection System, Version 2.0

Product	Size	Cat.#
Y Chromosome Deletion Detection System, Version 2.0	25 reactions	MD1531

For Research Use Only. Not for use in diagnostic procedures.

Description: The Y Chromosome Deletion Detection System, Version 2.0, provides a standardized screening panel amplifying only informative nonpolymorphic sequence tag sites (STS) on the human Y chromosome. The system amplifies key functional regions associated with AZoospermia Factor (AZF), namely the regions that flank AZFa and cover AZFb, AZFc, AZFd including *DAZ, KAL-Y, SMCY* and flanking loci for other key spermatogenesis-related genes (namely *RBM1, DFFRY* and *DBY*). This updated version 2.0 system contains additional STS making it fully compliant with European Molecular Genetic Quality Network (EMQN) recommendations.

Five Multiplex Master Mixes, with a total of 20 characterized Y-specific primer pairs, are included. Four of the multiplex primer sets contain a control primer pair that amplifies a fragment of the X-linked *SMCX* locus. One of the multiplex primer sets (Multiplex E Master Mix) contains a control primer pair that amplifies a unique region in both male and female DNA (ZFX/ZFY). Finally, a primer pair that amplifies a region of the SRY gene has been included in Multiplex E Master Mix as a control for the testis-determining factor on the short arm of the Y chromosome to detect XX males arising from Y to X translocations.

The Multiplex Master Mixes are designed to facilitate the simultaneous amplification of several different regions of the Y chromosome. The amplification products (83–496bp) of the five multiplex PCR amplifications can be clearly separated by agarose gel electrophoresis and visualized by ethidium bromide staining.

Failure to amplify specific regions of the Y chromosome is indicative of Y chromosome deletions in the test sample. Y chromosome deletions in the regions amplified by these primer sets have been associated with male infertility. The size control ladder provided minimizes analysis time and the possibility of misinterpreting molecular weight of amplification products.

Features:

- **Ease of Use:** Premixed Multiplex Master Mixes contain 20 primer pairs, including internal controls providing a standardized panel of results requiring no user optimization.
- **State-of-the-Art Analysis:** Primer pairs are compliant with current EMQN recommendations and include primer pairs to amplify SRY.
- **More Robust Reactions:** Improved formulation and use of GoTaq® DNA Polymerase minimizes dropouts.
- **Flexibility:** Amplify genomic DNA purified using various methods and with a PE480 (oil overlay) or PE9600/9700 (non-oil overlay) thermal cycler.
- **Complete System:** All required reagents are provided in the kit, except AmpliTaq Gold® (required for use with PE9600 or PE9700 thermal cycler protocol).

Protocol	Part#
Technical Manual	TM248

Storage Conditions: Store at –20°C.

Primer Sets in the Y Chromosome Deletion Detection System.

Multiplex	Locus/ STS 1	Locus/ STS 2	Locus/ STS 3	Locus/ STS 4	Locus/ STS 5
Master Mix A	*DAZ*/ SY254	DYS240/ SY157	DYS271/ SY81	DYS221/ SY130	*KAL-Y*/ SY182
Master Mix B	*SMCY*/ SYPR3	DYS218/ SY127	*DAZ*/ SY242		*DAZ*/ SY208
Master Mix C	DYS219/ SY128	DYS212/ SY121	DYF51S1/ SY145	*DAZ*/ SY255	
Master Mix D	DYS236/ SY152	DYS223/ SY133		DYS215/ SY124	
Master Mix E	SRY/ SY14	DYS224/ SY134	DYS148/ SY86	DYS273/ SY84	ZFX1/ ZFY

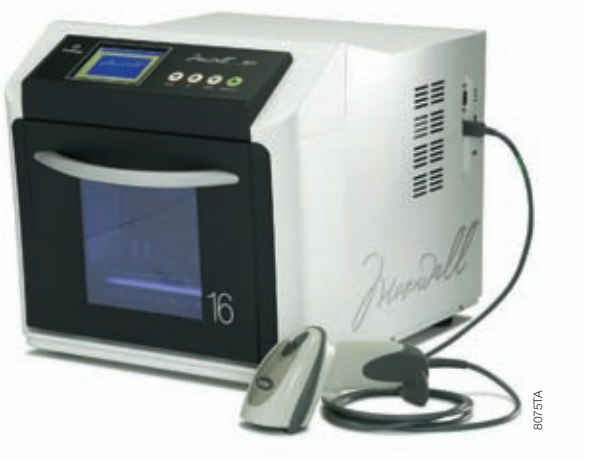
9492LA

Automated DNA, RNA, Viral Total Nucleic Acid and Polyhistidine-Tagged Protein Purification: Maxwell® 16 System

Product	Size	Cat.#
Maxwell® 16 Instrument	1 each	AS2000
Maxwell® 16 MDx Instrument	1 each	AS3000
Available Separately	**Size**	**Cat.#**
Maxwell® 16 Cartridge Rack	1 each	AS1201
Maxwell® 16 Magnetic Elution Rack	1 each	AS1202
Maxwell® 16 LEV Cartridge Rack	1 each	AS1251
Maxwell® 16 SEV Hardware Kit	1 each	AS1200
Maxwell® 16 LEV Hardware Kit	1 each	AS1250
Thermal Serial Printer and Universal Power Cable	1 each	E2821
Maxwell® 16 LEV Magnet	1 each	AS1261

Cat.# AS2000, AS3000 For Laboratory Use.

Maxwell® 16 Instrument (Cat. #AS2000).



Maxwell® 16 Instrument (Cat. #AS3000) with optional bar code reader.

Description: The Maxwell® 16 Instruments provide consistent, labor-saving automated purification of high-quality DNA, RNA, viral total nucleic acid or recombinant proteins for a broad range of downstream applications. The Maxwell® 16 Instrument can be purchased configured as an SEV Instrument (Standard Elution Volume 200–400µl) for maximum yield or LEV Instrument (Low Elution Volume 30–100µl) for maximum concentration. In addition, SEV and LEV instruments can be configured with the Flexi Method Firmware, allowing the user to program the Maxwell® 16 Instrument to further optimize performance. Your personal automation instrument configuration will be built to order. The Maxwell® 16 Instrument is preprogrammed with purification protocols, which when combined with kits containing prefilled reagent cartridges maximize simplicity and convenience. The instrument processes 1 to 16 samples in approximately 18–50 minutes (depending on sample type).

The Maxwell® 16 Instrument extracts DNA, RNA, viral total nucleic acid or recombinant proteins using paramagnetic particles, allowing optimal capture, washing and elution of the target material. Add samples or lysate directly to the prefilled reagent cartridges, and press start. Optimized reagent systems and automated methods are provided to purify from specified sample types to deliver maximum quality for downstream applications.

The Maxwell® 16 Instrument includes a 1-year basic warranty. Service programs are offered to extend coverage. If during the extended warranty period the instrument needs repair under normal use, Promega will be responsible for the repair. Service programs offer similar terms with the addition of the use of a temporary replacement instrument during the instrument repair period. Please contact Promega for complete warranty and service terms and limits.

The Maxwell® 16 Instrument is a General Purpose Medical Device (GPLE) in the USA. Visit: **promega.com/maxwell16/** for up-to-date information.

Features:

- **Recover Lost Time and Labor:** Automation gives you back your time and labor to complete your work.
- **Gain Confidence in Your Results:** Instrument design, optimized reagents and automated methods provide consistent yield and purity.
- **Improve Your Productivity:** Process up to 16 samples per instrument run in approximately 30–45 minutes.
- **Choose Your Sample Type:** Flexibility to purify from tissue, cells, blood and other samples.

Protocol	Part#
Maxwell® 16 Instrument (AS2000) Operating Manual	TM295
Maxwell® 16 MDx Instrument (AS3000) Operating Manual	TM320
Maxwell® Sample Track Software Technical Manual	TM314

Storage Conditions: Store at 22–25°C.

Automated DNA Purification: Maxwell® 16 System

Product	Size	Cat.#
Maxwell® 16 LEV Blood DNA Kit	48 preps	AS1290
Maxwell® 16 Blood DNA Purification Kit	48 preps	AS1010
Maxwell® 16 Cell DNA Purification Kit	48 preps	AS1020
Maxwell® 16 Tissue DNA Purification Kit	48 preps	AS1030
Maxwell® 16 Mouse Tail DNA Purification Kit	48 preps	AS1120
Maxwell® 16 FFPE Tissue LEV DNA Purification Kit	48 preps	AS1130
Maxwell® 16 Cell LEV DNA Purification Kit	48 preps	AS1140
Available Separately	**Size**	**Cat.#**
Maxwell® 16 MDx Instrument	1 each	AS3000
Maxwell® 16 Instrument	1 each	AS2000
Maxwell® 16 LEV Magnet	1 each	AS1261

Cat.# AS1290, AS1010, AS1020, AS1030, AS1130, AS1140, AS3000, AS2000 For Laboratory Use.

Description: The Maxwell® 16 Genomic DNA Purification Kits are designed for use with the Maxwell® 16 Instrument. Seven kits are provided for DNA purification with corresponding optimized automated methods. You get consistent yield and purity from easy-to-use automation.

For genomic DNA purification, the Maxwell® 16 System is the only system that makes purification from tissue as easy as purification from blood or cells. The action of the plunger grinds solid tissue samples directly in the lysis buffer in the prefilled reagent cartridges. Integrated grinding replaces time- and labor-intensive use of lytic enzymes such as proteinase K or manual tissue grinding prior to purification.

Kits for optimized DNA purification from eukaryotic tissue, blood, cells, mouse tail and FFPE tissue sections are available. Protocols for a variety of new samples are being developed. Visit: **www.promega.com/maxwell16/** for up-to-date information.

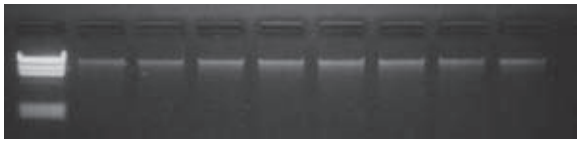
Features:

- **Achieve High Yield:** Efficient processing and higher sample capacity than comparable systems.
- **Enjoy Amazing Speed:** Hands-free purification of genomic DNA in 18–30 minutes.
- **Get More Time:** Easily process tissues and cells.

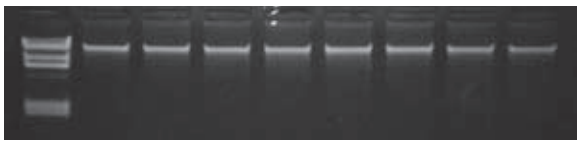
Protocol	Part#
Maxwell® 16 LEV Blood DNA Kit Technical Manual	TM333
Maxwell® 16 DNA Purification Kits (Cat# AS1010, AS1020, AS1030) Technical Manual	TM284
Maxwell® 16 Mouse Tail DNA Purification Kit (Cat# AS1120) Technical Manual	TM309
Maxwell® 16 FFPE Tissue LEV DNA Purification Kit (Cat# AS1130) Technical Bulletin	TB382
Maxwell® 16 Cell LEV DNA Purification Kit (Cat# AS1140) Technical Bulletin	TB383

Storage Conditions: Store at 5–40°C.

A.



B.



5513TB

L 1 2 3 4 5 6 7 8

Genomic DNA purified from 8 samples of 200µl of whole human blood (Panel A) and 8 samples of 1cm of mouse tail (Panel B) visualized on a 1% agarose gel stained with ethidium bromide. Lane L: Lambda DNA/HindIII Markers (Cat.# G1711). Lanes 1–8: 5µl of purified genomic DNA.

DNA Yields from Various Starting Materials.

Sample Type	Sample Size	Yield
Whole blood	200µl	4–9µg (>3pg/white blood cell)
Whole blood	400µl	8–15µg (>3pg/white blood cell)
Mouse tail	1.2cm	20µg
Animal tissue	20–25mg	60–100µg (mouse liver)
Tissue culture cells	5×10^6	10µg (HeLa)
Gram– bacteria	2×10^9	10µg (BL21)
Gram+ bacteria	2×10^9	1µg (*B. cereus*)
Plant leaf (tomato)	25mg	10µg

9482LA

Automated RNA Purification: Maxwell® 16 System

Product	Size	Cat.#
Maxwell® 16 Total RNA Purification Kit	48 preps	AS1050
Maxwell® 16 Tissue LEV Total RNA Purification Kit	48 preps	AS1220
Maxwell® 16 Cell LEV Total RNA Purification Kit	48 preps	AS1225
Available Separately	**Size**	**Cat.#**
Maxwell® 16 Instrument	1 each	AS2000

Cat.# AS1050, AS1220, AS1225, AS2000 For Laboratory Use.

Description: The Maxwell® 16 Total RNA Purification Kit, Maxwell® 16 Tissue LEV Total RNA Purification Kit and Maxwell® 16 Cell LEV Total RNA Purification Kit are designed for use with the Maxwell® 16 Instrument in either the standard or low elution volume (LEV) configuration. The kits provide high-quality, essentially DNA-free total RNA using novel approaches to selectively remove genomic DNA prior to automated RNA purification. You get enhanced sensitivity and improved confidence in your results for quantitative RT-PCR (qRT-PCR), RT-PCR, cDNA synthesis and other applications.

The Maxwell® 16 Total RNA Purification Kit extracts total RNA from white blood cell fraction of whole blood, tissue culture cell lines, PAXgene®-stabilized whole blood and plant leaf tissue and can be used in RNA cleanup applications from TRIzol® extractions or in vitro transcription reactions. Beta-Mercaptoethanol is included for use with certain sample types.

The Maxwell® 16 LEV Tissue and Cell Total RNA Purification Kits provide high-concentration (100ng/µl), essentially DNA-free total RNA from cultured cells, mammalian tissue and PAXgene®-stabilized white blood cells. It's personal automation working for you, so you can get more consistent gene expression analysis results with less hands-on labor.

The simple protocols require adding a cleared lysate to the reagent cartridge. Place the reagent cartridge into the Maxwell® 16 Instrument, and press start. Purified RNA is obtained in less than 45 minutes of hands-free instrument operation. No post-purification treatment with nuclease, cleanup or concentration is required to achieve superior performance in downstream applications.

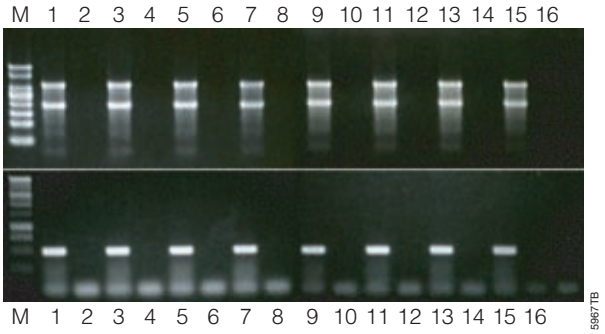
The Maxwell® 16 Total RNA Purification Kits are General Purpose Medical Devices (GPR) in the USA. Visit: **www.promega.com/maxwell16/** for up-to-date information.

Features:

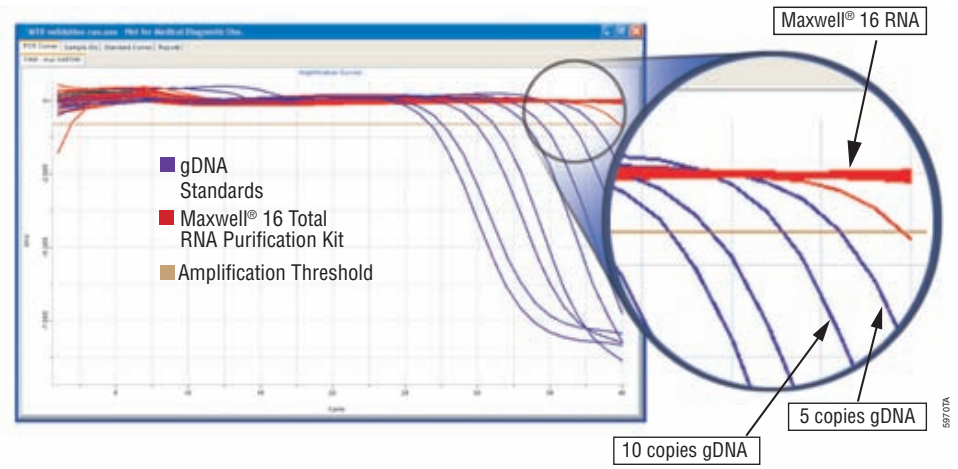
- **Enjoy Confidence in Your Application Results:** Essentially undetectable contaminating genomic DNA means fewer repeated experiments and unexplained or variable results.
- **Choose Your Sample Type:** Flexibility to purify from tissue, cells, blood and other samples.
- **Achieve High Yield and High Concentration:** High yields and high-concentration total RNA result in better performance in gene expression analysis applications.

Protocol	Part#
Maxwell® 16 Total RNA Purification Kit Technical Bulletin	TB351
Maxwell® 16 Tissue LEV Total RNA Purification Kit Technical Bulletin	TB367
Maxwell® 16 Cell LEV Total RNA Purification Kit Technical Bulletin	TB368

Storage Conditions: Store the kit components at room temperature (15–30°C). For Cat.# AS1225, upon receipt, remove the RNasin® Plus RNase Inhibitor and store at –20°C.



No detectable cross-contamination. Sixteen purification reactions were performed using an input of 25mg of mouse liver lysate (odd lanes) or SV RNA Lysis Buffer alone (even lanes). **Panel A.** Four-microliter aliquots of each purified sample were resolved by 1.2% agarose gel electrophoresis under denaturing conditions.Lane M, RNA Markers (Cat.# G3191). **Panel B.** Equivalent volumes (1µl) of each sample were amplified by endpoint RT-PCR using a primer pair specific for a portion of beta actin RNA. A total of five microliters of each amplification reaction was analyzed by 1.2% agarose gel electrophoresis and visualized by ethidium bromide staining. Lane M, 1kb DNA Ladder (Cat.# G5711).



Undetectable genomic DNA contamination. RNA was isolated from 24 replicate samples of 25mg of mouse liver and analyzed using the Plexor® qPCR System and a primer pair specific for a portion of the mouse glyceraldehyde-3-phosphate dehydrogenase (GAPDH) gene. An average of less than 0.1 copies of genomic DNA (gDNA) was observed in purified RNA.

Maxwell® 16 Flexi Method Firmware

Product	Size	Cat.#
Maxwell® 16 Flexi Method Firmware	1 each	AS6411

Description: Certain sample types present unique challenges for DNA, RNA or recombinant protein extraction. The Maxwell® 16 Flexi Method Firmware provides the flexibility and control to modify or create automated methods for the Maxwell® 16 Instrument. You have the ability to optimize multiple instrument parameters to tailor instrument operation to your unique needs. It's Personal Automation™ just the way you want it. The Maxwell® 16 Flexi Method Firmware allows users to change 5 key instrument operating parameters:

- Lysis time
- Binding
- Drying
- Elution
- Paramagnetic particle capture

You program the Maxwell® 16 Instrument by following on-screen prompts and entering changes through the instrument keypad; no external PC or programming knowledge is required. User-defined optimized methods are as easy to use as pushing the Start button. The Flexi Method Firmware also allows you to save and password-protect your unique methods. Make and save changes as you define the key instrument operating parameters that impact your successful results.

The Flexi Method Firmware can be installed on existing AS1000 and AS2000 Maxwell® 16 Instruments by purchasing the AS6411 CD-ROM, which contains the Firmware, installation software and Technical Manual. Flexi Method Firmware ordered with the purchase of a new AS2000 Instrument will be installed at the factory.

Features:

- **Achieve Confidence in your Results:** You control operation of key instrument operating parameters.
- **Address Key Unanswered Questions:** Flexibilty gives you the ability to optimize Maxwell® 16 operation to your sample and scientific needs.
- **Spend More Time Generating Data:** Follow simple on-screen prompts to program instrument from the keypad. Press Run to start.

Protocol	Part#
Technical Bulletin	TB381

Storage Conditions: Store at 22–25°C.

Automated Clinical Nucleic Acid Purification: Maxwell® 16 MDx System

Product	Size	Cat.#
Maxwell® 16 MDx Instrument	1 each	AS3000
Available Separately	**Size**	**Cat.#**
Maxwell® 16 Viral Total Nucleic Acid Purification Kit	48 preps	AS1150
Maxwell® 16 LEV Blood DNA Kit	48 preps	AS1290
Maxwell® 16 Blood DNA Purification Kit	48 preps	AS1010
Maxwell® 16 Cell DNA Purification Kit	48 preps	AS1020
Maxwell® 16 Tissue DNA Purification Kit	48 preps	AS1030
Maxwell® 16 FFPE Tissue LEV DNA Purification Kit	48 preps	AS1130
Maxwell® 16 Cell LEV DNA Purification Kit	48 preps	AS1140
Maxwell® 16 Total RNA Purification Kit	48 preps	AS1050
Maxwell® 16 Tissue LEV Total RNA Purification Kit	48 preps	AS1220
Maxwell® 16 Cell LEV Total RNA Purification Kit	48 preps	AS1225
Maxwell® 16 LEV Hardware Kit	1 each	AS1250
Maxwell® 16 LEV Cartridge Rack	1 each	AS1251
Maxwell® 16 SEV Hardware Kit	1 each	AS1200
Maxwell® 16 Cartridge Rack	1 each	AS1201
Thermal Serial Printer and Universal Power Cable	1 each	E2821
Maxwell® 16 LEV Magnet	1 each	AS1261

Cat.# AS3000, AS1150, AS1290, AS1010, AS1020, AS1030, AS1130, AS1140, AS1050, AS1220, AS1225 For Laboratory Use.

Description: The Maxwell® 16 MDx Instrument provides easy-to-use, consistent and reliable automated nucleic acid extraction of one to 16 samples, bar-code sample tracking, a touch-screen interface and UV decontamination. You choose either low elution volume (50–100µl, LEV) or standard elution volume (300–400µl, SEV) format. Run report data can be transferred from the Maxwell® 16 MDx Instrument to a PC or to an external printer. Data transferred to a PC is can be uploaded to a laboratory information management system (LIMS). The Maxwell® 16 MDx Instrument is labeled as General Purpose Laboratory Equipment (GPLE) in the USA. For the rest of the world, it is intended for research use only.

Features:

- **Fast, Hands-Free Purification:** Improves workflow, and allows staff to perform other value-added tasks.
- **Consistent, Reliable Performance:** Less rework; confidence in results.
- **Easy-to-Use:** Immediate productivity gains; minimal operator training required.
- **Small Size:** Takes up less room on the lab bench. Fits inside biosafety cabinet or hood.
- **Bar-Code Sample Tracking Capability:** Eliminates sample mixup, and data can be integrated into LIMS.
- **UV Light:** Helps decontamination.

Protocol	Part#
Maxwell® 16 MDx Instrument Technical Manual	TM320
Maxwell® Sample Track Software Technical Manual	TM314

Storage Conditions: Store at 15–40°C.

Maxwell® 16 Service and Support

Product	Size	Cat.#
Maxwell® 16 Premier Warranty	1 each	SA2000
Maxwell® 16 Standard Service Agreement	1 each	SA2010
Maxwell® 16 Premier Service Agreement	1 each	SA2015
Maxwell® 16 Preventative Maintenance	1 each	SA2020
Maxwell® 16 Installation Qualification	1 each	SA1001
Maxwell® 16 Operational Qualification	1 each	SA1011
Maxwell® 16 Installation and Operational Qualification	1 each	SA1021

Cat.# SA2000, SA2010, SA2015 Not available in all markets. Please contact your local representative for details.

Description: The **Standard Warranty**, included in the system price, covers all parts, labor and shipping to and from our repair location as well as a loaner instrument upon request. The loaner will be shipped via standard ground shipment and will arrive in 5 to 7 working days. We will repair your instrument and return it to you performing to original factory specifications.

The **Premier Warranty** (SA2000) covers all parts, labor and shipping to and from our depot repair location as well as your choice of a loaner instrument within 1 working day or on-site repair by a factory-trained service technician. We will repair your instrument and return it to you performing to original factory specifications. It also includes one preventive maintenance visit.

The **Standard Service Agreement** (SA2010) covers all parts, labor and shipping to and from our depot repair location as well as a loaner instrument upon request. The loaner will be shipped via standard ground shipment and will arrive in 5 to 7 working days. If your Maxwell® 16 Instrument needs repair, we will provide a box for shipment of the instrument back to our service facility. We will repair it and return it to you performing to original factory specifications.

The **Premier Service Agreement** (SA2015) includes all parts, labor and shipping to and from our depot repair location as well as your choice of a loaner instrument within 1 working day or on-site repair by a factory-trained service technician. You can utilize our depot repair and receive a loaner instrument in one working day or you can elect to have one of our service technicians service it in your lab. Additionally, it includes one annual preventive maintenance visit per year.

In order to keep the system operating at peak performance, Promega recommends that Maxwell® 16 Instruments receive a **Preventive Maintenance** (SA2020) check after 12 months of use. During this procedure, our qualified service personnel test the instrument, check parts for wear and replace them as needed. Additionally, the system is aligned and performance is verified. Documentation for your files is provided. The preventive maintenance service is performed by returning the instrument to the factory.

The **Installation Qualification** (SA1001) provides a series of formal on-site instrument checks, delivers written documentation of instrument functionality, and demonstrates that everything ordered with your instrument is supplied and installed in your laboratory. Upon delivery to the lab, the instrument and its components will be visually inspected and reviewed for completeness. Following the inspection, the instrument will be powered on to confirm that the system is properly functional.

The **Operational Qualification** (SA1011) demonstrates that the Maxwell® 16 will function according to its operational specifications. An instrument specialist will check the instrument's alignment and then perform an operational test run to ensure that all of the hardware modules function correctly. Following the documentation of these tests, familiarization training with the instrument's operators will occur. The specialist will also explain all of the sections of the instrument log book.

The **Installation and Operational Qualification** package (SA1021) includes all of the components from both SA1001 and SA1011 in one service product.

Features:

- **Multiple Options to Meet Your Needs:** Allows you to select the warranty coverage or service agreement that best meets the needs of your lab.
- **Factory-Trained Specialists:** Ensures your instrument is repaired quickly and effectively.
- **Expert Technical Service:** Promega experts can help you solve problems quickly.
- **Fixed-Cost Service Products:** Predictable support expenditures.
- **Ongoing System Documentation:** Allows audit tracing and compliance.
- **Comprehensive Service and Support:** Makes certain there is minimal instrument downtime.

Molecular Weight Markers

Premixed, Ready-to-Load DNA Markers	204
DNA Step Ladders	205
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Conventional DNA Markers	207
Markers for Autoradiography	208
Chromosomal DNA Marker	209
RNA Markers	209
Protein Markers	210
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Loading Dye	210

BenchTop DNA Markers

Product	Size	Cat.#
BenchTop ΦX174 DNA/HaeIII Markers	250µl (50 lanes)	G7511
BenchTop pGEM® DNA Markers	250µl (50 lanes)	G7521
BenchTop PCR Markers	300µl (50 lanes)	G7531
BenchTop 1kb DNA Ladder	600µl (100 lanes)	G7541
BenchTop 100bp DNA Ladder	300µl (50 lanes)	G8291

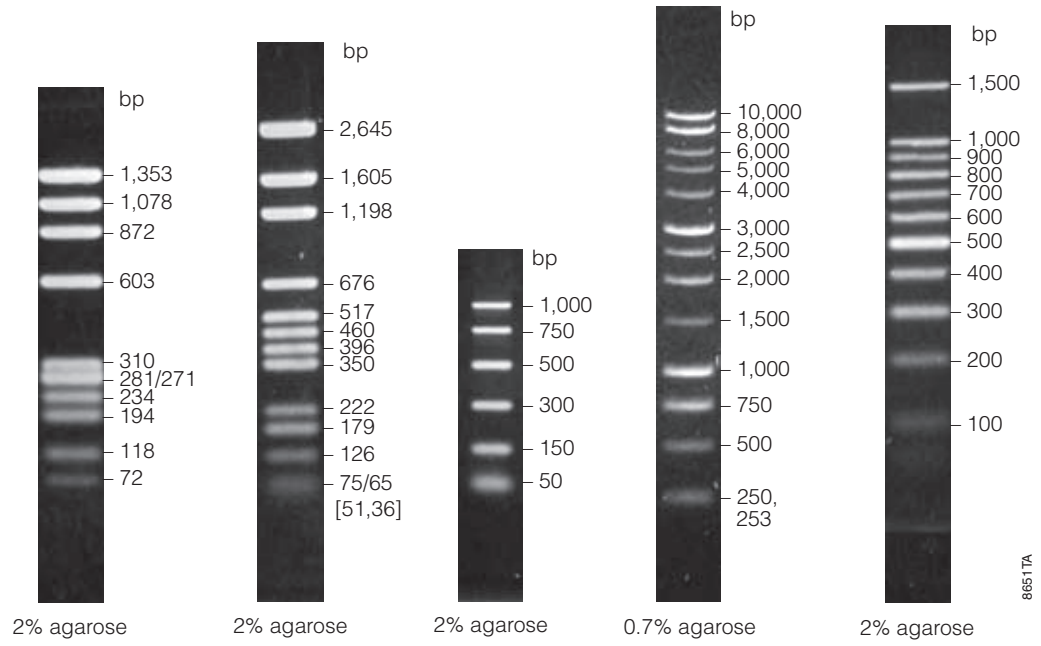
For Laboratory Use.

Description: The BenchTop DNA Markers offer the convenience of storage at room temperature (22–25°C) as well as the capability of direct loading onto agarose gels. The BenchTop DNA Markers are supplied in a stabilizing solution of 1X Blue/Orange Loading Dye, which circumvents any requirements for further manipulation.

Features:

- **Convenient:** Storage at 22–25°C.
- **Efficient:** Premixed with loading buffer. Ready to load onto agarose gels.
- **Recommended Loading:** Cat.# G7511, G7521: Load 5µl/lane. Cat.# G7531, G7541, G8291: Load 6µl/lane.

Storage Conditions: Store at 22–25°C.



DNA Step Ladders

Product	Size	Cat.#
10bp DNA Step Ladder	32.5µg (50 lanes)	G4471
25bp DNA Step Ladder	100µg (55 lanes)	G4511
50bp DNA Step Ladder	90µg (52 lanes)	G4521
100bp DNA Step Ladder	100µg (100 lanes)	G6951
200bp DNA Step Ladder	100µg (100 lanes)	G6961
1kb DNA Step Ladder	90µg (300 lanes)	G6941

For Laboratory Use.

Description: The DNA Step Ladders are ladders of defined sizes with exact incremental steps between bands. The ladders are not intended for use in quantitative analysis. Each ladder is provided with a tube of 6X Blue/Orange Loading Dye. The fragments may be stained with ethidium bromide.

10bp DNA Step Ladder: Ten blunt-ended DNA fragments ranging from 10bp to 100bp in exactly 10bp increments. All of the bands are of approximately equal intensity with the exception of the 10bp band, which may appear slightly less intense.

25bp DNA Step Ladder: Twelve DNA fragments ranging from 25bp to 300bp in 25bp increments. An 1,800bp "backbone" fragment is also visible. The 300bp band is ≈3 times more intense than all other bands.

50bp DNA Step Ladder: Sixteen DNA fragments ranging from 50bp to 800bp in 50bp increments plus an 1,800bp "backbone" fragment. All bands except the 800bp band are of equal intensity; the 800bp band is ≈3 times more intense.

100bp DNA Step Ladder: Forty blunt-ended DNA fragments ranging from 100bp to 4,000bp in 100bp increments. Two internal features facilitate band identification. First, a high-intensity 500bp band stands out at the lowest segment of the ladder (<1kb). Bands within each segment (<1kb, <2kb, <4kb) have approximately the same intensity.

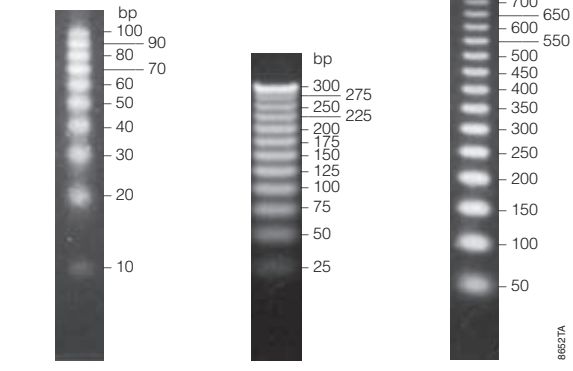
200bp DNA Step Ladder: Thirty-three blunt-ended DNA fragments ranging from 200bp to 6,600bp in 200bp increments. The 1,000bp band appears more intense than all other bands, which are of approximately equal intensity.

1kb DNA Step Ladder: Ten blunt-ended DNA fragments ranging from 1kb to 10kb in 1kb increments. All bands except the 5kb band are of equal intensity; the 5kb band is ≈3 times more intense.

Features:

- **Recommended Loading:** Cat.# G4471, G6951, G6961, G6941: Load 1µl/lane. Cat.# G4511, G4521: Load 5µl/lane.

Storage Conditions: Store at –20°C.



4% agarose (3% NuSieve® GTG® agarose/1% agarose)

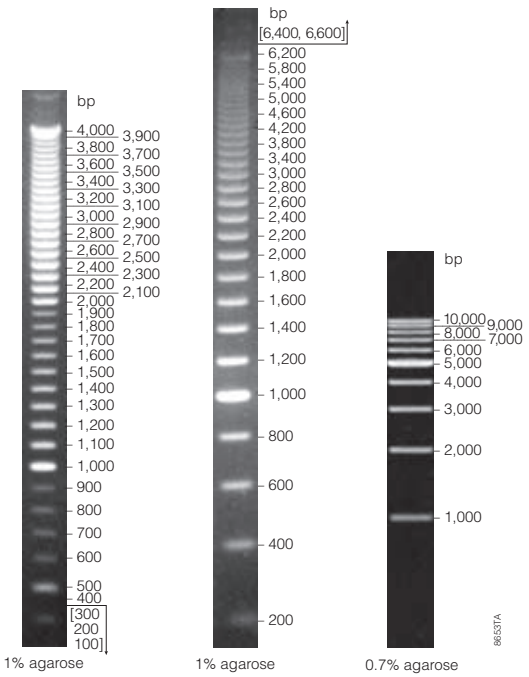
10bp DNA Step Ladder
Cat.# G4471
Load 1µl/lane.

2% agarose/TAE

25bp DNA Step Ladder
Cat.# G4511
Load 5µl/lane.

2% agarose/TAE

50bp DNA Step Ladder
Cat.# G4521
Load 5µl/lane.



1% agarose

100bp DNA Step Ladder
Cat.# G6951
Load 1µl/lane.

1% agarose

200bp DNA Step Ladder
Cat.# G6961
Load 1µl/lane.

0.7% agarose

1kb DNA Step Ladder
Cat.# G6941
Load 1µl/lane.

DNA Ladders

Product	Size	Cat.#
PCR Markers	250µl (50 lanes)	G3161
100bp DNA Ladder	250µl (50 lanes)	G2101
1kb DNA Ladder	500µl (100 lanes)	G5711

For Laboratory Use.

Description: The DNA Ladders are ladders with defined sizes. The ladders are not intended for use in quantitative analysis. Each ladder is provided with a tube of 6X Blue/Orange Loading Dye.

PCR Markers: Six bands of equal intensity of 50, 150, 300, 500, 750 and 1,000bp. The PCR Markers may be run on polyacrylamide gels with less loading volume; however, additional bands may be visible compared to those visible on agarose gels.

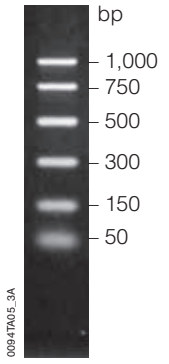
100bp DNA Ladder: Eleven fragments that range in size from 100bp to 1,000bp in 100bp increments with an additional band at 1,500bp. The 500bp fragment is present at increased intensity for easy identification. The ladder is dephosphorylated and ready for 5´ end-labeling with radioisotopes using T4 Polynucleotide Kinase, allowing visualization by autoradiography.

1kb DNA Ladder: Thirteen blunt-ended fragments with sizes ranging from 250bp to 10,000bp. The 1,000bp and 3,000bp fragments have increased intensity relative to the other bands on ethidium bromide-stained agarose gels for easy identification. All other fragments are of equal intensity. The ladder is dephosphorylated and ready for 5´ end-labeling with radioisotopes using T4 Polynucleotide Kinase, allowing visualization by autoradiography.

Features:

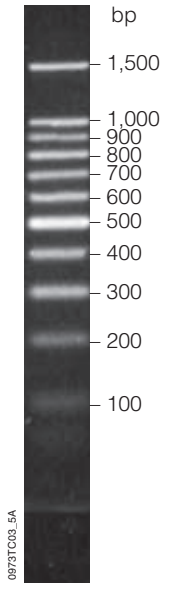
- **Recommended Loading:** Load 5µl/lane.

Storage Conditions: Store at –20°C.



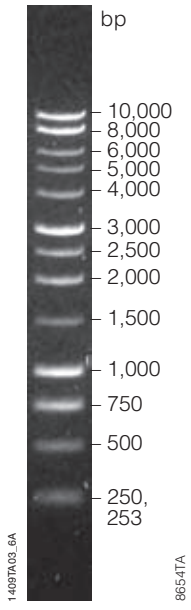
2% agarose

PCR Markers
Cat.# G3161
Load 5µl/lane.



2% agarose

100bp DNA Ladder
Cat.# G2101
Load 5µl/lane.



0.7% agarose

1kb DNA Ladder
Cat.# G5711
Load 5µl/lane.

Conventional DNA Markers

Product	Size	Cat.#
Lambda DNA/HindIII Markers	100µg (200 lanes)	G1711
Lambda DNA/EcoRI Markers	100µg (200 lanes)	G1721
Lambda DNA/EcoRI + HindIII Markers	100µg (200 lanes)	G1731
ΦX174 DNA/HaeIII Markers	50µg (50 lanes)	G1761
ΦX174 DNA/HinfI Markers	50µg (50 lanes)	G1751
pGEM® DNA Markers	50µg (50 lanes)	G1741

For Laboratory Use.

Description: The Conventional DNA Digest Markers are created by digesting either λ DNA, ΦX174 replicative form DNA, or plasmids to completion with one or more restriction enzymes. The enzymes are heat-inactivated, and the DNA fragments are either phenol-extracted, then ethanol-precipitated or just ethanol-precipitated. The precipitated fragments are resuspended in storage buffer. The markers are not intended for quantitative analysis. Each marker is supplied with a tube of 6X Blue/Orange Loading Dye.

λ DNA/HindIII Markers: Eight ethanol-precipitated DNA fragments ranging in size from 125bp to 23,130bp.

λ DNA/EcoRI Markers: Six ethanol-precipitated DNA fragments ranging in size from 3,530bp to 21,226bp.

λ DNA/EcoRI + HindIII Markers: Thirteen ethanol-precipitated DNA fragments ranging in size from 125bp to 21,226bp.

ΦX174 DNA/HaeIII Markers: Eleven phenol-extracted, ethanol-precipitated DNA fragments ranging in size from 72bp to 1,353bp.

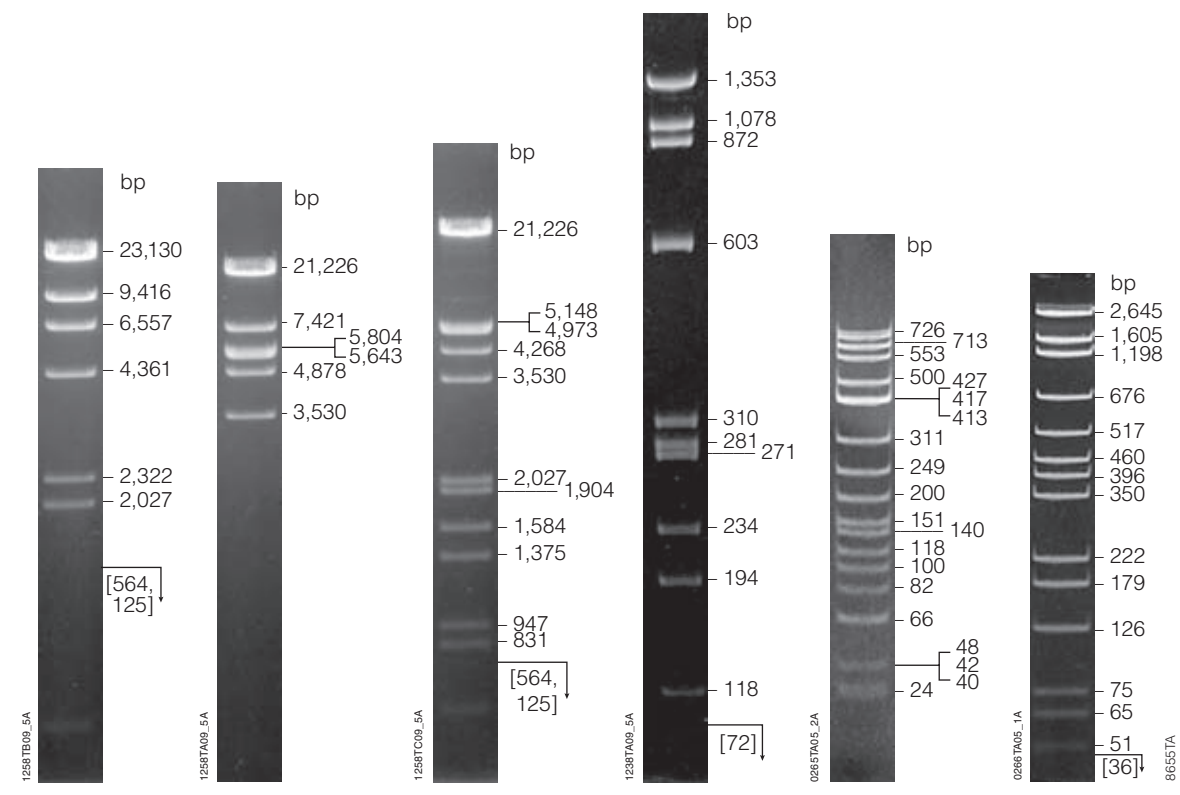
ΦX174 DNA/HinfI Markers: Twenty ethanol-precipitated DNA fragments ranging in size from 24bp to 726bp.

pGEM® DNA Markers: Fifteen phenol-extracted, ethanol-precipitated DNA fragments ranging in size from 36bp to 2,645bp. These unique markers are generated from separate digests of pGEM®-3 Vector DNA with HinfI, RsaI and SinI later combined to form the markers.

Features:

- **Recommended Loading:** Load 1µl/lane.

Storage Conditions: Store at –20°C.



0.7% agarose

Lambda DNA/ HindIII Markers Cat.# G1711 Load 1µl/lane.

0.7% agarose

Lambda DNA/ EcoRI Markers Cat.# G1721 Load 1µl/lane.

0.7% agarose

Lambda DNA/EcoRI + HindIII Markers Cat.# G1731 Load 1µl/lane.

8% acrylamide

ϕX174 DNA/ HaeIII Markers Cat.# G1761 Load 1µl/lane.

8% acrylamide

ϕX174 DNA/ HinfI Markers Cat.# G1751 Load 1µl/lane.

8% acrylamide

pGEM® DNA Markers Cat.# G1741 Load 1µl/lane.

ΦX174 DNA/HinfI Dephosphorylated Markers

Product	Size	Cat.#
ΦX174 DNA/HinfI Dephosphorylated Markers	2.5 µg	E3511
For Laboratory Use.		

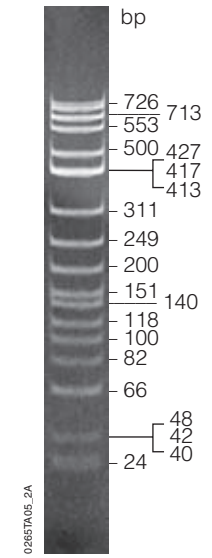
Description: ΦX174 DNA/HinfI Dephosphorylated Markers are prepared by digesting double-stranded ΦX174 DNA to completion with HinfI. The DNA fragments are then treated with calf intestinal alkaline phosphatase, phenol:chloroform-extracted, ethanol-precipitated and resuspended in TE buffer, making the markers ready for 5´ end-labeling. The 20 DNA fragments range in size from 24–726bp. The markers are not intended for use in quantitative analysis.

This marker is especially convenient for applications such as primer extension, requiring DNA or RNA size estimations.

Features:

- **Concentration:** 50µg/ml.
- **Range (bp):** 24–726.
- **Number of Bands:** 20.
- **Convenient:** Ready to label.

Storage Conditions: Store at –20°C.



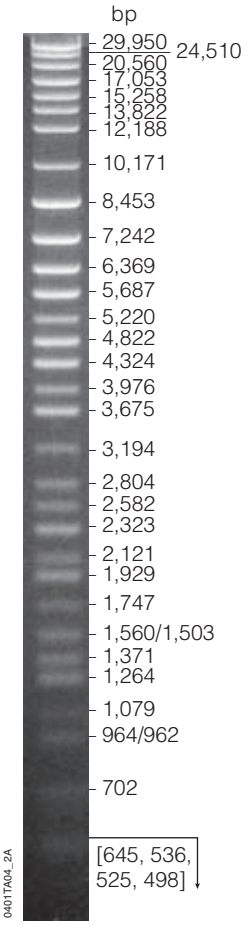
8% acrylamide

Analytical Marker DNA Wide Range

Product	Size	Conc.	Cat.#
Analytical Marker DNA Wide Range	2 µg	0.2 µg/µl	DG1931
For Laboratory Use.			

Description: The Analytical Marker DNA Wide Range, intended for auto-radiography, provides an evenly spaced distribution of 32 DNA fragments ranging from 702bp to 29,950bp in size and 4 smaller fragments (498, 525, 536 and 645bp) that are generally not visible unless the gel is overexposed. This marker is composed of a mixture of restriction enzyme digests of λ DNA (Cat.# D1501) and ΦX174 DNA (Cat.# D1531). The marker is not intended for use in quantitative analysis.

Storage Conditions: Store at –20°C.



0.5% agarose

ProMega-Markers® Lambda Ladders

Product	Size	Cat.#
ProMega-Markers® Lambda Ladders	40–60 lanes	G3011

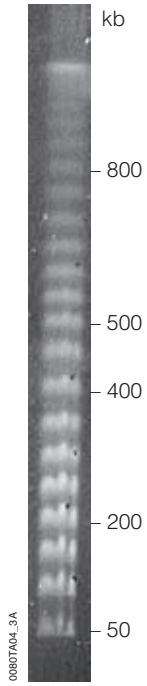
For Laboratory Use.

Description: ProMega-Markers® Lambda Ladders are prepared by concatemerization of λ phage DNA into multimers ranging in size from 50kb to 800kb and up, with each multimer, or rung, of the 20-step ladder differing in size by one λ genome (approximately 48.5kb). The ladders are embedded in dye-colored, 0.5% agarose string molds in 50mM EDTA. The ladders are not intended for use in quantitative analysis.

Features:

- **Concentration:** 0.5µg/5mm.
- **Range (bp):** 50,000–800,000 and up.

Storage Conditions: Store at 4°C. **Do not freeze.**



0.6% agarose

RNA Markers

Product	Size	Cat.#
RNA Markers	50 µl	G3191

For Laboratory Use.

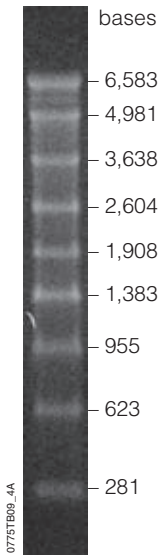
Description: Promega RNA Markers are suitable for size estimation of single-stranded RNA from 0.28–6.58kb in glyoxal or formaldehyde-agarose gels. The RNA Markers consist of a ladder of nine RNA transcripts that are synthesized in vitro from specific templates. The sizes are 281, 623, 955, 1,383, 1,908, 2,604, 3,638, 4,981 and 6,583 bases. The markers are not intended for use in quantitative analysis. After electrophoresis, the fragments can be visualized by ethidium bromide staining.

Features:

- **Recommended Loading:** 3µl (prepared in formaldehyde/MOPS buffer and separated onto a 1% formaldehyde-agarose gel using MOPS running buffer).
- **Range (bases):** 281–6,583.
- **Number of Bands:** 9.

Protocol	Part#
Promega Product Information	9PIG319

Storage Conditions: Store at –70°C.



1% formaldehyde-agarose

Broad Range Protein Molecular Weight Markers

Product	Size	Conc.	Cat.#
Broad Range Protein Molecular Weight Markers	100 lanes	5 µl/lane	V8491

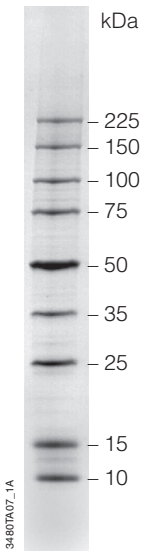
For Laboratory Use.

Description: The Broad Range Protein Molecular Weight Markers consist of nine clearly identifiable bands at convenient molecular weights. The protein sizes are 10, 15, 25, 35, 50, 75, 100, 150 and 225kDa. The band at 50kDa is of greater intensity for use as a reference point. These markers are intended for use as a size standard when performing SDS-PAGE (sodium dodecyl sulfate-polyacrylamide gel electrophoresis) for estimation of the molecular weight of the protein of interest. Note that they are not stained.

Features:

- **Reference Band:** Band at 50kDa is 3X intensity for use as a reference.
- **Convenient:** 9 bands at evenly spaced intervals.
- **Fast:** Ready to load.

Storage Conditions: Store at –20°C (weekly/monthly use) or 4°C (daily use).



4–20% Tris-Glycine SDS-PAGE gel

Genomic DNA

Product	Size	Cat.#
Human Genomic DNA: Male	100 µg	G1471
Human Genomic DNA: Female	100 µg	G1521
Human Genomic DNA	100 µg	G3041
Mouse Genomic DNA	100 µg	G3091

G1471, G1521, G3041 For Laboratory Use.

Description: Genomic DNA from selected species are purified, and greater than 90% of the DNA is longer than 50kb in size as measured by pulsed-field gel electrophoresis. The DNA is suitable for Southern blot hybridizations, genomic analysis (including PCR), and genomic library construction. The Mouse Genomic DNA is isolated from whole blood from disease-free mice. Human Genomic DNA comes from multiple anonymous donors.

Storage Conditions: Store at 4°C.

Blue/Orange Loading Dye, 6X

Product	Size	Cat.#
Blue/Orange Loading Dye, 6X	3ml (3 × 1 ml)	G1881

For Laboratory Use.

Description: Blue/Orange Loading Dye, 6X, is a convenient marker dye containing 0.4% orange G, 0.03% bromophenol blue, 0.03% xylene cyanol FF, 15% Ficoll® 400, 10mM Tris-HCl (pH 7.5) and 50mM EDTA (pH 8.0). It is provided in a premixed, ready-to-use form. The dye is used for loading DNA samples into gel electrophoresis wells and tracking migration during electrophoresis. In a 0.5–1.4% agarose gel in 0.5X TBE, xylene cyanol FF migrates at approximately 4kb, bromophenol blue at approximately 300bp and orange G at approximately 50bp.

Features:

- **Quality Tested:** Each lot of Blue/Orange Loading Dye, 6X, is tested and certified to be free of nuclease activity.

Storage Conditions: Store at –20°C.

Protein Expression, Purification and Functional Analysis

Vectors	**212**
Competent Cells	**215**
Sequencing Primers	**216**
Cell-Free Eukaryotic Systems	**216**
Cell-Free Prokaryotic Systems	**218**
Cell-Free Systems, Accessory Products	**223**
Cell-Based Systems	**224**
Protein Purification	**225**
Protein Labeling	**229**
Protein Modification	**231**
Protein Function, Protein Interactions	**234**

Flexi® Cloning System

Product	Size	Cat.#	
Flexi® System, Entry/Transfer	5 entry and 20 transfer reactions	C8640	223
Flexi® System, Transfer	100 transfer reactions	C8820	891
Carboxy Flexi® System, Transfer	50 transfer reactions	C9320	391
Available Separately	**Size**	**Cat.#**	
HaloTag® Cloning Starter System	1 each	G6050	350
10X Flexi® Enzyme Blend (SgfI & PmeI)	25 µl	R1851	85
	100 µl	R1852	310
Carboxy Flexi® Enzyme Blend (SgfI & EcoICRI)	50 µl	R1901	84
pF1A T7 Flexi® Vector	20 µg	C8441	223
pF1K T7 Flexi® Vector	20 µg	C8451	223
pFN2A (GST) Flexi® Vector	20 µg	C8461	251
pFN2K (GST) Flexi® Vector	20 µg	C8471	251
pF3A WG (BYDV) Flexi® Vector	20 µg	L5671	248
pF3K WG (BYDV) Flexi® Vector	20 µg	L5681	248
pF4A CMV Flexi® Vector	20 µg	C8481	251
pF4K CMV Flexi® Vector	20 µg	C8491	251
pF5A CMV-neo Flexi® Vector	20 µg	C9401	251
pF5K CMV-neo Flexi® Vector	20 µg	C9411	251
pFN6A (HQ) Flexi® Vector	20 µg	C8511	251
pFN6K (HQ) Flexi® Vector	20 µg	C8521	251
pFC7A (HQ) Flexi® Vector	20 µg	C8531	251
pFC7K (HQ) Flexi® Vector	20 µg	C8541	251
pFC14A HaloTag® CMV Flexi® Vector	20 µg	G9651	193
pFC14K HaloTag® CMV Flexi® Vector	20 µg	G9661	193
pFC15A HaloTag® CMV*d1* Flexi® Vector	20 µg	G1611	193
pFC15K HaloTag® CMV*d1* Flexi® Vector	20 µg	G1601	193
pFC16A HaloTag® CMV*d2* Flexi® Vector	20 µg	G1591	193
pFC16K HaloTag® CMV*d2* Flexi® Vector	20 µg	G1571	193
pFC17A HaloTag® CMV*d3* Flexi® Vector	20 µg	G1551	193
pFC17K HaloTag® CMV*d3* Flexi® Vector	20 µg	G1321	193
pFN18A HaloTag® T7 Flexi® Vector	20 µg	G2751	193
pFN18K HaloTag® T7 Flexi® Vector	20 µg	G2681	193
pFN19A HaloTag® T7 SP6 Flexi® Vector	20 µg	G1891	193
pFN19K HaloTag® T7 SP6 Flexi® Vector	20 µg	G1841	193
pFC20A HaloTag® T7 SP6 Flexi® Vector	20 µg	G1681	193
pFC20K HaloTag® T7 SP6 Flexi® Vector	20 µg	G1691	193
pFN21A HaloTag® CMV Flexi® Vector	20 µg	G2821	193
pFN21K HaloTag® CMV Flexi® Vector	20 µg	G2831	193
pFN22A HaloTag® CMV*d1* Flexi® Vector	20 µg	G2841	193
pFN22K HaloTag® CMV*d1* Flexi® Vector	20 µg	G2851	193
pFN23A HaloTag® CMV*d2* Flexi® Vector	20 µg	G2861	193
pFN23K HaloTag® CMV*d2* Flexi® Vector	20 µg	G2871	193
pFN24A HaloTag® CMV*d3* Flexi® Vector	20 µg	G2881	193
pFN24K HaloTag® CMV*d3* Flexi® Vector	20 µg	G2981	193
pF9A CMV *hRluc*-neo Flexi® Vector	20 µg	C9361	251
pFN10A (ACT) Flexi® Vector	20 µg	C9331	251
pFN11A (BIND) Flexi® Vector	20 µg	C9341	251

Description: The Flexi® Vector System is a simple, yet powerful, directional cloning method for protein-coding sequences. It is based on two rare-cutting restriction enzymes, SgfI and PmeI, and provides a rapid, efficient and high-fidelity way to transfer protein-coding regions between a variety of Flexi® Vectors without the need to resequence.

All Flexi® Vectors carry the lethal barnase gene, which is replaced by the DNA fragment of interest and acts as a positive selection for the successful ligation of the insert.

Unlike site-specific recombination vector systems, the Flexi® Vector Systems do not require appending multiple amino acids to the amino or carboxy termini of the protein of interest. In addition, the systems do not require an archival entry vector, and most applications allow direct entry into the vector suited to the experimental design.

C-terminal Flexi® Vectors allow expression of C-terminal-tagged proteins. While these vectors can act as acceptors of protein-coding regions flanked by SgfI and PmeI, they lack a PmeI site and contain a different blunt-ended site, EcoICRI. This joined sequence cannot be removed from the C-terminal Flexi® Vectors and transferred to other Flexi® Vectors.

Features:

- **Versatility:** You can choose between a variety of initial applications (e.g., bacterial protein, mammalian, or cell-free protein expression) and then transfer to others as required.
- **Time Savings:** Efficient transfer allows for direct use of recombinant clones, minimizing time wasted screening background colonies.
- **Enhanced Productivity:** Adaptable to high-throughput formats for large screening projects.
- **Easy Access:** No licensing fees or complicated transfer restrictions.

Protocol	Part#
Technical Manual	TM254

Storage Conditions: Cat.# C8640 is comprised of Cat.# C8641 and A9280. Store Cat.# C8641 at –20°C; store Cat.# A9280 at room temperature. Store Cat.# C8820 and C9320 at –20°C. Store vectors and enzyme blend at –20°C.

pCMVTNT™ Vector

Product	Size	Cat.#	
pCMVTNT™ Vector	20 µg	L5620	188

Description: The pCMVTNT™ Vector is designed for the convenient expression of cloned genes using both in vivo and in vitro expression systems. Both SP6 and T7 polymerase promoters lie in tandem adjacent to the multiple cloning site. This allows for gene expression from either an SP6- or T7-based coupled in vitro transcription/translation system. The presence of RNA phage promoters also allows for the highly efficient synthesis of RNA in vitro. The pCMVTNT™ Vector also contains a 5′ β-globin leader sequence that has been referenced for enhanced expression of certain genes in vitro. For in vivo expression, the vector contains a CMV enhancer/promoter region, which allows strong constitutive expression in many cell types. A β-globin/IgG chimeric intron is located downstream from the enhancer/promoter region. The late SV40 polyadenylation site is located downstream of the multiple cloning site.

Features:

- **In Vivo Expression:** The CMV enhancer/promoter region allows strong constitutive expression in many cell types.
- **Flexible:** The vector contains tandem SP6 and T7 phage promoters allowing use in the appropriate in vitro translation or transcription system.
- **Convenient:** Multiple cloning site provides a selection of restriction sites for cloning.

Protocol	Part#
Technical Bulletin	TB305

Storage Conditions: Store at –20°C.

pTNT™ Vector

Product	Size	Cat.#	
pTNT™ Vector	20 µg	L5610	155

Description: The pTNT™ Vector is designed for the convenient in vitro expression of cloned genes. Both SP6 and T7 polymerase promoters lie in tandem adjacent to the multiple cloning site. This permits gene expression from either an SP6- or T7-based coupled in vitro transcription/translation system. The presence of RNA phage promoters also allows for the highly efficient synthesis of RNA in vitro. The pTNT™ Vector also contains a 5′ β-globin leader sequence and synthetic poly(A)$_{30}$ tail, both of which have been shown to enhance expression of certain genes.

Features:

- **Flexible:** The vector contains tandem SP6 and T7 phage promoters allowing use in the appropriate in vitro translation or transcription system.
- **Convenient:** Multiple cloning site provides a selection of restriction sites for cloning.

Protocol	Part#
Technical Bulletin	TB304

Storage Conditions: Store at –20°C.

pAdVAntage™ Vector

Product	Size	Cat.#	
pAdVAntage™ Vector	20 µg	E1711	98

Description: Co-transfection of mammalian cells with the pAdVAntage™ Vector enhances transient protein expression in a variety of cell types by increasing translation initiation.

Transfection of mammalian cells with an expression vector often results in suboptimal expression of the protein of interest. Double-stranded RNA (dsRNA) generated during transfection is thought to activate the dsRNA-activated inhibitor (DAI), one of several enzymes involved in the host cell's antiviral defense system. DAI phosphorylates the translation initiation factor eIF-2, halting translation and therefore protein production.

However, DAI translation inhibition can be overcome with the adenoviral Virus Associated I RNA (VAI RNA) produced by RNA polymerase III following co-transfection with the pAdVAntage™ Vector. The VAI RNA binds to DAI, preventing its activation, thereby allowing translation and protein expression.

Features:

- **Increased Expression:** Co-transfection of pAdVAntage™ Vector with luciferase constructs showed at least a tenfold increase in luciferase expression in 293 and HeLa cell lines over transfections performed with the construct DNA alone.
- **Flexible:** Can be used in a variety of cell lines.

Protocol	Part#
Technical Bulletin	TB207

Storage Conditions: Store at –20°C.

pSI Mammalian Expression Vector

Product	Size	Cat.#	
pSI Mammalian Expression Vector	20 µg	E1721	290

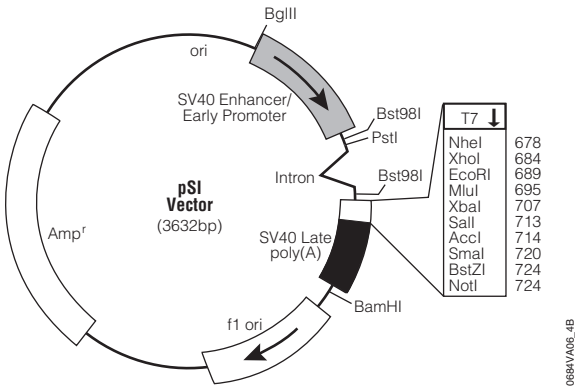
Description: The pSI Mammalian Expression Vector promotes constitutive expression of cloned DNA inserts in mammalian cells. The major difference between the pCI and pSI Mammalian Expression Vectors is the enhancer/promoter region controlling the expression of the inserted gene. The pSI Expression Vector contains the simian virus 40 (SV40) enhancer and early promoter region. This vector can be used for both transient and stable expression of genes. For stable expression, the pSI Vector must be co-transfected with an expression vector containing a selectable gene for mammalian cells.

Features:

- **Strong, Constitutive Expression:** The pSI Vector's SV40 enhancer/promoter region allows strong, constitutive expression in most cell lines. The vector is maintained as an episome in cells expressing the SV40 large T antigen, leading to even higher levels of expression. A β-globin/IgG chimeric intron located downstream from the enhancer/promoter region can further increase expression.
- **Increased Steady-State mRNA Levels:** The late SV40 polyadenylation signal increases the steady-state level of RNA approximately fivefold more than the early SV40 polyadenylation signal.
- **Convenient:** Multiple cloning sites exist for easy insertion of cDNA.
- **Versatile:** Synthesize transcripts in vitro using the T7 RNA polymerase promoter or generate single-stranded DNA in *E. coli* using the f1 origin of replication.

Protocol	Part#
Technical Bulletin	TB206

Storage Conditions: Store at –20°C.



pCI Mammalian Expression Vector

Product	Size	Cat.#	
pCI Mammalian Expression Vector	20 µg	E1731	290

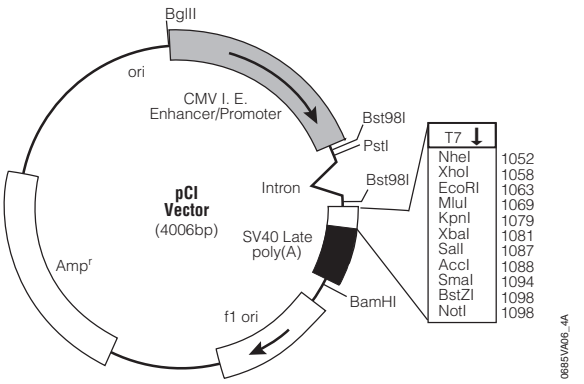
Description: The pCI Mammalian Expression Vector promotes constitutive expression of cloned DNA inserts in mammalian cells. The major difference between the pCI and pSI Mammalian Expression Vectors is the enhancer/promoter region controlling the expression of the inserted gene. The pCI Expression Vector contains the human cytomegalovirus (CMV) major immediate-early gene enhancer/promoter region. This vector can be used for both transient and stable expression of genes. For stable expression, the pCI Vector must be co-transfected with an expression vector containing a selectable gene for mammalian cells.

Features:

- **Strong, Constitutive Expression:** The pCI Vector's CMV enhancer/promoter region enables strong, constitutive expression in many cell types. A β-globin/IgG chimeric intron located downstream of the enhancer/promoter region can further increase expression.
- **Increased Steady-State mRNA Levels:** The late SV40 polyadenylation signal increases the steady-state level of RNA approximately fivefold more than the early SV40 polyadenylation signal.
- **Convenient:** Multiple cloning sites exist for easy insertion of cDNA.
- **Versatile:** Synthesize transcripts in vitro using the T7 RNA polymerase promoter or generate single-stranded DNA in *E. coli* using the f1 origin of replication.

Protocol	Part#
Technical Bulletin	TB206

Storage Conditions: Store at –20°C.



pCI-neo Mammalian Expression Vector

Product	Size	Cat.#	
pCI-neo Mammalian Expression Vector	20 µg	E1841	290

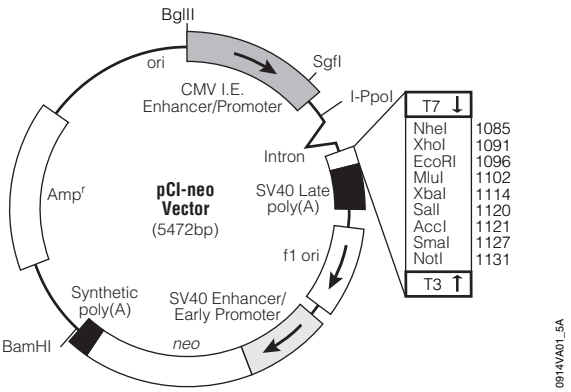
Description: The pCI-neo Mammalian Expression Vector carries the human cytomegalovirus (CMV) immediate-early enhancer/promoter region to promote constitutive expression of cloned DNA inserts in mammalian cells. This vector also contains the neomycin phosphotransferase gene, a selectable marker for mammalian cells. The pCI-neo Vector can be used for transient or stable expression by selecting transfected cells with the antibiotic G-418.

Features:

- **Strong, Constitutive Expression:** The human cytomegalovirus (CMV) immediate-early enhancer/promoter region produces strong, constitutive expression. A β-globin/IgG chimeric intron located downstream from the enhancer/promoter region can further increase expression. The vector is maintained as an episome in cells expressing the SV40 large T antigen, leading to even higher levels of expression.
- **Transient or Stable Expression:** The neomycin phosphotransferase gene allows selection of stable transfected cells.
- **Increased Steady-State mRNA Levels:** The late SV40 polyadenylation signal increases the steady-state level of RNA approximately fivefold more than the early SV40 polyadenylation signal.
- **Convenient:** Multiple cloning sites exist for easy insertion of cDNA.
- **Versatile:** Synthesize transcripts in vitro using the T7 RNA polymerase promoter or generate single-stranded DNA in *E. coli* using the f1 origin of replication.

Protocol	Part#
Technical Bulletin	TB215

Storage Conditions: Store at –20°C.



BL21(DE3)pLysS Competent Cells

Product	Size	Cat.#	
BL21(DE3)pLysS Competent Cells, >10^6cfu/µg	5 × 200 µl	L1191	142

Description: BL21(DE3)pLysS Competent Cells allow high-efficiency protein expression of any gene that is under the control of a T7 promoter and has a ribosome binding site. BL21(DE3)pLysS is lysogenic for λ-DE3, which contains the T7 bacteriophage gene I, encoding T7 RNA polymerase under the control of the *lac* UV5 promoter. BL21(DE3)pLysS also contains a plasmid, pLysS, which carries the gene encoding T7 lysozyme. T7 lysozyme lowers the background expression level of target genes under the control of the T7 promoter but does not interfere with the level of expression achieved following induction by IPTG. One milliliter is sufficient for ten transformations.

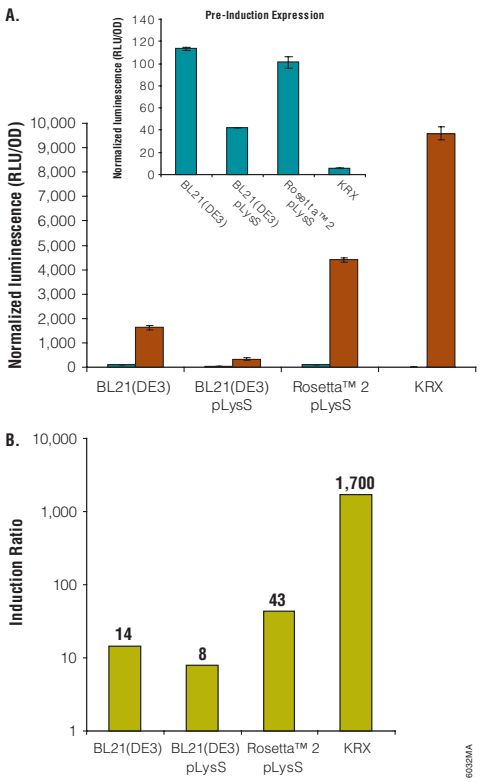
Genotype: F–, *ompT*, *hsd*S_B (r_B–, m_B–), *dcm*, *gal*, λ(DE3), pLysS, Cm^r.

Features:

- **T7 Promoter Expression:** Contains an IPTG-inducible gene for T7 RNA polymerase.
- **Convenient:** A time-saving alternative to making competent cells.

Protocol	Part#
Technical Bulletin	TB095

Storage Conditions: Store at –70°C.



Pre-induction and post-induction expression levels of firefly luciferase. Cells were transformed with the pF1K T7 Flexi® Vector containing the firefly luciferase gene.Cultures were grown at 37°C to an optical density ($O.D._{600}$) of 0.8–1.0 and then moved to a 25°C incubator shaker. When cultures reached an $O.D._{600}$ of 1.0–1.5, protein expression was induced using either 0.1% rhamnose or 1mM IPTG and grown overnight at 25°C.Samples for luciferase assays were removed prior to and after induction. **Panel A.** Firefly luciferase expression level was determined using the Bright-Glo™ Luciferase Assay Reagent. Pre- and post-induction firefly luciferase expression levels were normalized to cell number (n = 3). **Panel B.** Induction ratios were calculated by dividing the post-induction luminescence values by the pre-induction values.

Single Step (KRX) Competent Cells

Product	Size	Cat.#	
Single Step (KRX) Competent Cells	20 × 50 µl	L3002	243
Available Separately	**Size**	**Cat.#**	
L-Rhamnose Monohydrate	10 g	L5701	39
	50 g	L5702	138

Description: The Single Step (KRX) Competent Cells are designed for efficient transformation and tightly controlled protein expression. This one strain consolidates the best attributes of these two steps into one strain to evaluate protein expression in *E. coli*.

Transformation efficiencies are greater than 10^8 cfu/µg, similar to other highly competent cells. The single step cells are available in single transformation size (50µl). KRX also can be used for blue/white screening.

Single Step (KRX) is an *E. coli* K strain that contains a chromosomal copy of the T7 RNA polymerase driven by a rhamnose promoter (rhaBAD) to provide dramatic control of the proteins expressed via a T7 promoter. Pre-induced expression protein levels are significantly lower than those of BL21(DE3)-derived strains. This feature facilitates cloning and expression of proteins toxic to *E. coli*.

Genotype: [F′, *tra*D36, Δ*omp*P, *pro*A^+B^+, *lac*I^q, Δ(*lacZ*)M15] Δ*ompT*, *end*A1, *rec*A1, *gyr*A96 (Nal^r), *thi*-1, *hsd*R17 (r_k^-, m_k^+), e14$^-$ ($McrA^-$), *rel*A1, *sup*E44, Δ(*lac-pro*AB), Δ(*rha*BAD)::T7 RNA polymerase.

Features:

- **Save Time:** In two days, you can transform your vector into the Single Step (KRX) Competent Cells and be ready for protein expression.
- **Controlled Protein Expression:** For overall expression of cloned proteins, the Single Step (KRX) Competent Cells provide dramatic control of expressed protein-coding regions.
- **Achieve High Yields:** Protein expression levels were shown to be as high as or higher than levels expressed in BL21(DE3)-derived strains.
- **Blue/White Screening:** Convenient method for detecting recombinant clones.

Protocol	Part#
Technical Bulletin	TB352

Bacterial Strain BL21(DE3)pLysS

Product	Size	Cat.#	
Bacterial Strain BL21(DE3)pLysS, Glycerol Stock	500 µl	P9811	36

Description: Bacterial Strain BL21(DE3)pLysS allows high-efficiency protein expression of any gene that is under the control of a T7 promoter and has a ribosome binding site. BL21(DE3)pLysS is lysogenic for λ-DE3, which contains the T7 bacteriophage gene I, encoding T7 RNA polymerase under the control of the *lac* UV5 promoter. BL21(DE3)pLysS also contains a plasmid, pLysS, which carries the gene encoding T7 lysozyme. T7 lysozyme lowers the background expression level of target genes under the control of the T7 promoter but does not interfere with the level of expression achieved following induction by IPTG.

Genotype: F–, *ompT*, *hsd*S_B (r_B–,m_B–), *dcm*, *gal*, λ(DE3), pLysS, Cm^r.

Features:

- **T7 Promoter Expression:** Contains an IPTG-inducible gene for T7 RNA polymerase.

Storage Conditions: Store at –70°C.

RNA Polymerase Promoter Sequencing Primers

Product	Size	Conc.	Cat.#	
SP6 Promoter Primer	2 µg	10 µg/ml	Q5011	70
T7 Promoter Primer	2 µg	10 µg/ml	Q5021	70
T7 EEV Promoter Primer	2 µg	10 µg/ml	Q6700	63

Description: The SP6 and T7 Promoter Primers are designed for sequencing inserts cloned into the pGEM® Vectors. The SP6 Promoter Primer is designed for sequencing inserts cloned into the pALTER®-MAX and pCI-neo Vectors. The primers are designed to be annealed to single-stranded DNA or, after alkaline denaturation, to double-stranded DNA. The promoter primers are purified by gel electrophoresis or HPLC. The T7 EEV Promoter Primer is suitable for sequencing the pALTER®-MAX, pCMVTNT™ , pTNT™ and phMGFP Vectors, and the pCI/pSI series of mammalian expression vectors.

Primer Sequences
SP6: 5′-d(TATTTAGGTGACACTATAG)-3′
T7: 5′-d(TAATACGACTCACTATAGGG)-3′
T7 EEV: 5′-d(AAGGCTAGAGTACTTAATACGA)-3′

Storage Conditions: Store at –20°C.

pUC/M13 Sequencing Primers

Product	Size	Conc.	Cat.#	
pUC/M13 Primer, Forward (17mer)	2 µg	10 µg/ml	Q5391	70
pUC/M13 Primer, Reverse (17mer)	2 µg	10 µg/ml	Q5401	70
pUC/M13 Primer, Reverse (22mer)	2 µg	10 µg/ml	Q5421	70
pUC/M13 Primer, Forward (24mer)	2 µg	10 µg/ml	Q5601	70

Description: The pUC/M13 Primers are designed for sequencing inserts cloned into the M13 vectors and pUC plasmids developed by Messing. These primers also can be used for sequencing other *lacZ*-containing plasmids such as the pGEM®-Z and pGEM®-Zf Vectors. The primers are purified by gel electrophoresis or HPLC.

Primer Sequences
Forward (17mer): 5′-d(GTTTTCCCAGTCACGAC)-3′
Reverse (17mer): 5′-d(CAGGAAACAGCTATGAC)-3′
Reverse (22mer): 5′-d(TCACACAGGAAACAGCTATGAC)-3′
Forward (24mer): 5′-d(CGCCAGGGTTTTCCCAGTCACGAC)-3′

Storage Conditions: Store at –20°C. The primers are supplied in sterile water.

pTARGET™ Sequencing Primer

Product	Size	Cat.#	
pTARGET™ Sequencing Primer	2 µg	Q4461	68

Description: The pTARGET™ Sequencing Primer is designed for sequencing inserts cloned into the pTARGET™ Mammalian Expression Vector (Cat.# A1410). This sequencing primer hybridizes to the region of the *lacZ* gene from nucleotides 1367–1344 on the pTARGET™ Vector.

This primer can be used **only** for sequencing inserts in the pTARGET™ Vector. The primer sequence is **not** a binding site for any RNA polymerases and **cannot** be used to generate in vitro transcripts.

The sequence of the pTARGET™ Sequencing Primer is 5′-d(TTACGCCAAGTTA TTTAGGTGACA)-3′. It is supplied at a concentration of 10ng/µl (1.25pmol/µl) in sterile water.

Storage Conditions: Store at –20°C.

PinPoint™ Vector Sequencing Primer

Product	Size	Cat.#	
PinPoint™ Vector Sequencing Primer	2 µg	V4211	72
	2 µg	V4211	72

Description: The PinPoint™ Vector Sequencing Primer is designed for sequencing inserts cloned into the PinPoint™ Xa Vectors (components of Cat.# V2020). The primer hybridizes upstream of the Factor Xa site at nucleotides 325–343, approximately 40–50 base pairs upstream of the multiple cloning region and can be used to determine if an insert is cloned in-frame with the biotinylation purification tag of the PinPoint™ Xa Vectors. The sequence of the PinPoint™ Vector Sequencing Primer is 5′-d(CGTGACGCGGTGCAGGGCG)-3′. It is supplied dried.

Features:

- **Performance Tested:** The PinPoint™ Vector Sequencing Primer is tested in double-stranded sequencing reactions with circular PinPoint™ Vectors.

Storage Conditions: Store at –20°C.

TNT® T7 Insect Cell Extract Protein Expression System (cell free protein expression)

Product	Size	Cat.#	
TNT® T7 Insect Cell Extract Protein Expression System	10 reactions	L1101	150
	40 reactions	L1102	525
pF25A ICE T7 Flexi® Vector	20 µg	L1061	225
pF25K ICE T7 Flexi® Vector	20 µg	L1081	225

Description: The TNT® T7 Insect Cell Extract Protein Expression System is a convenient, quick, single-tube, coupled transcription/translation system for the cell-free expression of proteins. Protein synthesis reactions are initiated by the addition of a DNA template, eliminating the need for the time-consuming process of in vitro RNA synthesis.

The extract is made from the commonly used *Spodoptera frugiperda* Sf21 cell line. All components necessary for the transcription/translation are present in the TNT® T7 ICE Master Mix. To initiate protein synthesis, the only component that must be added is the DNA template. Reactions are incubated at 28–30°C and are complete within 4 hours.

Proteins are expressed from genes cloned downstream of the T7 promoter. Companion vectors have been designed to achieve optimal yield with this system (pF25A and pF25K). They contain untranslated region (UTR) sequences at the 5′ and 3′ ends of the gene coding region to enhance translation efficiency. Using the TNT® T7 Insect Cell Extract Protein Expression System and these vectors, 75µg/ml of functional protein can be produced.

Features:

- **Obtain Data Faster:** Protein is expressed in only 4 hours, not days as with cell-based expression.
- **Complete System:** No requirement to purchase additional reagents.
- **Achieve High Protein Yields:** Express up to 75µg/ml of protein for multiple applications.

Protocol	Part#
Technical Manual	TM305

Storage Conditions: Store at –70°C.

TnT® SP6 High-Yield Wheat Germ Protein Expression System (cell free protein expression)

Product	Size	Cat.#	
TnT® SP6 High-Yield Wheat Germ Protein Expression System	4 × 300 µl	L3260	528
	1 × 300 µl	L3261	164

Description: The TnT® SP6 High-Yield Wheat Germ Protein Expression System, based on an optimized wheat germ extract, is a single-tube, coupled transcription/translation system designed to express proteins in only two hours. Protein synthesized, in the range of 10–100µg/ml, can be used in multiple proteomic-based applications, as well as in high-throughput analysis.

All components necessary for transcription/translation are provided in the extract, with the exception of the plasmid DNA or PCR template. Optional protein-labeling reagents must also be supplied by the user.

Features:

- **Save Time:** You can generate protein in only two hours, as compared to days when using cell-based (*E. coli*) systems.
- **Choose Your Format:** Use plasmid or PCR-generated templates to generate protein.
- **Achieve High Yields:** Generate 10- to 20-fold more protein (10–100µg/ml) when compared to other cell-free systems.
- **Generate Usable Protein:** Generate soluble, full-length protein and avoid problems associated with *E. coli* systems.

Protocol	Part#
Technical Manual	TM282

Storage Conditions: Store at –70°C.

TnT® Quick Coupled Transcription/ Translation System (cell free protein expression)

Product	Size	Conc.	Cat.#	
TnT® T7 Quick Coupled Transcription/Translation System	40 reactions	–	L1170	470
TnT® T7 Quick Coupled Transcription/Translation System, Trial Size	5 reactions	–	L1171	128
TnT® SP6 Quick Coupled Transcription/Translation System	40 reactions	–	L2080	470
TnT® SP6 Quick Coupled Transcription/Translation System, Trial Size	5 reactions	–	L2081	
Available Separately	**Size**	**Conc.**	**Cat.#**	
Magnesium Acetate	100 µl	25 mM	L4581	
Potassium Chloride	200 µl	2.5 M	L4591	

Cat.# L1171, L4581, L4591 For Laboratory Use.

Description: The TnT® Quick Systems are convenient single-tube, coupled transcription/translation reactions for eukaryotic cell-free protein expression. These cell-free expression systems combine the RNA Polymerase, nucleotides, salts, amino acids and Recombinant RNasin® Ribonuclease Inhibitor with the reticulocyte lysate solution to form a single TnT® Quick Master Mix.

The TnT® Quick Coupled Transcription/Translation System is available in two configurations for the expression of genes cloned downstream from either the T7 or SP6 RNA polymerase promoters. To use these cell-free expression systems, 0.2–2.0µg of circular plasmid DNA containing a T7 or SP6 promoter, or a PCR-generated fragment containing a T7 promoter, is added to an aliquot of the TnT® Quick Master Mix and incubated in a 50µl reaction volume for 60–90 minutes at 30°C. The expression reaction produces significant quantities of protein for a variety of applications including GST pull-downs and gel shift assays.

Applications (of cell-free expression)

- GST pull-downs.
- Gel shift assays.
- Co-immunoprecipitation.
- Characterization of protein modifications.
- Protein:RNA interactions.
- Protein activity studies.
- Confirmation of gene products.
- Protein Truncation Test (PTT).

For more information about cell-free protein expression, see the Promega Protocols & Applications Guide at **www.promega.com/paguide**

Features:

- **Use in Multiple Applications:** The TnT® cell-free expression systems are widely used for protein:protein interaction, protein:nucleic acid interactions, and more.
- **Save Time:** Using a one-tube reaction, proteins are expressed in one hour, not days, as with in vivo methods.
- **Complete System:** All the reagents you need are provided (except radioisotopes).
- **Reliable:** Eliminate solubility issues by using a cell-free mammalian system.
- **Dependability You Can Count On:** The TnT® Systems are rigorously quality controlled to ensure the highest level of protein expression.

Protocol	Part#
Technical Manual	TM045

Storage Conditions: Store at –70°C. Do not freeze-thaw the lysate more than two times.

TnT® Coupled Reticulocyte Lysate Systems (cell free protein expression)

Product	Size	Cat.#
TnT® SP6 Coupled Reticulocyte Lysate System	40 reactions	L4600
TnT® SP6 Coupled Reticulocyte Lysate System, Trial Size	8 reactions	L4601
TnT® T7 Coupled Reticulocyte Lysate System	40 reactions	L4610
TnT® T7 Coupled Reticulocyte Lysate System, Trial Size	8 reactions	L4611
TnT® T3 Coupled Reticulocyte Lysate System	40 reactions	L4950
TnT® T7/T3 Coupled Reticulocyte Lysate System	40 reactions	L5010
TnT® T7/SP6 Coupled Reticulocyte Lysate System	40 reactions	L5020

Description: The TnT® Coupled Reticulocyte Lysate Systems offer researchers an alternative for eukaryotic cell-free protein expression: a single-tube, coupled transcription/translation system. The TnT® Lysate Systems greatly simplify the process and reduce the time required to obtain in vitro translation results. Standard rabbit reticulocyte lysate translations commonly use RNA synthesized in vitro from SP6, T3 or T7 RNA polymerase promoters and require three separate reactions with several steps between each reaction. The TnT® Systems bypass many of these steps by incorporating transcription directly in the translation mix.

Applications

- Protein:protein interactions.
- Protein:DNA interactions.
- Protein:RNA interactions.
- Protein activity studies.
- Confirmation of gene products.
- Characterization of protein modifications.
- Protein Truncation Test (PTT).

For more information, see the Promega Protocols & Applications Guide at: **www.promega.com/paguide**

Features:

- **Use in Multiple Applications:** The TnT® Systems are widely used for protein:protein interaction, protein:nucleic acid interactions, and more.
- **Save Time:** Using a one-tube reaction, proteins are generated in one hour, not days, as with in vivo methods.
- **Complete System:** All the reagents you need are provided (except radioisotopes).
- **Reliable:** Eliminate solubility issues by using an in vitro mammalian system.
- **Dependability You Can Count On:** The TnT® Systems are rigorously quality controlled to ensure the highest level of performance.

Protocol	Part#
Technical Bulletin	TB126

Storage Conditions: Store the polymerase at –20 to –70°C. Store Luciferase Assay Wells at room temperature. Store the other components at –70°C. Do not freeze-thaw the lysate more than two times.

TnT® Coupled Wheat Germ Extract System

Product	Size	Cat.#
TnT® T3 Coupled Wheat Germ Extract System	40 reactions	L4120
TnT® SP6 Coupled Wheat Germ Extract System	40 reactions	L4130
TnT® T7 Coupled Wheat Germ Extract System	40 reactions	L4140
TnT® T7/SP6 Coupled Wheat Germ Extract System	40 reactions	L5030
TnT® T7/T3 Coupled Wheat Germ Extract System	40 reactions	L5040

Cat.# L5030, 5040 For Laboratory Use.

Description: The TnT® Coupled Wheat Germ Extract Systems offer researchers an alternative for eukaryotic cell-free protein expression: a one-tube, coupled transcription/translation system. The TnT® Extract Systems greatly simplify the process and reduce the time required to obtain in vitro translation results. Standard wheat germ extract translations commonly use RNA synthesized in vitro from SP6, T3 or T7 RNA polymerase promoters. This entire process requires separate reactions with several steps between each reaction. The TnT® Extracts bypass many of these steps by incorporating transcription directly in the translation mix. Additionally, the TnT® Extract reactions often produce significantly more protein (two- to sixfold) in a 1.5-hour reaction than do standard in vitro wheat germ extract translations using RNA templates.

Magnesium Acetate, 25mM, and Potassium Chloride, 2.5M, can be used to optimize in vitro translation reactions in the TnT® T7 Quick Coupled Transcription/Translation System, Flexi® Rabbit Reticulocyte Lysate System and TnT® Coupled Wheat Germ Extract System.

Features:

- **Reliable:** The TnT® Systems are rigorously quality controlled to ensure the highest level of transcription/translation, whether your template is a linear (T3 or T7 only) or circular plasmid.
- **Convenient:** Single-tube procedure eliminates the time and effort required to prepare RNA for a standard wheat germ translation. Translation results can be visualized by autoradiography in 6–8 hours.
- **Versatile:** The T7 and T3 systems will produce protein from both linear and circular DNA (use only circular DNA with the SP6 system; for PCR templates use Cat.# L5540, TnT® T7 Quick for PCR DNA).
- **Controls Included:** Luciferase Control DNA and Luciferase Assay Reagents are included with the system as functional controls. Only full-length luciferase is active.

Protocol	Part#
Technical Bulletin	TB165

Storage Conditions: Store the polymerase at –20°C. Store the Luciferase Assay Wells at room temperature. Store the other components at –70°C. Avoid multiple freeze-thaw cycles.

Luciferase SP6/T7 Control DNAs

Product	Size	Cat.#
Luciferase SP6 Control DNA	20 µg	L4741
Luciferase T7 Control DNA	20 µg	L4821

Description: The Luciferase SP6 and T7 Control DNAs are used as functional controls in the TnT® Quick Coupled and TnT® Coupled Transcription/Translation Systems. The Control DNAs contain the gene for luciferase under transcriptional control of a phage RNA polymerase promoter. All constructs carry a 30-base pair poly[d(A)/d(T)] tail following the luciferase gene. Control reactions are monitored easily by the production of luminescence, which is generated from full-length luciferase and the addition of necessary components. Luciferase Control DNAs are supplied as 0.5mg/ml solutions in TE buffer.

Storage Conditions: Store at –20°C.

TnT® T7 Quick for PCR DNA

Product	Size	Cat.#
TnT® T7 Quick for PCR DNA	40 reactions	L5540
For Laboratory Use.		

Description: TnT® T7 Quick for PCR DNA is a rapid, convenient, coupled transcription/translation system designed for optimum protein expression from PCR templates. For most PCR templates, the TnT® T7 Quick for PCR DNA reactions produce up to 5 times more protein than other commercially available kits. The PCR-generated DNA can be used directly from the amplification reaction or purified by numerous commercially available kits and traditional methods.

Features:

- **Convenient:** Directly from PCR, no cleanup necessary.
- **High Yield:** Up to 5 times more expressed protein than standard translation reactions with linear templates.
- **Quick:** One-tube reaction.
- **Complete:** Reagents including Recombinant RNasin® Ribonuclease Inhibitor are included in the Quick Master Mix.
- **Good Value:** One-tube format means no leftover reagents.
- **Reliable:** The TnT® Systems are rigorously quality controlled to ensure the highest level of transcription/translation.

Protocol	Part#
Technical Manual	TM235

Storage Conditions: Store at –70°C. Do not freeze-thaw the Master Mix more than two times.

Rabbit Reticulocyte Lysate System, Nuclease Treated

Product	Size	Cat.#
Rabbit Reticulocyte Lysate System, Nuclease Treated	30 reactions	L4960
Each system contains 2 × 200µl of both Rabbit Reticulocyte Lysate and Wheat Germ Extract; this is sufficient for 12 reactions using the Rabbit Reticulocyte Lysate and 12 reactions using the Wheat Germ Extract.		

Description: Rabbit Reticulocyte Lysate Translation Systems are utilized in the identification of mRNA species, the characterization of their protein products and the investigation of transcriptional and translational control. Rabbit Reticulocyte Lysate is prepared from New Zealand white rabbits using a standard protocol that ensures reliable and consistent reticulocyte production in each lot. After the reticulocytes are lysed, the extract is treated with micrococcal nuclease to destroy endogenous mRNA and thus reduce background translation to a minimum. The lysate contains the cellular components necessary for protein synthesis (tRNA, ribosomes, amino acids, initiation, elongation and termination factors).

Features:

- **Consistent:** Reliable and consistent translation with each lot.
- **Optimized and Ready to Use:** The treated Rabbit Reticulocyte Lysate is optimized for translation and contains an energy-regenerating system (phosphocreatine/phosphocreatine kinase), a mixture of tRNAs (to expand the range of mRNAs that can be translated), hemin (to prevent inhibition of initiation), and potassium chloride and magnesium acetate.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Manual	TM232

Storage Conditions: Store at –70°C or below. Do not freeze-thaw the lysate more than two times.

Flexi® Rabbit Reticulocyte Lysate System

Product	Size	Cat.#
Flexi® Rabbit Reticulocyte Lysate System	30 reactions	L4540

Description: The Flexi® Rabbit Reticulocyte Lysate System allows translation reactions to be optimized for a wide range of parameters, including Mg^{2+} and K^{+} concentrations and the choice of adding DTT. To help optimize Mg^{2+} for a specific message, the endogenous Mg^{2+} concentration of each lysate batch is stated in the product information included with this product. The Flexi® System also offers the choice of three amino acid mixtures and includes a control RNA encoding the firefly luciferase gene.

Features:

- **Improved Efficiency:** In an optimized system, the quantity of protein produced can be increased as much as fourfold over that of a standard lysate reaction.
- **Easy Optimization:** To aid in optimizing magnesium concentrations, the endogenous magnesium concentration is provided for each lot of Flexi® Lysate.
- **Choice:** The Flexi® System contains three Amino Acid Mixtures, which enable different choices of radioisotopes.
- **Control Included:** Luciferase Control RNA and Luciferase Assay Reagent are included with the system as a functional control. Only full-length luciferase is active.

Protocol	Part#
Technical Bulletin	TB127

Storage Conditions: Store at –70°C, except Luciferase Assay Wells, which can be stored at room temperature. Do not freeze-thaw the lysate more than two times.

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Rabbit Reticulocyte Lysate/Wheat Germ Extract Combination System

Product	Size	Cat.#
Rabbit Reticulocyte Lysate/Wheat Germ Extract Combination System	24 reactions	L4330

Description: The Rabbit Reticulocyte Lysate/Wheat Germ Extract Combination System contains both Rabbit Reticulocyte Lysate and Wheat Germ Extract for comparing in vitro translation systems. Reticulocyte Lysate is prepared from New Zealand white rabbits. The Wheat Germ Extract is prepared by grinding wheat germ in an extraction buffer followed by centrifugation to remove cellular debris. Both systems contain the cellular components necessary for protein synthesis. The systems have been treated with micrococcal nuclease, which destroys endogenous mRNA and results in minimal background translation.

Features:

- **Choice:** Test both Rabbit Reticulocyte Lysate and Wheat Germ Systems to find optimal translation systems.
- **Consistent:** Rigorous quality control ensures minimal lot-to-lot variability.
- **Optimal Expression:** Potassium Acetate is provided to enhance the Wheat Germ Extract System for a wide range of mRNAs.

Protocol	Part#
Rabbit Reticulocyte Lysate System Technical Manual	TM232
Wheat Germ Extract Technical Manual	TM230

Storage Conditions: Store at –70°C or below. Do not freeze-thaw the lysate more than two times.

Wheat Germ Extract Plus

Product	Size	Cat.#
Wheat Germ Extract Plus	40 reactions	L3250
	10 reactions	L3251
Available Separately	**Size**	**Cat.#**
pF3A WG (BYDV) Flexi® Vector	20 µg	L5671
pF3K WG (BYDV) Flexi® Vector	20 µg	L5681

Description: Wheat Germ Extract Plus is a highly efficient, single-tube protein expression system designed to give yields in the range of 10–80µg/ml. The extract contains all the cellular components necessary for protein synthesis (tRNA, ribosomes, amino acids, and initiation, elongation and termination factors). Potassium acetate is also included at a concentration to enhance translation for a wide range of mRNAs. Protein synthesis is initiated by addition of the appropriate mRNA template, and the reaction is incubated for 2 hours at 25°C. Synthesized proteins can be analyzed by SDS-polyacrylamide gel electrophoresis or used directly in numerous applications. There is no requirement for dialysis or specific incubation equipment.

The **pF3A WG (BYDV) and pF3K WG (BYDV) Flexi® Vectors** are designed for in vitro expression in wheat germ extracts and are available for use with Wheat Germ Extract Plus. These vectors contain sequences from the barley yellow dwarf virus (BYDV), an RNA plant virus, upstream and downstream of the protein coding region of interest. The BYDV elements interact with each other, form a closed loop and act synergistically to stimulate translation in wheat germ extracts, bypassing mRNA cap and polyadenylation dependencies. See the *Flexi® Vector Systems Technical Manual* #TM254 for more information on the pF3A WG (BYDV) and pF3K WG (BYDV) Flexi® Vectors.

Features:

- **Save Time:** Generate proteins in a few hours rather than days.
- **Versatility to Suit Your Needs:** You can label, purify or use protein directly for many applications.
- **Increased Solubility:** No need to worry about solubility issues associated with *E. coli* systems.
- **Perform Expanded Applications:** Wheat Germ Extract Plus can generate up to 20-fold more protein (80µg/ml) than other standard wheat germ systems.
- **No Need to Purchase Expensive Instrumentation:** Use in batch format.

Protocol	Part#
Technical Manual	TM066

Storage Conditions: Store at –70°C.

Wheat Germ Extract

Product	Size	Cat.#
Wheat Germ Extract	5 × 200 µl	L4380

Description: Wheat Germ Extract contains the cellular components necessary for protein synthesis (tRNA, ribosomes, initiation, elongation and termination factors). Wheat Germ Extract is prepared by grinding wheat germ in an extraction buffer followed by centrifugation to remove cell debris. The supernatant is subjected to chromatography that separates endogenous amino acids and plant pigments from the extract. The extract is also treated with micrococcal nuclease to destroy endogenous mRNA and thus reduce background translation to a minimum.

Features:

- **Optimized:** Extract contains an energy-regenerating system (phosphocreatine/phosphocreatine kinase), spermidine (to stimulate the efficiency of chain elongation), magnesium acetate and potassium acetate.
- **Flexible:** Three Amino Acid Mixtures are provided, which enable different choices of radioisotopes.
- **Robust:** Potassium Acetate is provided to enhance translation for a wide range of mRNAs.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Manual	TM230

Storage Conditions: Store at –70°C or below. Avoid freeze-thaw cycles.

T7 Sample System

Product	Size	Cat.#
T7 Sample System	1 each	L5900

Description: The T7 Sample System is designed to facilitate the optimization of individual gene expression by offering four unique in vitro translation systems to evaluate. The system consists of samples of: TnT® T7 Quick for PCR DNA, TnT® T7 Quick Coupled Transcription/Translation System, TnT® Coupled Wheat Germ Extract System and *E. coli* T7 S30 Extract System for Circular DNA.

All of the coupled systems utilize RNA generated by a T7 phage promoter. Criteria such as post-translational modifications, ionic optimization and detection methods (i.e., non-isotopic) should be considered when choosing an in vitro system. In some cases only direct experimental results will confirm which system is best for specific genes.

Features:

- **Variety:** Four major in vitro translation systems to evaluate.
- **Value:** No requirement for the purchase of several large expensive systems.
- **Reliability:** Comprised of rigorously quality-controlled reagents to ensure the highest level of transcription/translation.
- **Optimization:** Determine which system is best for individual genes.

Protocol	Part#
Technical Bulletin	TB293

Storage Conditions: Store at –70°C.

Rabbit Reticulocyte Lysate, Untreated

Product	Size	Cat.#
Rabbit Reticulocyte Lysate, Untreated	1 ml	L4151

Description: Untreated Rabbit Reticulocyte Lysate contains the cellular components necessary for protein synthesis (tRNA, ribosomes, amino acids, initiation, elongation and termination factors) but has not been treated with micrococcal nuclease. Untreated Lysate is used primarily for the isolation of these components and as an abundant source of endogenous globin mRNA. Untreated Lysate is prepared from New Zealand white rabbits in the same manner as treated lysates with the exception that it is not treated with micrococcal nuclease.

Features:

- **Reliable:** Consistent reticulocyte production in each lot.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Manual	TM232

Storage Conditions: Store at –70°C or below.

S30 T7 High-Yield Protein Expression System (cell free expression)

Product	Size	Cat.#
S30 T7 High-Yield Protein Expression System	24 reactions	L1110
	8 reactions	L1115

Description: The *E. coli* S30 T7 High-Yield Protein Expression System is designed to express up to 500µg/ml of protein in 1 hour from plasmid vectors containing a T7 promoter and a ribosome binding site. The protein expression system provides an extract that contains T7 RNA polymerase for transcription and is deficient in OmpT endoproteinase and Ion protease activity. All other necessary components in the system are optimized for protein expression. This results in greater stability and enhanced expression of target proteins.

Features:

- **Obtain Data Faster:** Protein expression in only one hour, not days as with cell-based expression.
- **Complete System:** No requirement to purchase additional reagents.
- **Achieve High Protein Expression:** Express up to 500µg/ml of protein for multiple applications.
- **Scalable:** Convenient screening protocol for high-throughput protein expression.
- **Flexible:** Detect expressed proteins by Coomassie® staining or incorporation of a fluorescence or biotinylated modified tRNA.

Protocol	Part#
Technical Manual	TM306

Storage Conditions: Store at –70°C.

E. coli T7 S30 Extract System for Circular DNA

Product	Size	Cat.#
E. coli T7 S30 Extract System for Circular DNA	30 reactions	L1130

Description: The *E. coli* T7 S30 Extract System for Circular DNA simplifies the transcription/translation of DNA sequences cloned in plasmid or λ vectors containing a T7 promoter by providing an extract that contains T7 RNA polymerase for transcription and all components needed for translation. The investigator only supplies cloned DNA containing a T7 promoter and a ribosome binding site. This product is prepared by modifications of the method described by Zubay from an *E. coli* strain B deficient in *Omp*T endoproteinase and *lon* protease activity. This results in greater stability of expressed proteins that would otherwise be degraded by proteases if expressed in vivo.

Features:

- **Flexible:** Can translate using any clone that has a T7 promoter and a ribosome binding site. Other S30 extracts require an *E. coli* promoter.
- **Greater Stability:** Reduced chance of expressed proteins degrading.
- **Complete:** Contains all components needed for coupled transcription/translation.
- **Low Background:** Synthesizes very low levels of endogenous proteins.
- **Optimized:** Premix is optimized for each lot of S30 Extract and contains all other required components (except amino acids), such as ribonucleotides, tRNAs, PEP (phosphoenol pyruvate) and salts.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB219

Storage Conditions: Store extract at –70°C. Check individual components for storage temperatures.

E. coli S30 Extract System for Linear Templates

Product	Size	Cat.#
E. coli S30 Extract System for Linear Templates	30 reactions	L1030

Description: The *E. coli* S30 Extract System for Linear Templates is prepared using minor modifications of the protocol described by Lesley and colleagues and allows successful transcription/translation of linear DNA templates. The investigator need only provide linear DNA containing a prokaryotic *E. coli*-like promoter (such as *lac*UV5, *tac*, λPL (con) and λ-P_R). A ribosome binding site is required to direct the synthesis of proteins in vitro. In vitro-generated RNA from DNA templates lacking an *E. coli* promoter may also be used in this system, but protein yields will be decreased to 1–10% of that produced from linear DNA templates.

Features:

- **Flexible:** Many templates can be used: DNA fragments, PCR-synthesized DNA, ligated overlapping oligonucleotides, in vitro-generated RNA and prokaryotic RNA.
- **Greater Stability:** Reduced chance of expressed proteins degrading.
- **Complete:** Contains all necessary components for coupled transcription/translation.
- **Low Background:** System synthesizes very low levels of endogenous proteins.
- **Optimized:** Premix is optimized for each lot of S30 Extract and contains all other required components (except amino acids), such as ribonucleotides, tRNAs, PEP (phosphoenol pyruvate) and salts.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB102

Storage Conditions: Store at –70°C.

E. coli S30 Extract System for Circular DNA

Product	Size	Cat.#
E. coli S30 Extract System for Circular DNA	30 reactions	L1020

Description: The *E. coli* S30 Extract for Circular DNA simplifies the transcription/translation of DNA sequences cloned in plasmid or λ vectors, providing a powerful tool for identifying and characterizing polypeptides. The investigator needs only to supply the cloned DNA containing the appropriate prokaryotic promoter and ribosome binding sites. The S30 Extract for Circular DNA Templates is prepared by modifications of the method described by Zubay from an *E. coli* strain B deficient in *Omp*T endoproteinase and *lon* protease activity. This results in a greater stability of expressed proteins that would otherwise be degraded by proteases if expressed in vivo. The S30 in vitro system also allows higher expression levels of proteins that are normally expressed at low levels in vivo due to the action of host-encoded repressors.

Features:

- **Greater Stability:** Reduced chance of expressed proteins degrading.
- **Complete:** Contains all necessary components for coupled transcription/translation.
- **Low Background:** System synthesizes very low levels of endogenous proteins.
- **Optimized:** Premix is optimized for each lot of S30 Extract and contains all other required components (except amino acids), such as ribonucleotides, tRNAs, PEP (phosphoenol pyruvate) and salts.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB092

Storage Conditions: Store at –70°C.

Canine Pancreatic Microsomal Membranes

Product	Size	Cat.#
Canine Pancreatic Microsomal Membranes	50 µl	Y4041

Description: Microsomal vesicles are used to study co-translational and initial post-translational processing of proteins. Processing events such as signal peptide cleavage, membrane insertion, translocation and core glycosylation can be examined by the translation of the appropriate mRNA in vitro in the presence of these microsomal membranes. In addition, processing and glycosylation events may be studied by the transcription/translation of the appropriate DNA in the TNT® Lysate Systems when used with Canine Pancreatic Microsomal Membranes. To assure consistent performance with minimal translational inhibition and background, microsomes have been isolated free from contaminating membrane fractions and stripped of endogenous membrane-bound ribosomes and mRNA. Membrane preparations are assayed for both signal peptidase and core glycosylation activities using two different control mRNAs. The two control mRNAs supplied with this system are the precursor for β-lactamase (or ampicillin resistance gene product) from *E. coli* and the precursor for α-mating factor (or α-factor gene product) from *S. cerevisiae.*

The Signal Sequence Control mRNA (*E. coli* β-lactamase) is transcribed by SP6 RNA polymerase from a plasmid bearing the coding region for the *E. coli* gene encoding the precursor to β-lactamase (the ampicillin resistance gene product). The RNA is synthesized without a cap analog. This control mRNA is used to assay for signal peptidase activity and is supplied with the Canine Pancreatic Microsomal Membranes System.

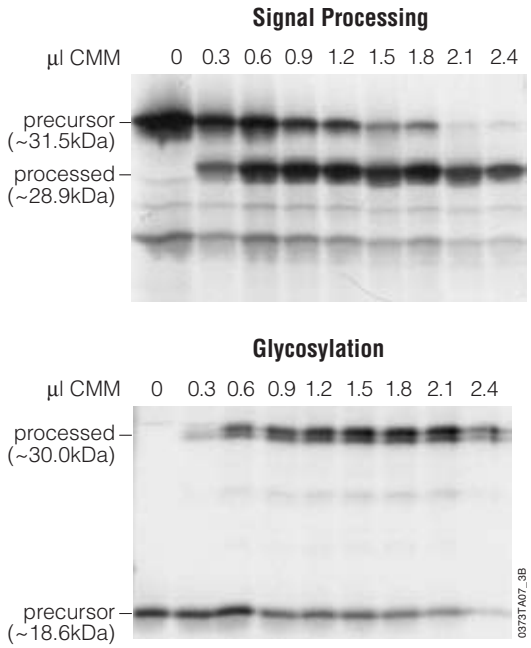
The Core Glycosylation Control mRNA (*S. cerevisiae* α-factor) is transcribed by SP6 RNA polymerase from a plasmid bearing the coding region for the *S. cerevisiae* α-mating factor. The RNA is synthesized without a cap analog. This control mRNA is used to assay for core glycosylation activity and is supplied with the Canine Pancreatic Microsomal Membranes System.

Features:

- **Reliable:** Microsomes are stripped of endogenous membrane-bound ribosomes and mRNA to ensure consistent performance with minimal translational inhibition and background. Performance tested in rabbit reticulocyte lysate.

Protocol	Part#
Technical Manual	TM231

Storage Conditions: Store at –70°C or below. Membranes are stable at –70°C for 1 year. After thawing, unused portions should be rapidly refrozen in liquid nitrogen. No detectable loss of activity results after two freeze-thaw cycles.



Processing and glycosylation activity of Canine Pancreatic Microsomal Membranes (CMM). The positive control mRNAs (0.5µg each of *E.coli* β-lactamase and *S.cerevisiae* α-factor) were translated using Rabbit Reticulocyte Lysate in a 25µl reaction for 60 minutes in the presence of the indicated amounts of CMM (3µl).Translation products were analyzed by gel electrophoresis followed by autoradiography of the [^{35}S]-labeled proteins.

Amino Acid Mixtures

Product	Size	Conc.	Cat.#
Amino Acid Mixture, Complete	175 µl	1 mM	L4461
Amino Acid Mixture Minus Cysteine	175 µl	1 mM	L4471
Amino Acid Mixture Minus Methionine and Cysteine	175 µl	1 mM	L5511
Amino Acid Mixture Minus Leucine	175 µl	1 mM	L9951
Amino Acid Mixture Minus Methionine	175 µl	1 mM	L9961

For Laboratory Use.

Description: The Amino Acid Mixture, Complete, is an aqueous solution containing 1mM each of the 20 essential amino acids. This mixture is compatible for use in the Flexi® Lysate, TNT® Lysate and standard Rabbit Reticulocyte Lysate Systems as well as in the Wheat Germ Extract and *E. coli* S30 Systems. Amino Acid Mixtures are also available lacking cysteine, methionine and cysteine, leucine or methionine.

Storage Conditions: Store at –70°C.

pGEM® β-Gal Control DNA

Product	Size	Cat.#
pGEM® β-Gal Control DNA	20 µg	L4731

Description: pGEM® β-Gal Control DNA contains the coding sequence of β-galactosidase downstream of an *E. coli* wildtype *lac*Z promoter. pGEM® β-Gal Control DNA can be used as a positive control in the *E. coli* S30 Extract System for Circular DNA. The wildtype *lac*Z promoter is not efficient for initiating transcription from a linear DNA template. Supplied as a 0.5mg/ml solution in TE buffer.

Storage Conditions: Store at –20°C.

Luciferase Control RNA

Product	Size	Conc.	Cat.#
Luciferase Control RNA	20 µg	1 mg/ml	L4561

Description: Luciferase Control RNA is a unique functional control for in vitro translation reactions. Luciferase Control RNA is an uncapped in vitro-transcribed RNA containing a 30-base poly(A) tail that produces functional luciferase when translated. Control reactions are monitored easily by a luciferase assay for the production of luminescence generated from the full-length luciferase.

Features:

- **Convenient:** Control reactions are easily monitored by a luciferase assay for luminescence.
- **Safe:** Non-radioactive format to monitor control activity.

Storage Conditions: Store at –70°C.

Regulated Mammalian Expression System

Product	Size	Cat.#
Regulated Mammalian Expression System	1 system	C9470
pReg neo Vector	20 µg	C9421
pF12A RM Flexi® Vector	20 µg	C9431
pF12K RM Flexi® Vector	20 µg	C9441
Available Separately	**Size**	**Cat.#**
Coumermycin A1	5 mg	C9451
Novobiocin Sodium Salt	1 g	C9461

Description: The Regulated Mammalian Expression System features low basal levels, robust and rapid induction, and downregulation of gene expression in mammalian cells. The Regulated Mammalian Expression System is based on a novel on/off switch that relies on the rapid and sensitive modulation by coumerin-related compounds of a chimeric transactivator protein. Nanomolar concentrations of the antibiotic coumermycin promote homodimerization of a chimeric transactivator that, in turn, binds to lambda operator sequences located upstream of a minimal promoter driving transcription of coding sequences for a protein of interest. The levels of protein expression can be regulated by adjusting the coumermycin concentration. More significantly, this expression can be promptly and effectively switched off by adding novobiocin, which acts as an antagonist by dissociating the dimerized transactivator protein.

The protein coding region of interest is cloned into either the pF12A RM Flexi® Vector or pF12K RM Flexi® Vector, both of which are specially designed for Regulated Mammalian (RM) protein expression. These vectors incorporate regulatory promoter sequences upstream of the protein-coding region and are compatible with the Flexi® Vector System. In transient transfection paradigms, the pF12A or pF12K RM Flexi® Vector containing the protein-coding region of interest is co-transfected into mammalian cells together with the pReg neo Vector. The pReg neo Vector is designed to express a chimeric transactivator protein that interacts with the regulatory promoter region in the pF12A and pF12K RM Flexi® Vectors in a regulated fashion in response to coumermycin and novobiocin. Additionally, the pReg neo Vector encodes a neomycin phosphotransferase gene that allows stable cell selection and generation with the antibiotic G-418.

Features:

- **Enhanced Data:** High level of controlled induction combined with low basal protein expression.
- **Regulated Expression:** Dose-response induction of protein expression; rapid and sensitive on/off switch for protein expression.
- **Versatility:** Compatible with other Flexi® Vectors.

Protocol	Part#
Technical Manual	TM289

Storage Conditions: Store at –20°C.

HaloTag® Protein Purification System

Product	Size	Cat.#	
HaloTag® Protein Purification System	1 each	G6280	
HaloTag® Protein Purification System Sample Pack	1 each	G6270	
Available Separately	**Size Conc.**	**Cat.#**	
HaloTag® TMR Ligand	15 µl 5 mM	G8252	249
pFN18A HaloTag® T7 Flexi® Vector	20 µg	G2751	193
pFN18K HaloTag® T7 Flexi® Vector	20 µg	G2681	193
Single Step (KRX) Competent Cells	20 × 50 µl	L3002	243

Cat.# G6280 contains 100ml of a 25% slurry of HaloLink™ Resin, TEV Protease and HisLink™ Resin. Cat.# G6270 contains 10ml of a 25% slurry of HaloLink™ Resin, TEV Protease and HisLink™ Resin.

Competitive pricing is available for bulk quantities of HaloLink™ Resin (e.g., more than 100 ml). Please inquire.

Description: The HaloTag® Protein Purification System is designed to purify proteins fused to HaloTag®, a novel protein tag that enhances the expression and solubility of recombinant proteins. HaloTag® Technology enables the covalent, efficient and specific capture of a protein of interest onto HaloLink™ Resin, thus overcoming the equilibrium-based limitations associated with affinity tags (i.e., poor capture of proteins expressed at low levels and protein loss during washing of the purification resin.

The HaloTag® technology offers a quick and convenient way to test protein expression of HaloTag® fusion proteins as well as monitor the efficiency of immobilization to HaloLink™ Resin by labeling with fluorescent HaloTag® TMR Ligand followed by SDS-PAGE analysis. Guidelines can be found in the HaloLink™ Resin Technical Manual #TM250, the HaloLink™ Protein Array Technical Manual #TM310, and the HaloCHIP™ System Technical Manual #TM075.

Outline of Procedure

The HaloTag® protein, a 34kDa mutated hydrolase, covalently attaches to HaloLink™ Resin via an immobilized chloroalkane ligand. TEV Protease cleaves the target protein from the HaloLink™ Resin. The TEV Protease, which has an N-terminal (HQ) tag, is removed from the protein of interest using HisLink™ Resin, and the purified protein of interest is recovered. The appropriate vector that encodes the HaloTag® protein and expresses protein optimally in *E. coli* is pFN18A HaloTag® T7 Flexi® Vector (G2751) or pFN18K HaloTag® T7 Flexi® Vector (G2681). These vectors can be purchased separately.

Features:

- **Experience Superior Yield, Purity and Specific Activity of Soluble, Functional Proteins Compared to His-Tag, GST and MBP Affinity Tags:** Specific and covalent HaloTag® fusion protein capture and immobilization on HaloLink™ Resin.
- **Achieve Enhanced Target Protein Expression in Prokaryotic, Mammalian and Cell-Free Systems:** Proteins are expressed as HaloTag® fusion proteins.
- **Purify Poorly Expressed Fusion Proteins:** Rapid, specific and covalent capture of HaloTag® protein onto HaloLink™ Resin is a nonequilibrium process.
- **Efficiently Recover Tag-Free Target Protein using TEV Protease Cleavage:** Optimized TEV protease recognition site within the interconnecting polypeptide separating the HaloTag® protein and the fusion partner. HaloTag® protein remains immobilized on the resin due to covalent capture.
- **Save Time:** One buffer compatible with downstream applications for all purification steps.
- **Perform Easy In-Gel Detection and Quantification of Protein Expression Levels with Fluorescent HaloTag® Ligands:** Highly stable HaloTag® protein-ligand interaction permits boiling with SDS sample buffer followed by resolving on SDS-PAGE.

Protocol	Part#
Technical Manual	TM312

Storage Conditions: Store the HaloLink™ Resin and HisLink™ Resin at 4°C. Do not freeze the resins. Store the TEV Protease at –20°C.

Anti-HaloTag® pAb

Product	Size	Conc.	Cat.#
Anti-HaloTag® pAb	200 µg	1 mg/ml	G9281

Description: The Anti-HaloTag® pAb is a purified rabbit polyclonal antibody raised against the HaloTag® protein. The antibody is purified using Protein G affinity resin and supplied at 1mg/ml in PBS. The antibody detects HaloTag® fusion proteins in Western blot hybridization and immunocytochemistry applications with high sensitivity and specificity. The HaloTag® protein is not endogenous to mammalian, plant and *E. coli* cells. *E. coli* and mammalian cell extracts demonstrate low cross-reactivity with the Anti-HaloTag® pAb.

Features:

- **Specificity:** The Anti-HaloTag® pAb is specific for HaloTag® protein and exhibits low cross-reactivity with *E. coli* and mammalian cell extracts.

Protocol	Part#
Technical Manual	TM260

Storage Conditions: Store at –20°C.

ProTEV Protease

Product	Size	Conc.	Cat.#
ProTEV Protease	1,000 u	10 u/µl	V6051
	10,000 u	10 u/µl	V6052

Description: The ProTEV Protease is an improved 50kDa version of the Nla protease from Tobacco Etch Virus (TEV), which has been engineered to be more stable than native TEV protease for prolonged enzymatic stability. The protease is used to cleave affinity tags from fusion proteins after protein purification. ProTEV Protease is highly site-specific, recognizing the seven amino acid sequence EXXYXQ(G/S), most commonly ENLYFQG, with cleavage occurring between glutamine and glycine or serine (see figures). The protease cleaves sequences with a variety of amino acids at the G/S (or P1′) position, giving the option to place the desired N-terminal amino acid on the C-terminal fusion partner after cleavage. Optimum activity is obtained at pH 7.0 and 30°C, but ProTEV Protease is active over a wide range of pH values (5.5–8.5) and temperatures (4–30°C), allowing the choice of conditions amenable to the protein of interest. ProTEV Protease is easily removed after cleavage using the HQ tag located at the N-terminus of the protein. It also can be used to cleave the protein of interest from its affinity tag while still immobilized on the affinity resin.

Features:

- **Active Over a Wide Range of pH and Temperatures:** Cleave individual fusion proteins using optimal conditions to maintain activity and correct conformation.
- **HQ-Tagged:** Convenient removal of ProTEV Protease using Ni-based affinity resins after cleavage.
- **Cleaves Fusion Proteins Directly in Solution or Immobilized on Affinity Resins:** ProTEV Protease is easy to use in multiple experimental formats.

Protocol	Part#
Promega Product Information	9PIV605

Storage Conditions: Store at –20°C.

Maxwell® 16 Polyhistidine Protein Purification Kit

Product	Size	Cat.#
Maxwell® 16 Polyhistidine Protein Purification Kit	48 preps	AS1060
Available Separately	**Size**	**Cat.#**
Maxwell® 16 Instrument	1 each	AS2000

Cat.# AS2000 For Laboratory Use.

Description: The Maxwell® 16 Polyhistidine Protein Purification Kit is used with the Maxwell® 16 Instrument to provide an easy method for the efficient, automated purification of polyhistidine-tagged protein from bacterial cultures and other sample types including mammalian and insect cells. With minor modifications, the reagents can also be used for purification of HQ-tagged proteins from bacterial cultures.

The Maxwell® Instrument is supplied with a preprogrammed purification procedure and reagent cartridges specifically designed to maximize simplicity and convenience. The instrument can process up to 16 samples in approximately 40 minutes. The purified protein is compatible with downstream applications such as gel electrophoresis and Western blot analysis.

Features:

- **Choose Your Sample Type:** Flexibility to purify from multiple starting cultures including bacterial culture, mammalian cells, insect cells and culture medium.
- **Have Confidence in Your Results:** Achieve consistent purification across all samples.
- **Save Hands-On Time:** Prefilled cartridges eliminate reagent preparation, multiple pipetting steps, centrifugation and additional sample manipulation.

Protocol	Part#
Technical Manual	TM285

Storage Conditions: Store at 4°C.

MagneGST™ Protein Purification System

Product	Size	Cat.#
MagneGST™ Protein Purification System	40 reactions	V8600
	200 reactions	V8603
Available Separately	**Size**	**Cat.#**
MagneGST™ Glutathione Particles	4 ml	V8611
	20 ml	V8612

This product requires the use of a magnetic stand.

Description: The MagneGST™ Protein Purification System provides a simple, rapid and reliable method for the purification of glutathione-S-transferase (GST) fusion proteins. Immobilized glutathione paramagnetic particles (MagneGST™ Particles) are used to isolate GST-tagged protein directly from a crude or cleared lysate using either a manual or automated procedure and requires use of a magnetic stand. GST-tagged proteins can be purified on a small scale from 1ml of culture or on a large scale using more than 50ml of culture. Samples also can be processed using a robotic platform.

Features:

- **Simple:** One-step purification of multiple samples with easy handling.
- **Quick:** After cell lysis, no requirement for high-speed centrifugation to clear lysate.
- **Scalable:** Scalable protocol using 1–50ml of cell culture.
- **Efficient:** Achieve high yields with little or no nonspecific background.
- **Complete:** System contains all necessary components except magnetic stand.

Protocol	Part#
Technical Manual	TM240

Storage Conditions: The complete system consists of two individual parts, each with a different storage condition. Store individual boxes at specified temperatures of 4°C and –70°C.

MagneHis™ Protein Purification System

Product	Size	Cat.#
MagneHis™ Protein Purification System	65 reactions	V8500
	325 reactions	V8550
Available Separately	**Size**	**Cat.#**
MagneHis™ Ni-Particles	2 ml	V8560
	10 ml	V8565

This product requires the use of a magnetic stand.

Description: The MagneHis™ Protein Purification System provides a simple, rapid and reliable method for the purification of polyhistidine- or HQ-tagged, expressed proteins. Paramagnetic precharged nickel particles (MagneHis™ Ni-Particles) are used to isolate polyhistidine- or HQ-tagged protein directly from a crude cell lysate using either a manual (requires use of a magnetic stand) or automated procedure. Using a tube format, polyhistidine- or HQ-tagged protein can be purified on a small scale using less than 1ml of culture or on a large scale using more than 1 liter of culture. Samples can be processed in a high-throughput manner using a robotic platform such as the Beckman Coulter Biomek® 2000 or FX.

Features:

- **Simple:** No centrifugation or vacuum is required once the cells are lysed.
- **Flexible:** MagneHis™ Ni-Particles are compatible with a variety of common buffers.
- **Quick:** No long incubations with lysozyme are required for cell lysis.
- **Scalable:** Volumes can be adjusted to correspond to the amount of material to be purified, 1ml to 1 liter of cell culture.
- **Efficient:** Binding capacity is up to 1mg of polyhistidine-tagged protein per 1ml of MagneHis™ Ni-Particles.
- **Versatile:** Purification can be performed manually (requires use of a magnetic stand) or using an automated platform.
- **Convenient:** Complete system that includes all necessary components, including a unique lysis buffer.
- **Automate This Assay:** Validated automated methods available for Beckman Coulter Biomek® 2000 and FX and Tecan Genesis® RSP at: **www.promega.com/automethods/**

Protocol	Part#
Technical Manual	TM060

Storage Conditions: Store at 4°C.

MagZ™ Protein Purification System

Product	Size	Cat.#
MagZ™ Protein Purification System	30 reactions	V8830

This product requires the use of a magnetic stand.

Description: The MagZ™ Protein Purification System provides a simple, rapid and reliable method for the purification of expressed polyhistidine- or HQ-tagged proteins, which are 99% free of hemoglobin contamination, from rabbit reticulocyte lysate. Based on the use of proprietary, paramagnetic precharged particles, polyhistidine- or HQ-tagged protein can be isolated from 50–500µl of TnT® Coupled Transcription/Translation reactions. Polyhistidine- or HQ-tagged proteins bind to the particles in minutes, while unbound proteins are washed away, and the target protein is eluted with imidazole.

Features:

- **Specific:** Minimal hemoglobin
- **Quick:** No long incubations are required.
- **Versatile:** Binding/wash and elution conditions can be further optimized for individual polyhistidine- or HQ-tagged proteins.

Protocol	Part#
Technical Bulletin	TB336

Storage Conditions: Store at 4°C.

FastBreak™ Cell Lysis Reagent

Product	Size	Cat.#
FastBreak™ Cell Lysis Reagent, 10X	15 ml	V8571
	60 ml	V8572
	100 ml	V8573

Description: FastBreak™ Cell Lysis Reagent is designed for the efficient, gentle lysis of *E. coli* cultures without the need for centrifugation or mechanical cell disruption. The reagent is provided as a 10X concentrate and contains a proprietary nonionic detergent to facilitate lysis. Add the reagent directly to *E. coli* cultures. Following a brief incubation, the cells are disrupted, and the protein of interest is released. Recombinant proteins can be directly screened in the cell extract or purified by the addition of the appropriate affinity matrix such as the MagneHis™ Protein Purification System. This product is suitable for both manual and automated protocols.

Features:

- **Save Time:** Eliminate centrifugation or mechanical disruption.
- **Easy to Use:** Add and incubate.
- **Flexible:** Use manually or on a robotic platform.

Protocol	Part#
Promega Product Information	9PIV857

Storage Conditions: Store at 4–25°C.

HisLink™ Protein Purification Resin

Product	Size	Cat.#
HisLink™ Protein Purification Resin	5 ml	V8823
	50 ml	V8821

Volumes above represent total volume of a 50% resin slurry.

Description: HisLink™ Protein Purification Resin is a macroporous silica resin modified to contain a high level of tetradentate chelated nickel (>20mmol Ni/ml settled resin). The resin is designed to efficiently capture and purify bacterially expressed polyhistidine- or HQ-tagged proteins. The HisLink™ Resin also may be used for general applications requiring an immobilized metal affinity chromatography (IMAC) matrix. HisLink™ Resin may be used in either column or batch purification formats. The lysate flows by gravity over a column of packed HisLink™ Resin at a rate sufficient for complete capture and efficient elution of polyhistidine- or HQ-tagged proteins from cleared lysate. The resin may be used in vacuum filtration devices (e.g., Vac-Man® Vacuum Manifold [Cat.# A7231]), allowing simultaneous processing of multiple columns. HisLink™ Resin is also an excellent choice for affinity purification of polyhistidine- or HQ-tagged proteins with low- to medium-pressure liquid chromatography systems such as FPLC. In batch format, HisLink™ Resin may be separated easily from the lysate without filtration (or centrifugation), allowing processing of larger quantities of lysate and the ability to purify protein without clearing the lysate of insoluble cellular debris.

Features:

- **Optimize Yields:** High binding capacity (>15mg/ml).
- **Save Time:** Purify polyhistidine- or HQ-tagged proteins from cleared or crude cell lysates.
- **Flexible:** Use standard gravity column chromatography or automated applications such as FPLC.

Protocol	Part#
Technical Bulletin	TB327

Storage Conditions: Store at 4°C.

HisLink™ 96 Protein Purification System

Product	Size	Cat.#
HisLink™ 96 Protein Purification System	1 × 96	V3680
	5 × 96	V3681

Description: HisLink™ 96 Protein Purification System provides a simple, quick and robust method of purifying multiple polyhistidine- or HQ-tagged expressed proteins from *E. coli* using a vacuum-based method. The system is designed to purify expressed polyhistidine- or HQ-tagged proteins directly from deep-well, 96-well plates. The HisLink™ 96 System is amenable to manual or automated methods, such as the Beckman Coulter Biomek® 2000 or FX for high-throughput applications.

Using the provided FastBreak™ Cell Lysis Reagent, bacterial cells containing polyhistidine- or HQ-tagged protein are lysed directly in culture. The HisLink™ Resin is added directly to the lysate, and the polyhistidine- or HQ-tagged proteins bind in a matter of minutes. Samples are then transferred to a filtration plate, unbound proteins are washed away and the target protein is recovered by elution with imidazole.

Features:

- **Simple:** No centrifugation required; lysis buffer is added directly to cells in culture medium.
- **Quick:** No long lysozyme incubations are required for cell lysis.
- **Versatile:** Perform purification manually or on an automated platform.
- **Efficient:** Binding capacity of 1mg of polyhistidine-tagged protein per well.

Protocol	Part#
Technical Bulletin	TB342

Storage Conditions: Store the system at 4°C. Plates may be stored at 4°C or room temperature.

HisLink™ Spin Protein Purification System

Product	Size	Cat.#
HisLink™ Spin Protein Purification System	25 reactions	V1320

Description: The HisLink™ Spin Protein Purification System provides a simple, quick and robust method of purifying polyhistidine- or HQ-tagged expressed proteins from *E. coli* using either a centrifuge- or vacuum-based method.

Proteins can be purified directly from culture medium containing bacterial cells expressing polyhistidine- or HQ-tagged protein. The bacterial cells are lysed using the FastBreak™ Cell Lysis Reagent, followed immediately by the addition of the HisLink™ Protein Purification Resin to the culture. The addition of these reagents results in simultaneous bacterial lysis and binding of the polyhistidine- or HQ-tagged proteins. Samples are then transferred to a HisLink™ Spin Column, where unbound protein is washed away and the target protein of interest is recovered by elution.

Features:

- **Versatile:** Choose between vacuum or centrifugation formats.
- **Convenient:** Complete system containing all necessary components.
- **Productive:** High binding capacity per column.

Protocol	Part#
Technical Manual	TM281

Storage Conditions: Store the system at 4°C. Store Spin Columns, Collection Tubes and FastBreak™ Cell Lysis Reagent at room temperature. After reconstitution, store the DNase I in aliquots at –20°C.

SoftLink™ Soft Release Avidin Resin

Product	Size	Cat.#
SoftLink™ Soft Release Avidin Resin	1 ml	V2011
	5 ml	V2012

Description: SoftLink™ Avidin Resin can be used for the isolation and purification of biotinylated molecules. SoftLink™ Resin is a rigid, methacrylate polymeric gel filtration matrix, functionalized with covalently bound, monomeric avidin. Monomeric avidin binds biotin with a K_d value of 10^{-7}M, allowing reversible binding of bound biotinylated proteins under mild elution conditions. Native, or tetrameric, avidin binds biotin with a very strong affinity ($K_d = 10^{-15}$M), which in turn requires strong denaturing conditions for eluting bound material. Monomeric avidin allows the specificity of capture but also the mildness of release appropriate for the purification of sensitive biological materials.

Features:

- **Sensitive:** Binds 20–40nmol of biotinylated protein per milliliter of resin.
- **Easy to Use:** Bound biotinylated molecules can be eluted under mild nondenaturing conditions (5mM biotin).
- **Versatile:** Retains biotin binding ability following exposure to a wide range of pH, low or high ionic strength, 6M guanidine and 1% SDS.
- **Reusable:** Regenerates at least 10 times without loss of binding capacity.
- **Robust:** Supports high flow rates (300cm/hour) and centrifugal forces (1,500 × *g*) in batch applications.
- **Flexible:** Purifications by batch or column method.

Protocol	Part#
Promega Product Information	9PIV201

Storage Conditions: Store at 4°C.

TetraLink™ Tetrameric Avidin Resin

Product	Size	Cat.#
TetraLink™ Tetrameric Avidin Resin	1 ml	V2591
	5 ml	V2592

Description: TetraLink™ Tetrameric Avidin Resin binds and immobilizes biotinylated proteins from complex mixtures of proteins. The high affinity of the avidin-biotin interaction ($K_d = 10^{-15}$M) results in efficient capture of biotinylated proteins and stability of the complex under a wide variety of wash conditions. The resin is useful for the production of affinity resins containing immobilized proteins. TetraLink™ Resin is complementary to the PinPoint™ Xa Protein Purification System, which provides a convenient way to express recombinant proteins with a biotin purification tag.

Features:

- **High-Affinity Binding:** The strength of the biotin-avidin interaction makes binding of biotinylated proteins essentially irreversible.
- **Versatile:** Retains biotin binding ability following exposure to a wide range of pH, low or high ionic strength, 6M guanidine and 1% SDS.
- **Reusable:** When used as an affinity column, can be reused without loss of capacity.
- **Robust:** Supports high flow rates (300cm/hour) and withstands centrifugal forces (1,500 × *g*) in batch applications.

Protocol	Part#
Technical Bulletin	TB230

Storage Conditions: Store at 4°C.

PinPoint™ Xa Protein Purification System

Product	Size	Cat.#
PinPoint™ Xa Protein Purification System	1 system	V2020
Available Separately	**Size**	**Cat.#**
PinPoint™ Xa-1 Vector	10 µg	V2031

Description: The PinPoint™ Xa Protein Purification System is designed for the production and purification of fusion proteins that are biotinylated in vivo. The DNA coding for the protein of interest is cloned into a PinPoint™ Vector downstream of a sequence encoding a peptide that becomes biotinylated in vivo. Biotinylated fusion proteins are produced in *E. coli* and are affinity-purified using the SoftLink™ Soft Release Avidin Resin. This proprietary resin allows elution of the fusion protein under nondenaturing conditions. The PinPoint™ Vectors feature the encoded endoproteinase Factor Xa (pronounced "ten a") proteolytic site that provides a way to separate the purification tag from the native protein, and the vectors carry a convenient multiple cloning region for ease in construction of fusion proteins.

The system contains vectors in all possible sense reading frames, an avidin-conjugated resin, Streptavidin-Alkaline Phosphatase, a purification column and biotin. The PinPoint™ Xa Control Vector contains the chloramphenicol acetyltransferase (CAT) gene and is provided as a means of monitoring protein expression, purification and processing conditions. The system generally yields 1–5mg of protein per liter of culture.

Features:

- **In vivo Biotinylation Tag:** Allows purification of fusion proteins; many proteins produced have been soluble.
- **Easy to Use:** Purification of biotinylated proteins with the SoftLink™ Resin can be performed by column or batch purification.
- **Easy Detection:** Streptavidin Alkaline Phosphatase can be used to detect the biotinylated fusion protein in a pseudo-Western format to monitor purification.
- **Flexible:** PinPoint™ Vectors are supplied for all reading frames.
- **Gentle Release Conditions:** SoftLink™ Resin allows release of the fusion protein under nondenaturing conditions.
- ***tac* Promoter:** Allows tightly regulated expression.

Protocol	Part#
Technical Manual	TM028

Storage Conditions: Store the PinPoint™ Purification Column at room temperature. Store all remaining components at 4°C. The vectors may be stored at –20°C.

PinPoint™ Vector Sequencing Primer

Product	Size	Cat.#
PinPoint™ Vector Sequencing Primer	2 µg	V4211
	2 µg	V4211

Description: The PinPoint™ Vector Sequencing Primer is designed for sequencing inserts cloned into the PinPoint™ Xa Vectors (components of Cat.# V2020). The primer hybridizes upstream of the Factor Xa site at nucleotides 325–343, approximately 40–50 base pairs upstream of the multiple cloning region and can be used to determine if an insert is cloned in-frame with the biotinylation purification tag of the PinPoint™ Xa Vectors. The sequence of the PinPoint™ Vector Sequencing Primer is 5′-d(CGTGACGCGGTGCAGGGCG)-3′. It is supplied dried.

Features:

- **Performance Tested:** The PinPoint™ Vector Sequencing Primer is tested in double-stranded sequencing reactions with circular PinPoint™ Vectors.

Storage Conditions: Store at –20°C.

Transcend™ Non-Radioactive Translation Detection Systems

Product	Size	Cat.#
Transcend™ Colorimetric Non-Radioactive Translation Detection System	30 reactions	L5070
Transcend™ Chemiluminescent Non-Radioactive Translation Detection System	30 reactions	L5080
Available Separately	**Size**	**Cat.#**
Transcend™ tRNA	30 µl	L5061

For Laboratory Use.

Description: The Transcend™ Non-Radioactive Translation Detection Systems allow non-radioactive detection of proteins synthesized in vitro. Using these systems, biotinylated lysine residues are incorporated into nascent proteins during translation, eliminating the need for labeling with [^{35}S]methionine or other radioactive amino acids. This biotinylated lysine is added to the translation reaction as a precharged ε-labeled biotinylated lysine-tRNA complex (Transcend™ tRNA) rather than a free amino acid. After SDS-PAGE and electroblotting, the biotinylated proteins can be visualized by binding either Streptavidin-Alkaline Phosphatase (Streptavidin-AP) or Streptavidin-Horseradish Peroxidase (Streptavidin-HRP), followed either by colorimetric or chemiluminescent detection. Typically, these methods can detect 0.5–5ng of protein within 3–4 hours after gel electrophoresis. This sensitivity is equivalent to that achieved with [^{35}S]methionine incorporation and autoradiographic detection 6–12 hours after gel electrophoresis.

Features:

- **Sensitive:** The biotin tag allows detection of 0.5–5ng of translated protein.
- **Safe:** No radioisotope handling, storage or disposal is required.
- **Fast:** Labeled proteins can be detected 3–4 hours after gel electrophoresis.
- **Flexible:** Results can be visualized by using colorimetric or chemiluminescent detection.

Protocol	Part#
Technical Bulletin	TB182

Storage Conditions: Store Transcend™ tRNA at –70°C. Do not subject the Transcend™ tRNA to more than five freeze-thaw cycles. Store all other components at 4°C.

FluoroTect™ Green$_{Lys}$ in vitro Translation Labeling System

Product	Size	Cat.#
FluoroTect™ Green$_{Lys}$ in vitro Translation Labeling System	40 reactions	L5001

Description: The FluoroTect™ Green$_{Lys}$ in vitro Translation Labeling System allows for the fluorescent labeling and detection of proteins synthesized in vitro. The system is based on a lysine-charged tRNA that is labeled at the ε position of the lysine with the fluorophore BODIPY®-FL. Fluorescent lysine residues will be incorporated into synthesized proteins during in vitro translation reactions, eliminating the need for radioactivity.

Detection of the labeled proteins is accomplished in 2–5 minutes directly "in-gel" by use of a laser-based fluorescent gel scanner. This eliminates any requirements for protein gel manipulation such as fixing/drying or any safety, regulatory and waste disposal issues associated with the use of radioactively labeled amino acids use. The convenience of "in-gel" detection also avoids the time-consuming electroblotting and detection steps of conventional non-isotopic systems.

Features:

- **Fast:** Data can be obtained in minutes, eliminating overnight exposures associated with radioactive-based systems or time-consuming steps utilized by traditional non-isotopic methodologies.
- **Convenient:** Results based on "in-gel" detection. No requirement to transfer, fix, or dry gels.
- **Non-Radioactive:** No safety, regulatory or waste disposal issues associated with radioactivity.
- **Flexible:** The modified charged tRNA can be used with a variety of Promega translation systems including: Rabbit Reticulocyte Lysate, TNT® Coupled Transcription/Translation System, Wheat Germ Extract and *E. coli* S30 Extract.

Protocol	Part#
Technical Bulletin	TB285

Storage Conditions: Store at –70°C.

PhosphoCatch™ Phosphopeptide Enrichment System

Product	Size	Cat.#
PhosphoCatch™ Phosphopeptide Enrichment System	10 pack	V1531
	20 pack	V1532

Description: PhosphoCatch™ Phosphopeptide Enrichment System is uniquely formulated to enhance the phosphopeptide profile by combining titanium oxide and zirconium oxide resins, since these metal oxides differentially enrich mono- and multiphosphorylated peptides. The resin combination along with a unique bind/wash buffer provides high specificity for phosphopeptides and low nonspecific binding. After packing the resins in the provided spin column, the digested sample is added. Following a brief washing step, the sample is eluted and analyzed by mass spectrometry.

Features:

- **Enrichment of Mono- and Multiphosphorylated Peptides:** Accurate representation of multiple phosphorylation sites using one technology.
- **Reduced Nonspecific Binding:** Enhanced data from minimal sample material.
- **Convenient:** Easy-to-use spin-column format.
- **Quick:** The entire procedure can be accomplished in 15 minutes.

Protocol	Part#
Technical Manual	TM324

Storage Conditions: Store PhosphoCatch™ Elution Buffer and PhosphoCatch™ 2X Bind/Wash Buffer at –20°C; store all other components at 4°C.

ProteaseMAX™ Surfactant, Trypsin Enhancer

Product	Size	Cat.#
ProteaseMAX™ Surfactant, Trypsin Enhancer	1 mg	V2071
	5 × 1 mg	V2072

Description: ProteaseMAX™ Surfactant, Trypsin Enhancer, is designed to improve in-gel and in-solution protein digestion. ProteaseMAX™ Surfactant ensures fast and efficient protein digestion with proteases such as Trypsin, Chymotrypsin and Lys-C. For in-gel protein digestion, ProteaseMAX™ Surfactant offers time and labor savings. Digestion step is complete in 1 hour, and the surfactant provides concurrent extraction of peptides from gels, eliminating the need for post-digestion peptide extraction. The surfactant also improves recovery of longer peptides that are retained in the gel under a standard extraction protocol.

For in-solution digestions, ProteaseMAX™ Surfactant solubilizes proteins, including difficult proteins (i.e., membrane proteins), and enhances protein digestion by providing a denaturing environment prior to protease addition.

ProteaseMAX™ Surfactant degrades over the course of a digestion reaction, yielding products that are compatible with downstream methods such as mass spectrometry (MS) and liquid chromatography (LC). No long-term negative effect of the residual surfactant on the ion optics and capillary of mass spectrometers has been observed. ProteaseMAX™ Surfactant can be used with existing in-gel or in-solution digestion protocols.

Features:

- **No Requirement for Peptide Extraction following In-Gel Digestions:** Increase the number of samples processed.
- **One-Hour Digestion:** Save time by avoiding overnight digestions.
- **Improved Peptide Recovery from Gels:** Increase in protein sequence coverage, thus increasing confidence of protein identification.
- **Enhanced Protein Solubilization:** Solubilize complex proteins such as membrane proteins at room temperature, avoiding high temperatures and preventing precipitation.
- **Self-Degradable:** Use directly for mass spectrometry analysis without additional inactivation steps such as heating or acid treatment.

Protocol	Part#
Technical Bulletin	TB373

Storage Conditions: Store lyophilized ProteaseMAX™ Surfactant at –20°C.

Immobilized Trypsin

Product	Size	Cat.#
Immobilized Trypsin	2 ml	V9012
	4ml (2 × 2 ml)	V9013

Description: Immobilized Trypsin provides a fast and convenient method for digesting a range of concentrations of purified protein or complex protein mixtures. Digested peptides are easily separated from the Immobilized Trypsin as they flow through the spin column into the collection tube. Immobilized Trypsin is easily removed from the peptide solution because the trypsin does not pass though the column frit. Trypsin is a proteolytic enzyme, which cleaves at the carboxyl side of positively charged Lysine (Lys) and Arginine (Arg). When these amino acids are followed by the nonpolar Proline (Pro), the digestion of the site is not efficient. When Lys and Arg are followed by acids [Aspartic Acid (Asp) and Glutamic Acid (Glu)] the digestion is also not as efficient.

Features:

- **Fast:** Digestions can be accomplished in as little as 30 minutes.
- **Scalable:** Easily adjustable protocol to accommodate various protein concentrations.
- **Easy-to-Use:** No shaking or water baths necessary.

Protocol	Part#
Technical Manual	TM077

Storage Conditions: Store at 4°C.

Chymotrypsin, Sequencing Grade

Product	Size	Cat.#
Chymotrypsin, Sequencing Grade	25 µg	V1061
	100µg (4 × 25 µg)	V1062

Description: Chymotrypsin is a highly-purified serine endopeptidase derived from bovine pancreas that preferentially hydrolyzes at the carboxyl side of aromatic amino acids: Tyr, Phe and Trp. Cleavage may also be observed, but at a lower rate, at Leu and Met. Chymotrypsin activity is optimal in the pH range of 7.0–9.0. This sequencing grade enzyme can be used alone or in combination with other proteases to produce protein digests for peptide mapping applications or protein identification by peptide mass fingerprinting or MS/MS spectral matching. It is suitable for digestion reactions in-solution or in-gel.

Protocol	Part#
Promega Product Information	9PIV106

Storage Conditions: Store at 4°C.

Trypsin Gold, Mass Spectrometry Grade

Product	Size	Cat.#
Trypsin Gold, Mass Spectrometry Grade	100 µg	V5280

Description: Trypsin is a serine protease that specifically cleaves at the carboxylic side of lysine and arginine residues. The stringent specificity of trypsin is essential for protein identification. Native trypsin is subject to autolysis, generating pseudotrypsin, which exhibits a broadened specificity including a chymotrypsin-like activity. Such autolysis products, present in a trypsin preparation, would result in additional peptide fragments that could interfere with database analysis of the mass of fragments detected by mass spectrometry. Trypsin Gold, Mass Spectrometry Grade, has been manufactured to provide maximum specificity. Lysine residues in the porcine trypsin have been modified by reductive methylation, yielding a highly active and stable molecule that is extremely resistant to autolytic digestion. The specificity of the purified trypsin is further improved by TPCK treatment, which inactivates chymotrypsin. The treated trypsin is then purified by affinity chromatography and lyophilized to yield Trypsin Gold, Mass Spectrometry Grade. Trypsin is often used for in-gel digestion. The digestion products are purified and concentrated, then analyzed by mass spectrometry to determine their molecular weights. Database searches can then be performed, using the mass of the peptides to identify the protein(s) resolved on the gel. Each lot of quality-tested Trypsin Gold, Mass Spectrometry Grade, is qualified for use with in-gel digestion and mass spectrometric analysis.

Features:

- **Pure:** TPCK treatment followed by affinity purification.
- **Application Qualified:** Each lot is qualified by mass spectrometry.
- **Convenient:** Provided in one vial.
- **Good Value:** Stability ensured up to five freeze-thaw cycles, thus minimizing leftover reagents.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Technical Bulletin	TB309

Storage Conditions: Store the lyophilized powder at –20°C. Reconstitute powder in 50mM acetic acid and store at –20°C. For long-term storage, freeze reconstituted trypsin at –70°C. Limit the number of freeze-thaw cycles to five.

Sequencing Grade Modified Trypsin

Product	Size	Cat.#
Sequencing Grade Modified Trypsin	100 µg	V5111
For Laboratory Use.		

Description: Trypsin specifically hydrolyzes peptide bonds at the carboxylic sides of lysine and arginine residues. Unmodified trypsin is subject to autolysis, generating fragments that can interfere with protein sequencing, HPLC or mass spectrometry analysis of the peptides. In addition, autolysis can result in the generation of pseudotrypsin, which has been shown to exhibit an additional chymotrypsin-like specificity. Promega Trypsin has been modified by reductive methylation, rendering it extremely resistant to autolytic digestion. In functional stability tests, modified trypsin retains at least two times as much activity as unmodified trypsin after a 3-hour incubation at 37°C.

The sequencing grade of modified trypsin has been further improved by TPCK treatment followed by affinity purification yielding a highly active and stable molecule. Sequencing Grade Modified Trypsin is provided as a lyophilized powder in convenient 20µg aliquots with a stability-optimized resuspension buffer. A protease:protein ratio of 1:100 to 1:20 (w/w) is recommended.

Recommended Reaction Buffer: 50mM NH_4HCO_3 (pH 7.8).

Online Protocols Referring to Promega Sequencing Grade Modified Trypsin

www.cbs.umn.edu/msp/protocols/insolution.shtml
www.umdnj.edu/proweb/methods/Sypro-Trypsin.pdf
www.biol.yorku.ca/cm/proteomics/in-gel.rtf
www.biochem.purdue.edu/psal/protocol.html
proteomics.unc.edu/docs/Trypsin_Digestion_Protocol.pdf
www.its.caltech.edu/~ppmal/sample_prep/digest.html
www.abrf.org/ResearchGroups/ProteinIdentification/EPosters/pirgprotocol.html
biology.berkeley.edu/crl/mass_spec/in_gel_digestion.pdf
www.healthsystem.virginia.edu/internet/biomolec/ingeldigest.cfm
www.utexas.edu/pharmacy/divisions/pharmtox/core/protocol2.html

Features:

- **Pure:** TPCK treatment followed by affinity purification.
- **Easy to Use:** Resuspension buffer provided.
- **Convenient:** Five tubes of lyophilized product are provided.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Promega Product Information	9PIV511

Storage Conditions: Store lyophilized at –20°C.

Sequencing Grade Modified Trypsin, Frozen

Product	Size	Cat.#
Sequencing Grade Modified Trypsin, Frozen	100 µg	V5113

For Laboratory Use.

Description: Trypsin specifically hydrolyzes peptide bonds at the carboxylic sides of lysine and arginine residues. Unmodified trypsin is subject to autolysis, generating fragments that can interfere with protein sequencing, HPLC or mass spectrometry analysis of the peptides. In addition, autolysis can result in the generation of pseudotrypsin, which has been shown to exhibit an additional chymotrypsin-like specificity. Promega Trypsin has been modified by reductive methylation, rendering it extremely resistant to autolytic digestion. In functional stability tests, modified trypsin retains at least two times as much activity as unmodified trypsin after a 3-hour incubation at 37°C.

The sequencing grade of modified trypsin has been further improved by TPCK treatment followed by affinity purification yielding a highly active and stable molecule. Sequencing grade modified trypsin is provided as a frozen liquid in convenient 20µg aliquots with a stability-optimized dilution buffer. A protease:protein ratio of 1:100 to 1:20 (w/w) is recommended for protein sequencing.

Recommended Reaction Buffer: 50mM NH_4HCO_3 (pH 7.8).

Protocol	Part#
Promega Product Information	9PIV5113

Storage Conditions: Store at –70°C.

Endoproteinase Lys-C, Sequencing Grade

Product	Size	Cat.#
Endoproteinase Lys-C, Sequencing Grade	5 µg	V1071

Description: Endoproteinase Lys-C is a sequencing grade serine protease isolated from Lysobacter enzymogenes as a highly purified protease that hydrolyzes specifically at the carboxyl side of Lys. Lys-C activity is optimal in the pH range of 7.0–9.0. This sequencing grade enzyme can be used alone or in combination with other proteases to produce protein digests for peptide mapping applications or protein identification by peptide mass fingerprinting or MS/MS spectral matching. It is suitable for digestion reactions in-solution or in-gel.

Protocol	Part#
Promega Product Information	9PIV107

Storage Conditions: Store at 4°C.

Asp-N, Sequencing Grade

Product	Size	Cat.#	
Asp-N, Sequencing Grade	2 µg	V1621	150

Description: Asp-N, Sequencing Grade, is an endoproteinase that hydrolyzes peptide bonds on the N-terminal side of aspartic and cysteic acid residues: Asp and Cys. Asp-N activity is optimal in the pH range of 4.0–9.0. This sequencing grade enzyme can be used alone or in combination with other proteases to produce protein digests for peptide mapping applications or protein identification by peptide mass fingerprinting or MS/MS spectral matching. It is suitable for digestion reactions in solution or in gel.

Protocol	Part#
Promega Product Information	9PIV162

Storage Conditions: Store at 4°C.

Proteinase K

Product	Size	Cat.#
Proteinase K	100 mg	V3021

For Laboratory Use.

Description: Proteinase K, produced by the fungus *Tritirachium album* Limber, is a serine protease that exhibits broad cleavage activity. It cleaves peptide bonds adjacent to the carboxylic group of aliphatic and aromatic amino acids and is useful for general digestion of protein in biological samples. It has been purified to remove RNase and DNase activities. The stability of Proteinase K in urea and SDS and its ability to digest native proteins make it useful for a variety of applications including preparation of chromosomal DNA for pulsed-field gel electrophoresis, protein fingerprinting and removal of nucleases from preparations of DNA and RNA. A typical working concentration for Proteinase K is 50–100µg/ml.

Form: Lyophilized powder.

Recommended Reaction Buffer: 50mM Tris-HCl (pH 8.0), 10mM $CaCl_2$.

Features:

- **Stable:** Active over a pH range of 4.3–12.0, in 0.5% SDS or 1% Triton® X-100 and retains >80% of its activity at temperatures up to 60°C.

Protocol	Part#
Promega Product Information	9PIV302

Storage Conditions: Store lyophilized powder desiccated at –20°C.

Factor Xa Protease

Product	Size	Cat.#
Factor Xa Protease	50 µg	V5581

Description: Factor Xa Protease is purified from bovine plasma and activated by treatment with the activating enzyme from Russell's viper venom. Factor Xa Protease preferentially cleaves after the arginine residue in the amino acid sequence Ile-Glu-Gly-Arg.

Recommended Reaction Buffer: 20mM Tris-HCl (pH 7.4), 0.1M NaCl.

Storage Conditions: Store in aliquots at –20°C.

HaloLink™ Protein Array Systems

Product	Size	Cat.#
HaloLink™ Array (TNT® T7 Quick) Two Slide System	two 50 -well arrays	G6140
HaloLink™ Array (TNT® SP6 Wheat Germ) Two Slide System	two 50 -well arrays	G6180
HaloLink™ Array Six Slide System	6 slides	G6190
HaloTag® Standard Protein	30 µg	G4491

Available Separately	Size	Conc.	Cat.#
HaloTag® TMR Ligand	15 µl	5 mM	G8252

Description: Protein arrays enable parallel analysis of multiple protein:protein, protein:drug or protein:nucleic acid interactions. The HaloLink™ Protein Array Systems provide a new way to create homebrew (on-demand) protein arrays by combining innovative HaloTag® technology, surface engineering and cell-free protein expression systems.

The HaloTag® protein is a mutated hydrolase that forms a covalent bond with HaloTag® ligands. Under physiological conditions binding is rapid and highly specific, yielding a complex that is stable even under stringent conditions. Using the HaloLink™ Protein Array Systems, HaloTag® fusion proteins are expressed in a cell-free expression system and then covalently captured on hydrogel-coated glass slides derivatized with HaloTag® Ligands. The fusion proteins are captured directly from the expression reaction mixture without prior purification. Using this approach, multiple fusion proteins can be rapidly synthesized and immobilized in parallel on the slide surface, and an entire experiment including protein expression, custom array formation and protein interaction analysis can be completed in less than eight hours.

The HaloLink™ Array Two Slide Systems (Cat.# G6140 and G6180) contain HaloLink™ Slides, HaloLink™ Array Gaskets for creating 50-well arrays, a HaloTag® Standard Protein, Anti-HaloTag® Antibody and a cell-free expression system [either the TNT® T7 Quick Coupled Transcription/Translation System (Cat.# G6140) or the TNT® SP6 High-Yield Wheat Germ Protein Expression System (Cat.# G6180)].

The HaloLink™ Array Six Slide System contains HaloLink™ Slides, HaloLink™ Array Gaskets and Anti-HaloTag® Antibody. To use the Six Slide System you will need to provide your own protein expression system or order the TNT® T7 Quick Coupled Transcription/Translation System (Cat.# L1170 or L1171) or TNT® SP6 High-Yield Wheat Germ Protein Expression System (Cat.# L3260 or L3261). The HaloTag® Standard Protein (Cat.# G4491) is not included with the Six Slide System but can be ordered separately.

The HaloTag® technology offers a quick and convenient way to test protein expression of HaloTag® fusion proteins by labeling with fluorescent HaloTag® TMR Ligand followed by SDS-PAGE analysis. Guidelines can be found in the HaloLink™ Protein Array Technical Manual #TM310.

Features:

- **Fast Protein Production:** Cell-free expression systems allow quick, single-tube, coupled transcription/translation for the production of the proteins of interest to be used in the protein array experiment.
- **Irreversible Binding of the Captured Protein:** Unlike other affinity tags, which tend to dissociate from the surface, HaloTag® fusion proteins are covalently bound to the HaloLink™ Slide.
- **No Protein Purification Step:** The protein of interest is immobilized directly from the cell-free expression reaction.
- **Reduced Nonspecific Binding:** HaloLink™ Slides have a unique hydrogel coating that is designed to prevent nonspecific binding while preserving the functionality of specifically captured proteins.
- **Extensive Washing Allowed:** Covalent binding of HaloTag® fusion proteins to the HaloLink™ Slide allows extensive, stringent washing that results in reduced background and a lower incidence of false positives.
- **No Need for a Robotic Arrayer:** The unique 50-well configuration allows multiple interactions to be studied in parallel without the need for a complex robotic arrayer.

Protocol	Part#
Technical Manual	TM310

Storage Conditions: Store the TNT® lysates at –70°C. Store the HaloTag® Standard Protein and the Anti-HaloTag® Antibody at –20°C. The HaloLink™ Protein Array Slides should be stored at –20°C and opened just before use. After opening, unused slides should be stored at –20°C and used within one month. Store the HaloLink™ Array Gaskets at room temperature.

- **Do not store the TNT® lysate at any temperature other than –70°C. Storage at other temperatures (e.g., –20°C) for even a short time will dramatically reduce activity.**
- **Do not freeze-thaw the TNT® lysate more than two times.**
- **Do not store the TNT® lysate in the presence of dry ice. Prolonged exposure to dry ice can cause significant loss of activity.**

HaloLink™ Magnetic Beads

Product	Size	Cat.#
HaloLink™ Magnetic Beads	40 reactions	G9311

Description: HaloLink™ Magnetic Beads provide a rapid and reliable method to covalently capture and immobilize HaloTag® fusion proteins to a paramagnetic particle. Immobilization through the HaloTag® Ligand provides for consistent, surface-directed orientation of the fusion protein, thus providing more reliable results in applications such as protein:protein interaction studies with the fusion protein. The result is a paramagnetic bead with very high binding capacity for HaloTag® fusion proteins and very low nonspecific protein binding. The paramagnetic feature allows for streamlined assay development and facilitates applications with automated assays developed on robotic platforms.

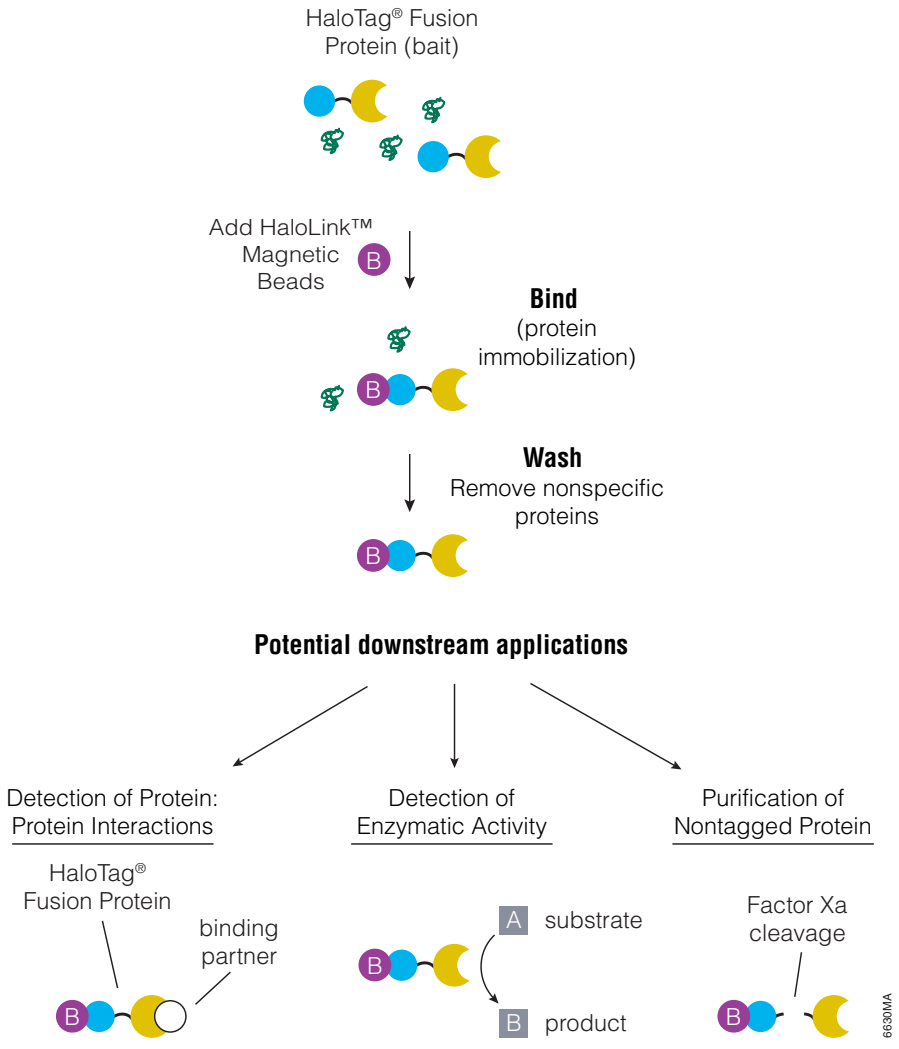
A variety of vectors for the expression of HaloTag® fusion proteins in bacterial, mammalian or cell-free systems are available.

Features:

- **Magnetic-Based:** Easy and quick immobilization of HaloTag® fusion proteins with minimal loss of sample material.
- **Covalent Bond:** Allows stringent washing of immobilized HaloTag® fusion protein, removing nonspecific proteins and enhancing overall data.
- **Multiple Applications:** Immobilized HaloTag® fusion proteins can be used for protein interaction analysis, enzyme assays and purification of fusion proteins containing a protease cleavage site.

Protocol	Part#
Technical Manual	TM291

Storage Conditions: Store at 4°C.



Overview of the HaloLink™ Magnetic Beads immobilization protocol.

HaloCHIP™ System

Product	Size	Cat.#	
HaloCHIP™ System	20 reactions	G9410	
Available Separately	**Size**	**Cat.#**	
pFC14A HaloTag® CMV Flexi® Vector	20 µg	G9651	193
pFC14K HaloTag® CMV Flexi® Vector	20 µg	G9661	193
pFC15A HaloTag® CMV*d1* Flexi® Vector	20 µg	G1611	193
pFC15K HaloTag® CMV*d1* Flexi® Vector	20 µg	G1601	193
pFC16A HaloTag® CMV*d2* Flexi® Vector	20 µg	G1591	193
pFC16K HaloTag® CMV*d2* Flexi® Vector	20 µg	G1571	193
pFC17A HaloTag® CMV*d3* Flexi® Vector	20 µg	G1551	193
pFC17K HaloTag® CMV*d3* Flexi® Vector	20 µg	G1321	193
pFN21A HaloTag® CMV Flexi® Vector	20 µg	G2821	193
pFN21K HaloTag® CMV Flexi® Vector	20 µg	G2831	193
pFN22A HaloTag® CMV*d1* Flexi® Vector	20 µg	G2841	193
pFN22K HaloTag® CMV*d1* Flexi® Vector	20 µg	G2851	193
pFN23A HaloTag® CMV*d2* Flexi® Vector	20 µg	G2861	193
pFN23K HaloTag® CMV*d2* Flexi® Vector	20 µg	G2871	193
pFN24A HaloTag® CMV*d3* Flexi® Vector	20 µg	G2881	193
pFN24K HaloTag® CMV*d3* Flexi® Vector	20 µg	G2981	193
HaloTag® Flexi® Vectors—CMV Deletion Series Sample Pack	9 × 2 µg	G3780	193

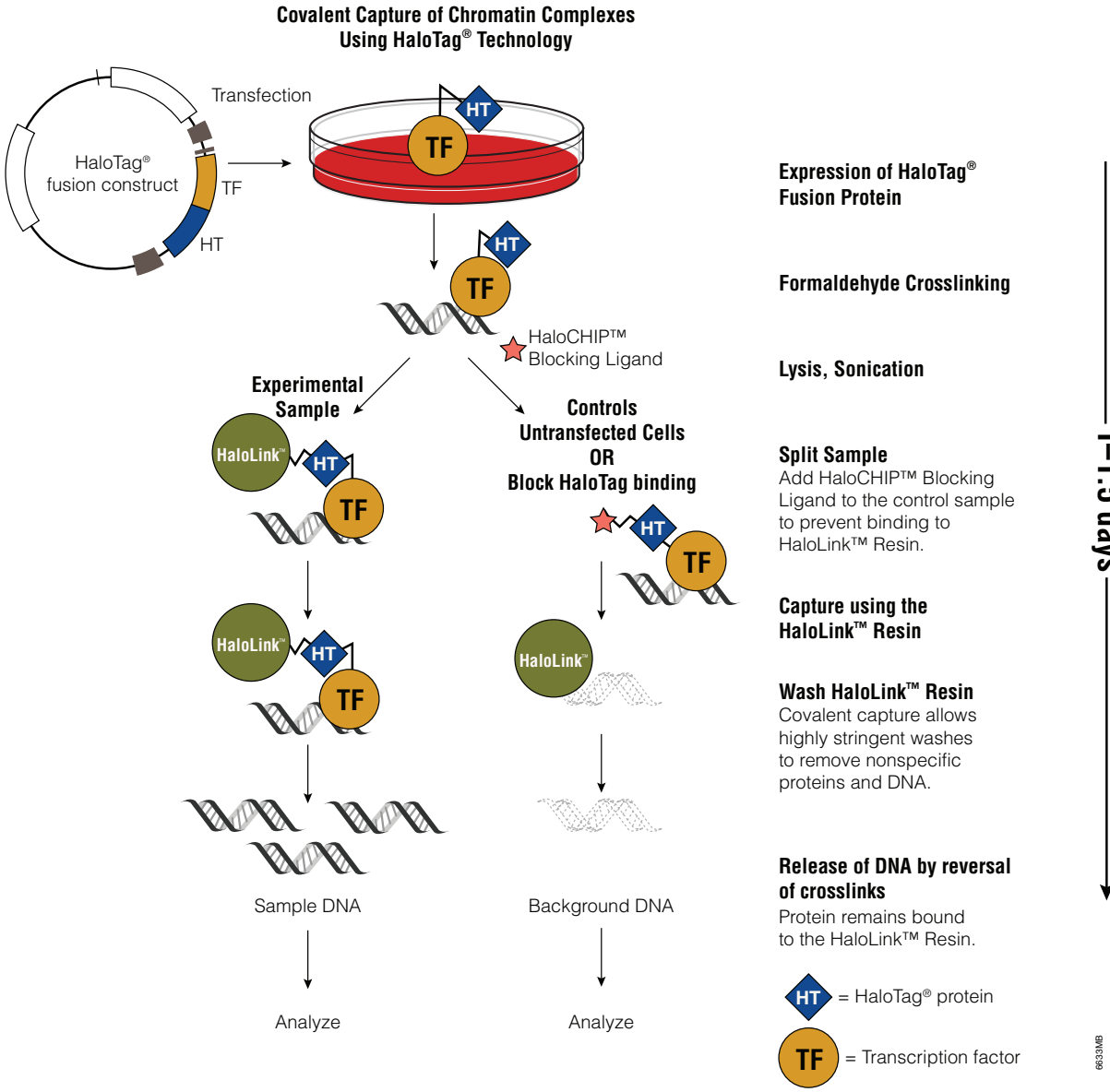
Description: The HaloCHIP™ System is a novel method designed for the covalent capture of intracellular protein:DNA complexes without the use of antibodies and offers an efficient and robust alternative to the standard chromatin immunoprecipitation (ChIP) method. Proteins of interest are expressed in cells as HaloTag® fusion proteins, crosslinked to DNA with formaldehyde and then captured on HaloLink™ Resin, which forms a highly specific, covalent interaction with the HaloTag® portion of the fusion protein. Stringent washing removes nonspecific proteins and DNA, and heating reverses the crosslinks between the DNA and the fusion protein and releases the captured DNA fragment, which subsequently can be purified.

Features:

- **No Requirement for Antibody:** No need to make your own or purchase expensive, qualified antibodies.
- **Obtain Results Faster:** Obtain data in 24–48 hours with fewer steps to minimize potential experimental errors.
- **Improved Signal-to-Noise Ratios:** Enables detection of small changes in protein binding patterns using a minimal number of cells.

Protocol	Part#
Technical Manual	TM075

Storage Conditions: The TE Buffer (pH 8.0), Reversal Buffer and Nuclease-Free Water may be stored at room temperature. Store the HaloLink™ Resin, Mammalian Lysis Buffer and High Salt Wash Buffer at 4°C. Store the HaloCHIP™ Blocking Ligand at –20°C.



Schematic diagram of the HaloCHIP™ System

HaloLink™ Resin

Product	Size	Cat.#
HaloLink™ Resin	2ml (0.5ml settled resin)	G1911
	5ml (1.25ml settled resin)	G1912
	10ml (settled resin)	G1914

Available Separately	Size	Conc.	Cat.#
HaloTag® Protein Purification System	1 each		G6280
HaloTag® TMR Ligand	15 µl	5 mM	G8252

Description: The HaloLink™ Resin provides a method for covalent and oriented attachment of HaloTag® fusion proteins onto a solid surface. The resin consists of a HaloTag® ligand bound to Sepharose® beads that specifically and rapidly binds HaloTag® fusion proteins. HaloLink™ Resin has high binding capacity. Due to covalent linkage, HaloTag® fusion proteins cannot be eluted from the resin, allowing extensive washing to remove nonspecifically bound protein without the danger of eluting HaloTag® fusion proteins. The binding rate is very rapid and equivalent to biotin-streptavidin.

The HaloLink™ Resin can be used in a variety of applications including: detection and analysis of protein:protein interactions (in vivo and in vitro), detection of enzymatic activity of immobilized HaloTag® fusions and one-step purification of fusion protein in conjunction with proteolytic cleavage. A variety of vectors for the expression of HaloTag® fusion proteins in bacterial, mammalian or cell-free systems are available.

The HaloTag® technology offers a quick and convenient way to test protein expression of HaloTag® fusion proteins as well as monitor the efficiency of immobilization to HaloLink™ Resin by labeling with fluorescent HaloTag® TMR Ligand followed by SDS-PAGE analysis. Guidelines can be found in the HaloLink™ Resin Technical Manual #TM250.

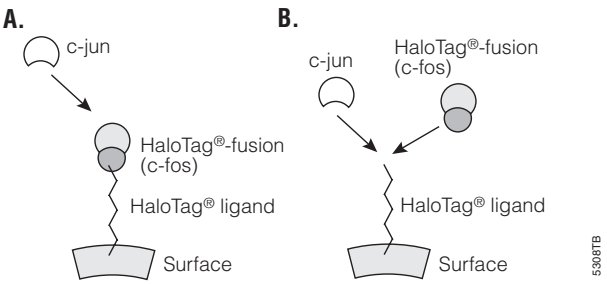
Features:

- **Covalent Attachment:** Enables stringent washing, minimizing nonspecific background without dissociation of bound HaloTag® fusion proteins.
- **Fast Binding Kinetics:** Enhances the detection of protein:protein interactions and enables binding of proteins at low concentrations.
- **Oriented Immobilization:** Allows maximal enzyme activity of bound protein.
- **High Binding Capacity:** One milliliter of settled resin binds >7mg of HaloTag® fusion proteins.

Protocol	Part#
Technical Manual	TM250

Note: For protein expression in mammalian cells when using a protease inhibitor cocktail, we recommend BaculoGold® Protease Inhibitor Cocktail (BD Biosciences, Cat# 554779) Some protease inhibitor cocktails, especially those containing 4-(2-aminoethyl) benzenesulfonyl fluoride hydrochloride (AEBSF), will reduce binding efficiency

Storage Conditions: Store at 4°C.



Detection of protein:protein interactions using the HaloLink™ Resin. **Panel A.** HaloTag® fusion protein (bait, HaloTag® c-fos) is expressed in TNT® T7 Quick Coupled Transcription/Translation System and immobilized to the HaloLink™ Resin. The partner (prey) c-jun is expressed in TNT® reactions and mixed with the immobilized HaloTag® c-fos and allowed to bind.**Panel B.** Both interacting partners, bait and prey HaloTag® fusions are expressed in individual TNT® reactions, mixed and allowed to bind; then the HaloLink™ Resin is added, and the complex is captured.

HaloTag® Mammalian Pull-Down Systems

Product	Size	Cat.#
HaloTag® Mammalian Pull-Down and Labeling System	24 reactions	G6500
HaloTag® Mammalian Pull-Down System	24 reactions	G6504
Available Separately	**Size**	**Cat.#**
HaloTag® Control Vector	20 µg	G6591
Protease Inhibitor Cocktail, 50X	1 ml	G6521
Mammalian Lysis Buffer	40 ml	G9381

Description: The **HaloTag® Mammalian Pull-Down and Labeling System** (Cat.# G6500 and G6504) is designed to capture and purify intracellular binary and higher order protein complexes, including transient or weakly interacting partners. This system includes the HaloTag® TMRDirect™ Ligand, which allows correlative cellular localization and real-time imaging studies with the same genetic construct as a means to further understand overall protein function. The **HaloTag® Mammalian Pull-Down System** does not include the HaloTag® TMRDirect™ Ligand.

The **HaloTag® Control Vector** provides protein expression of the HaloTag® protein in mammalian cells, *E. coli* or in vitro expression systems dependent on human cytomegalovirus (CMV) intermediate early enhancer, T7 or SP6 RNA polymerase promoters. It can be used as a control for any HaloTag® experimental system and can be used for both stable and transient HaloTag® expression in mammalian cells; for stable expression, co-transfection with a vector containing a selectable marker is required.

The **Protease Inhibitor Cocktail, 50X**, is a mixture of six different protease inhibitors with different target protease specificities. This product is provided in a freeze-dried format and can be reconstituted using either 100% ethanol or DMSO.

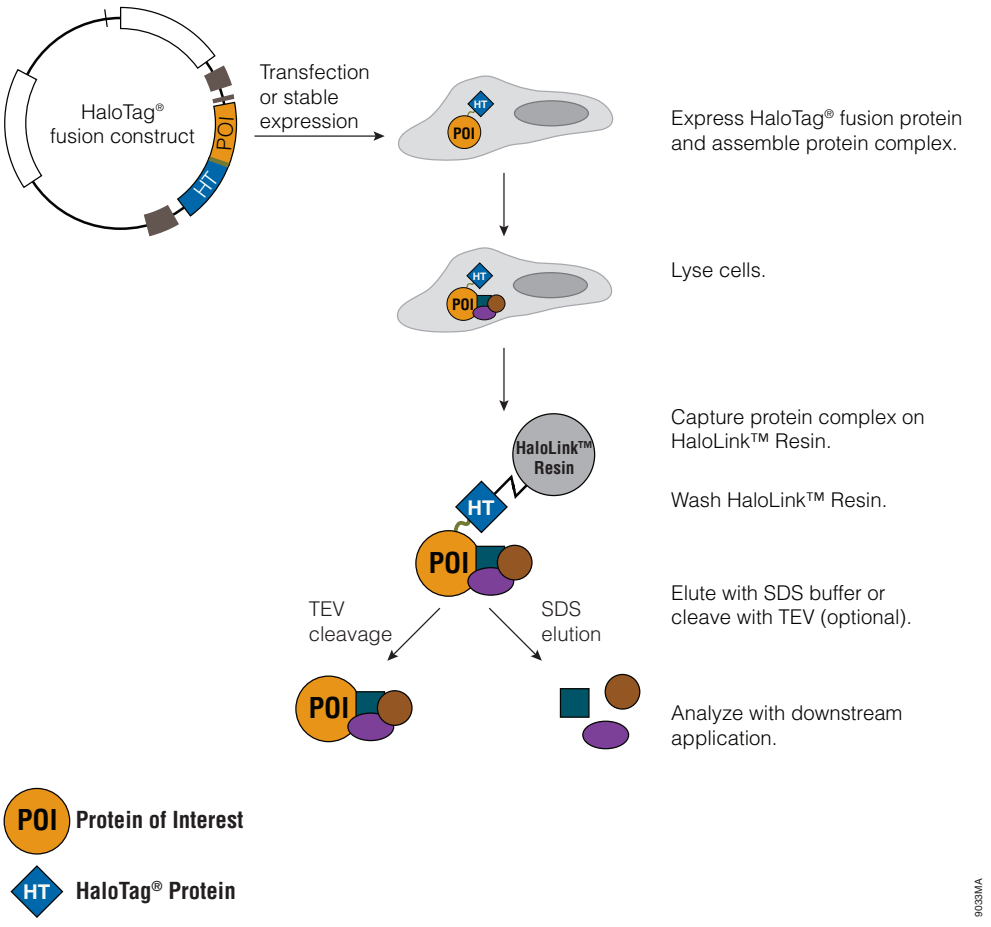
The **Mammalian Lysis Buffer** is designed for use with HaloTag® Mammalian-based expression systems such as the HaloTag® Mammalian Pull-Down and Labeling Systems (referenced here) as well as the HaloCHIP™ System (Cat.# G9410). Formulation consists of 50mM Tris-HCl, 150mM NaCl, 1% Triton® X-100 and 0.1% sodium deoxycholate (pH 7.5).

Features:

- **Rapid, Efficient and Covalent Capture of Binary and Higher Order Complexes Directly from Lysates:** Improved capture of protein partners, including transient interactions.
- **High Purity and Low Background:** Improved accuracy in identification of proteins; covalent attachment allows bait protein to remain behind if desired.
- **Ability to Fluorescently Label the Same Genetic Fusion:** Correlate complex capture with cellular localization.
- **Compatibility with All Downstream Methods of Analysis:** Freedom to identify complexes in variety of applications including mass spectrometry.

Protocol	Part#
Technical Manual	TM342

Storage Conditions: Store the 10X TBS Buffer and SDS Elution Buffer at room temperature. Store the HaloLink™ Resin and Mammalian Lysis Buffer at 4°C. Store the Protease Inhibitor Cocktail and HaloTag® TMRDirect™ Ligand at –20°C.



Representation of the HaloTag® mammalian pull-down assay using HaloTag® fusion protein as bait.

MagneGST™ Pull-Down System (GST Pull-Downs)

Product	Size	Cat.#
MagneGST™ Pull-Down System	80 reactions	V8870

This product requires the use of a magnetic stand.

Description: The MagneGST™ Pull-Down System is designed for detection of protein interactions between GST-fusion proteins expressed in bacterial lysates and prey proteins expressed in the TNT® Systems. Prey protein synthesized in the TNT® Quick Coupled Transcription/Translation Reaction is captured using bait protein (GST-fusion protein) immobilized on MagneGST™ Particles. Nonspecifically bound proteins are then washed away, and the prey protein is analyzed. Prey proteins can be detected by incorporating radioactively labeled methionine in the TNT® Quick reaction, followed by SDS-PAGE and autoradiography or by incorporating the supplied non-radioactive methionine in the TNT® reaction and detecting by Western blotting with protein-specific antibodies.

Protocol	Part#
Technical Manual	TM249

Storage Conditions: Store the TNT® T7 Quick Master Mix and Methionine at –70°C. Store the RQ1 RNase-Free DNase at –20°C. Store the Nuclease-Free Water, MagneGST™ Glutathione Particles, MagneGST™ Binding/Wash Buffer and Cell Lysis Reagent at 4°C.

CheckMate™/Flexi® Vector Mammalian Two-Hybrid System

Product	Size	Cat.#
CheckMate™/Flexi® Vector Mammalian Two-Hybrid System	1 each	C9360
Available Separately	**Size**	**Cat.#**
pFN10A (ACT) Flexi® Vector	20 µg	C9331
pFN11A (BIND) Flexi® Vector	20 µg	C9341
pGL4.31[*luc2P/GAL4*UAS/Hygro] Vector	20 µg	C9351
CheckMate™ Positive Control Vectors	1 set	C9370
CheckMate™ Negative Control Vectors	1 set	C9380
Flexi® System, Entry/Transfer	5 entry and 20 transfer reactions	C8640
JM109 Competent Cells, >10^7cfu/µg	5 × 200 µl	L1001

Description: The CheckMate™/Flexi® Vector Mammalian Two-Hybrid System provides a means to confirm, validate and study suspected interactions between two proteins or domains and can also be used to generate stable cell lines for cell-based assays. Developed primarily for mammalian proteins of interest, the system can allow protein expression and post-translational modifications in an environment mimicking the native cell milieu. It is patterned on the yeast two-hybrid system with one protein of interest ("X") fused to a DNA-binding domain and the other protein ("Y") fused to a transcriptional activation domain.

The system uses three plasmids that are co-transfected into mammalian cells, each plasmid has unique features. The pFN10A (ACT) Flexi® Vector contains a herpes simplex virus VP16 transcriptional activation domain upstream of the cloning site, and the pFN11A (BIND) Flexi® Vector contains the yeast GAL4DNA-binding domain upstream of the cloning site. The pFN11A (BIND) Flexi® Vector also expresses the *Renilla reniformis* luciferase under the control of the SV40 promoter, allowing normalization for differences in transfection efficiency. The third vector, pGL4.31[*luc2P/GAL4*UAS/Hygro] Vector, contains five GAL4 binding sites upstream of a minimal TATA box, which is upstream of a firefly luciferase gene that acts as a reporter for interactions between proteins X and Y.

This system differs from the original CheckMate™ Mammalian Two-Hybrid System in that the vectors are compatible with the Flexi® Vector System, which allows directional cloning and rapid, efficient and high-fidelity transfer of protein coding regions between a variety of Flexi® Vectors.

Features:

- **Mammalian-Based System:** Interactions can be studied in the cell line of choice. Proteins are more likely to be in their native conformation. Post-translational modifications, such as glycosylation, phosphorylation and acylation, are better maintained.
- **Versatile:** Vectors are based on the Flexi® Cloning technology, enabling convenient transfer of protein-coding regions for additional functional proteomics applications.
- **Convenient:** The Dual-Luciferase® Reporter Assay System is used for detection.

Protocol	Part#
Technical Manual	TM283

Storage Conditions: Store at –20°C.

CheckMate™ Mammalian Two-Hybrid System

Product	Size	Cat.#
CheckMate™ Mammalian Two-Hybrid System	1 system	E2440

Description: Two-hybrid systems are extremely powerful methods for detecting protein:protein interactions in vivo. The basis of two-hybrid systems is the modular domains found in some transcription factors: a DNA-binding domain, which binds to a specific DNA sequence, and a transcriptional activation domain, which interacts with the basal transcriptional machinery. A transcriptional activation domain in association with a DNA-binding domain will promote the assembly of RNA polymerase II complexes at the TATA box and increase transcription. In the CheckMate™ Mammalian Two-Hybrid System the DNA-binding domain and the transcriptional activation domain, produced by separate plasmids, are closely associated when one protein ("X") fused to a DNA-binding domain interacts with a second protein ("Y") fused to a transcriptional activation domain. In this system, interaction between proteins X and Y results in transcription of a reporter gene.

Features:

- **Mammalian System:** Interactions can be studied in the cell line of choice. Proteins are more likely to be in their native conformation. Post-translational modifications, such as glycosylation, phosphorylation and acylation, are better maintained.
- **Convenient Quantitation:** The Dual-Luciferase® Reporter Assay System is used for detection.
- **Internal Control:** *Renilla* luciferase normalizes transfection efficiency.
- **Fast Transient Assay:** Results obtained two days after transfection, as compared to 3–4 days with the yeast system.
- **Stable Transfectants:** The pACT Vector contains the neomycin phosphotransferase gene, which allows for selection of stable transfectants.

Protocol	Part#
Technical Manual	TM049

Storage Conditions: Store at –20°C.

Restriction/Modifying Enzymes and RNase Inhibitors

Restriction Enzymes	**242**
Restriction Enzyme Accessory Products	**251**
DNA Polymerases	**252**
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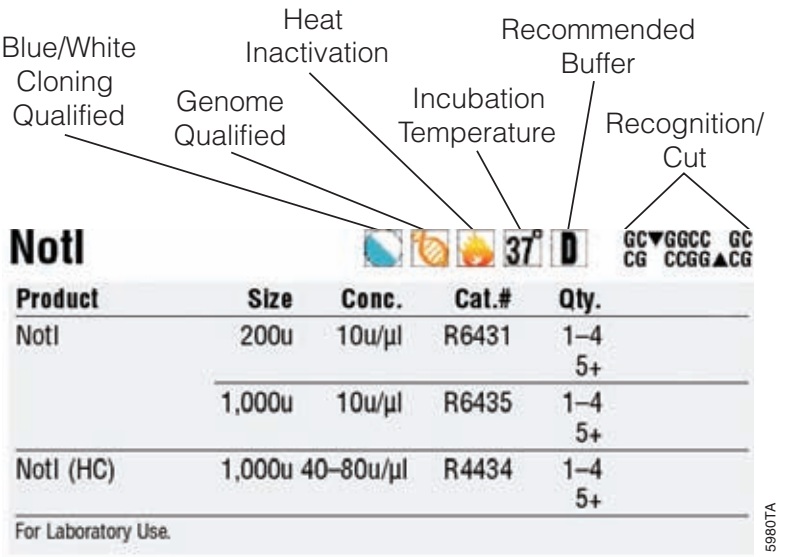
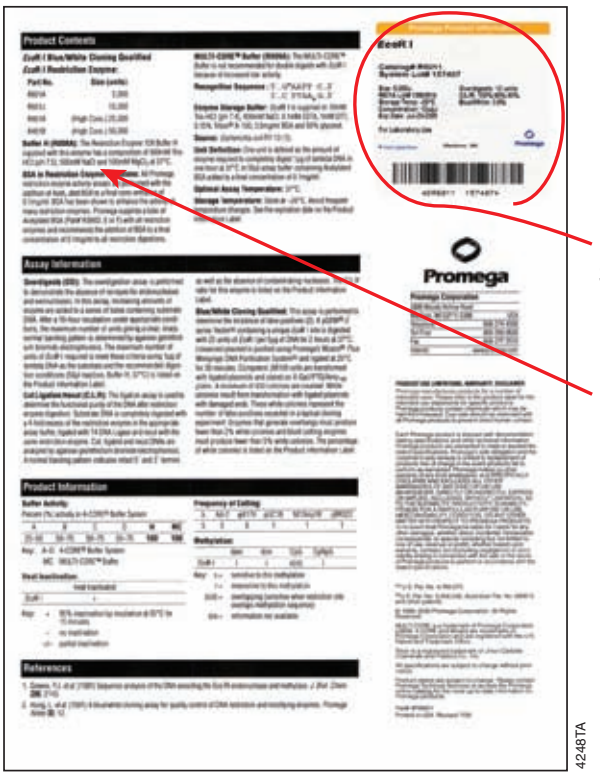
Additional Enzymes and Proteins	**260**
RNasin® Ribonuclease Inhibitors	**261**

All the Information You Need—At a Glance

On the following pages, restriction enzyme information is organized using icons to help you quickly and easily identify the features of each enzyme. See the diagram to the right to identify the meaning of the icons used.



Product Usage, Quality Control and Lot-Specific Information



Each enzyme comes in recyclable packaging that holds the enzyme, buffers (if applicable) and a lot-specific Product Information Sheet. The Product Information Sheet contains details of the quality control assays performed, product storage and usage information, protocols and references. Lot-specific information is printed on a removable sticker that can be pasted into a notebook or log book, simplifying your record-keeping.

AatII

37 J

Product	Size	Conc.	Cat.#
AatII	50 u	3–5 u/µl	R6541
	250 u	3–5 u/µl	R6545

For Laboratory Use.

Description: G ACGT▼C
C▲TGCA G
Storage Conditions: Store at –20°C.

AccI

37 G

Product	Size	Conc.	Cat.#
AccI	100 u	3–10 u/µl	R6411
	500 u	3–10 u/µl	R6415

For Laboratory Use.

Description: GT▼(A/C)(T/G) AC
CA (T/G)(A/C)▲TG
Storage Conditions: Store at –20°C.

AccIII

65 F

Product	Size	Conc.	Cat.#
AccIII	200 u	10 u/µl	R6581

For Laboratory Use.

Description: T▼CCGG A
A GGCC▲T
Storage Conditions: Store at –20°C. **Do not freeze.**

Acc65I

37 D

Product	Size	Conc.	Cat.#
Acc65I	1,500 u	10 u/µl	R6921

For Laboratory Use.

Description: G▼GTAC C
C CATG▲G
Storage Conditions: Store at –20°C.

AccB7I

37 E

Product	Size	Conc.	Cat.#
AccB7I	200 u	12 u/µl	R7081

For Laboratory Use.

Description: CCAN NNN▼NTGG
GGTN▲NNN NACC
Storage Conditions: Store at –20°C.

AgeI

37 K

Product	Size	Conc.	Cat.#
AgeI	100 u	3–10 u/µl	R7251

For Laboratory Use.

Description: A▼CCGG T
T GGCC▲A
Storage Conditions: Store at –20°C.

AluI

37 B

Product	Size	Conc.	Cat.#
AluI	500 u	10 u/µl	R6281

For Laboratory Use.

Description: AG▼CT
TC▲GA
Storage Conditions: Store at –20°C.

ApaI

37 A

Product	Size	Conc.	Cat.#
ApaI	5,000 u	10 u/µl	R6361
ApaI (HC)	25,000 u	40–80 u/µl	R4364

For Laboratory Use.

Description: G GGCC▼C
C▲CCGG G
Storage Conditions: Store at –20°C.

AvaI

37 B

Product	Size	Conc.	Cat.#
AvaI	200 u	8–12 u/µl	R6091
	1,000 u	8–12 u/µl	R6095

For Laboratory Use.

Description: C▼(T/C)CG(A/G) G
G (A/G)GC(T/C)▲C
Storage Conditions: Store at –20°C.

AvaII

37 C

Product	Size	Conc.	Cat.#
AvaII	100 u	1–10 u/µl	R6131
	1,000 u	1–10 u/µl	R6135

For Laboratory Use.

Description: G▼G(A/T)C C
C C(T/A)G▲G
Storage Conditions: Store at –20°C.

BalI

37 G

Product	Size	Conc.	Cat.#
BalI	50 u	2–10 u/µl	R6691
	250 u	2–10 u/µl	R6695

For Laboratory Use.

Description: TGG▼CCA
ACC▲GGT
Storage Conditions: Store at –20°C.

BamHI

Product	Size	Conc.	Cat.#
BamHI	2,500 u	10 u/µl	R6021
	12,500 u	10 u/µl	R6025
BamHI (HC)	12,500 u	40–80 u/µl	R4024
	50,000 u	40–80 u/µl	R4027

For Laboratory Use.

Description: G▼GATC C
C CTAG▲G

Storage Conditions: Store at –20°C.

BanI

Product	Size	Conc.	Cat.#
BanI	200 u	8–12 u/µl	R6891

For Laboratory Use.

Description: G▼G(T/C)(A/G)C C
C C(A/G)(T/C)G▲G

Storage Conditions: Store at –20°C.

BanII

Product	Size	Conc.	Cat.#
BanII	1,000 u	8–12 u/µl	R6561

For Laboratory Use.

Description: G (A/G)GC(T/C)▼C
C▲(T/C)CG(A/G) G

Storage Conditions: Store at –20°C.

BbuI

Product	Size	Conc.	Cat.#
BbuI	200 u	10 u/µl	R6621
BbuI (HC)	1,000 u	40–80 u/µl	R4624

For Laboratory Use.

Description: G CATG▼C
C▲GTAC G

Storage Conditions: Store at –20°C.

BclI

Product	Size	Conc.	Cat.#
BclI	1,000 u	10 u/µl	R6651

For Laboratory Use.

Description: T▼GATC A
A CTAG▲T

Storage Conditions: Store at –20°C.

BglI

Product	Size	Conc.	Cat.#
BglI	1,000 u	10 u/µl	R6071
	5,000 u	10 u/µl	R6077
BglI (HC)	5,000 u	40–80 u/µl	R4074

For Laboratory Use.

Description: GCCN NNN▼NGGC
CGGN▲NNN NCCG

Storage Conditions: Store at –20°C.

BglII

Product	Size	Conc.	Cat.#
BglII	500 u	10 u/µl	R6081
	2,500 u	10 u/µl	R6085
	10,000 u	10 u/µl	R6087

For Laboratory Use.

Description: A▼GATC T
T CTAG▲A

Storage Conditions: Store at –20°C.

BsaMI

Product	Size	Conc.	Cat.#
BsaMI	500 u	10 u/µl	R6991

For Laboratory Use.

Description: GAATG CN▼
CTTAC▲GN

Storage Conditions: Store at –20°C.

Bsp1286I

Product	Size	Conc.	Cat.#
Bsp1286I	500 u	10 u/µl	R6741

For Laboratory Use.

Description: G (G/A/T)GC(C/A/T)▼C
C▲(C/T/A)CG(G/T/A) G

Storage Conditions: Store at –20°C.

BsrSI

Product	Size	Conc.	Cat.#
BsrSI	500 u	10 u/µl	R7241

For Laboratory Use.

Description: ACTG GN▼
TGAC▲CN

Storage Conditions: Store at –20°C.

BssHII

50° H

Product	Size	Conc.	Cat.#
BssHII	100 u	10 u/µl	R6831
	500 u	10 u/µl	R6835

For Laboratory Use.

Description: G▼CGCG C
C GCGC▲G
Storage Conditions: Store at –20°C.

Bst98I

37° D

Product	Size	Conc.	Cat.#
Bst98I	500 u	8–12 u/µl	R7141

For Laboratory Use.

Description: C▼TTAA G
G AATT▲C
Storage Conditions: Store at –20°C.

BstEII

60° D

Product	Size	Conc.	Cat.#
BstEII	2,000 u	10 u/µl	R6641

For Laboratory Use.

Description: G▼GTNAC C
C CANTG▲G
Storage Conditions: Store at –20°C.

BstOI

60° C

Product	Size	Conc.	Cat.#
BstOI	2,000 u	10 u/µl	R6931

For Laboratory Use.

Description: CC▼(A/T) GG
GG (T/A)▲CC
Storage Conditions: Store at –20°C.

BstXI

50° D

Product	Size	Conc.	Cat.#
BstXI	250 u	8–12 u/µl	R6471
	1,000 u	8–12 u/µl	R6475

For Laboratory Use.

Description: CCAN NNNN▼NTGG
GGTN▲NNNN NACC
Storage Conditions: Store at –20°C.

BstZI

50° D

Product	Size	Conc.	Cat.#
BstZI	500 u	10 u/µl	R6881

For Laboratory Use.

Description: C▼GGCC G
G CCGG▲C
Storage Conditions: Store at –20°C.

Bsu36I

37° E

Product	Size	Conc.	Cat.#
Bsu36I	500 u	10 u/µl	R6821

For Laboratory Use.

Description: CC▼TNA GG
GG ANT▲CC
Storage Conditions: Store at –20°C.

CfoI

37° B

Product	Size	Conc.	Cat.#
CfoI	3,000 u	10 u/µl	R6241

For Laboratory Use.

Description: G CG▼C
C▲GC G
Storage Conditions: Store at –20°C.

ClaI

37° C

Product	Size	Conc.	Cat.#
ClaI	500 u	10 u/µl	R6551
	2,500 u	10 u/µl	R6555

For Laboratory Use.

Description: AT▼CG AT
TA GC▲TA
Storage Conditions: Store at –20°C.

CspI

30° K

Product	Size	Conc.	Cat.#
CspI	100 u	10 u/µl	R6671
	500 u	10 u/µl	R6675

For Laboratory Use.

Description: CG▼G(A/T)C CG
GC C(T/A)G▲GC
Storage Conditions: Store at –20°C.

Csp45I

37° B

Product	Size	Conc.	Cat.#
Csp45I	2,500 u	10 u/µl	R6571

For Laboratory Use.

Description: TT▼CG AA
AA GC▲TT
Storage Conditions: Store at –20°C.

DdeI

37° D

Product	Size	Conc.	Cat.#
DdeI	200 u	10 u/µl	R6291
	1,000 u	10 u/µl	R6295

For Laboratory Use.

Description: C▼TNA G
G ANT▲C
Storage Conditions: Store at –20°C.

DpnI

Product	Size	Conc.	Cat.#
DpnI	200 u	10 u/µl	R6231
For Laboratory Use.			

Description: G^{me}A▼TC
CT▲^{me}AG

Storage Conditions: Store at –20°C.

DraI

Product	Size	Conc.	Cat.#
DraI	2,000 u	10 u/µl	R6271
For Laboratory Use.			

Description: TTT▼AAA
AAA▲TTT

Storage Conditions: Store at –20°C.

Eco47III

Product	Size	Conc.	Cat.#
Eco47III	50 u	2–5 u/µl	R6731
For Laboratory Use.			

Description: AGC▼GCT
TCG▲CGA

Storage Conditions: Store at –20°C.

EcoICRI

Product	Size	Conc.	Cat.#
EcoICRI	1,000 u	10 u/µl	R6951
EcoICRI (HC)	5,000 u	40–80 u/µl	R4954
For Laboratory Use.			

Description: GAG▼CTC
CTC▲GAG

Storage Conditions: Store at –20°C.

EcoRI

Product	Size	Conc.	Cat.#
EcoRI	5,000 u	12 u/µl	R6011
	15,000 u	12 u/µl	R6017
EcoRI (HC)	25,000 u	40–80 u/µl	R4014
	50,000 u	40–80 u/µl	R4017
For Laboratory Use.			

Description: G▼AATT C
C TTAA▲G

Storage Conditions: Store at –20°C.

EcoRV

Product	Size	Conc.	Cat.#
EcoRV	2,000 u	10 u/µl	R6351
	10,000 u	10 u/µl	R6355
EcoRV (HC)	10,000 u	40–80 u/µl	R4354
For Laboratory Use.			

Description: GAT▼ATC
CTA▲TAG

Storage Conditions: Store at –20°C.

HaeII

Product	Size	Conc.	Cat.#
HaeII	1,000 u	10 u/µl	R6661
For Laboratory Use.			

Description: (A/G) GCGC▼(T/C)
(T/C)▲CGCG (A/G)

Storage Conditions: Store at –20°C.

HaeIII

Product	Size	Conc.	Cat.#
HaeIII	2,500 u	10 u/µl	R6171
	10,000 u	10 u/µl	R6175
HaeIII (HC)	12,500 u	40–80 u/µl	R4174
For Laboratory Use.			

Description: GG▼CC
CC▲GG

Storage Conditions: Store at –20°C.

HhaI

Product	Size	Conc.	Cat.#
HhaI	1,000 u	10 u/µl	R6441
For Laboratory Use.			

Description: G CG▼C
C▲GC G

Storage Conditions: Store at –20°C.

HincII

Product	Size	Conc.	Cat.#
HincII	200 u	10 u/µl	R6031
	1,000 u	10 u/µl	R6035
	5,000 u	10 u/µl	R6037
For Laboratory Use. Please Note: High Concentration HincII (Cat.# R4034) has been discontinued. Please order a HincII at standard concentration as listed above.			

Description: GT(T/C)▼(A/G)AC
CA(A/G)▲(T/C)TG

Storage Conditions: Store at –20°C.

HindIII

37° E

Product	Size	Conc.	Cat.#
HindIII	5,000 u	10 u/µl	R6041
	15,000 u	10 u/µl	R6045
HindIII (HC)	25,000 u	40–80 u/µl	R4044
	50,000 u	40–80 u/µl	R4047

For Laboratory Use.

Description: A▼AGCT T
T TCGA▲A
Storage Conditions: Store at –20°C.

HinfI

37° B

Product	Size	Conc.	Cat.#
HinfI	1,000 u	10 u/µl	R6201
	5,000 u	10 u/µl	R6205
HinfI (HC)	5,000 u	40–80 u/µl	R4204

For Laboratory Use.

Description: G▼ANT C
C TNA▲G
Storage Conditions: Store at –20°C.

HpaI

37° J

Product	Size	Conc.	Cat.#
HpaI	100 u	3–10 u/µl	R6301
	500 u	3–10 u/µl	R6305

For Laboratory Use.

Description: GTT▼AAC
CAA▲TTG
Storage Conditions: Store at –20°C.

HpaII

37° A

Product	Size	Conc.	Cat.#
HpaII	1,000 u	10 u/µl	R6311
	5,000 u	10 u/µl	R6315

For Laboratory Use.

Description: C▼CG G
G GC▲C
Storage Conditions: Store at –20°C.

Hsp92I

37° F

Product	Size	Conc.	Cat.#
Hsp92I	500 u	10 u/µl	R7151

For Laboratory Use.

Description: G(A/G)▼CG (T/C)C
C(T/C) GC▲(A/G)G
Storage Conditions: Store at –20°C.

Hsp92II

37° K

Product	Size	Conc.	Cat.#
Hsp92II	1,000 u	10 u/µl	R7161

For Laboratory Use.

Description: CATG▼
▲GTAC
Storage Conditions: Store at –20°C.

I-PpoI (Intron-Encoded Endonuclease)

37°

Product	Size	Conc.	Cat.#
I-PpoI	10,000 u	100–200 u/µl	R7031

For Laboratory Use.

Description: CTCTC TTAA▼GGTAGC
GAGAG▲AATT CCATCG
Storage Conditions: Store at –20°C.

KpnI

37° J

Product	Size	Conc.	Cat.#
KpnI	2,500 u	8–12 u/µl	R6341
	10,000 u	8–12 u/µl	R6345
KpnI (HC)	12,500 u	40–80 u/µl	R4344

For Laboratory Use.

Description: G GTAC▼C
C▲CATG G
Storage Conditions: Store at –20°C.

MboI

37° C

Product	Size	Conc.	Cat.#
MboI	200 u	8–12 u/µl	R6711

For Laboratory Use.

Description: ▼GATC
CTAG▲
Storage Conditions: Store at –20°C.

MboII

37° B

Product	Size	Conc.	Cat.#
MboII	100 u	2–10 u/µl	R6723

For Laboratory Use.

Description: GAAGA$(N)_8$▼
CTTCT$(N)_7$▲
Storage Conditions: Store at –20°C.

MluI

37° D

Product	Size	Conc.	Cat.#
MluI	1,000 u	10 u/µl	R6381

For Laboratory Use.

Description: A▼CGCG T
T GCGC▲A
Storage Conditions: Store at –20°C.

MspI

Product	Size	Conc.	Cat.#
MspI	2,000 u	10 u/µl	R6401
	10,000 u	10 u/µl	R6405
MspI (HC)	10,000 u	40–80 u/µl	R4404

For Laboratory Use.

Description: C▼CG G
G GC▲C

Storage Conditions: Store at –20°C.

MspA1I

Product	Size	Conc.	Cat.#
MspA1I	1,000 u	10 u/µl	R7021

For Laboratory Use.

Description: C(A/C)G▼C(G/T)G
G(T/G)C▲G(C/A)C

Storage Conditions: Store at –20°C.

NaeI

Product	Size	Conc.	Cat.#
NaeI	250 u	4 u/µl	R7131
	1,000 u	4 u/µl	R7135

For Laboratory Use.

Description: GCC▼GGC
CGG▲CCG

Storage Conditions: Store at –20°C.

NarI

Product	Size	Conc.	Cat.#
NarI	200 u	10 u/µl	R6861

For Laboratory Use.

Description: GG▼CG CC
CC GC▲GG

Storage Conditions: Store at –20°C.

NciI

Product	Size	Conc.	Cat.#
NciI	1,000 u	10 u/µl	R7061

For Laboratory Use.

Description: CC▼(C/G) GG
GG (G/C)▲CC

Storage Conditions: Store at –20°C.

NcoI

Product	Size	Conc.	Cat.#
NcoI	200 u	10 u/µl	R6513
	1,000 u	10 u/µl	R6515

For Laboratory Use.

Description: C▼CATG G
G GTAC▲C

Storage Conditions: Store at –20°C.

NdeI

Product	Size	Conc.	Cat.#
NdeI	500 u	10 u/µl	R6801

For Laboratory Use.

Description: CA▼TA TG
GT AT▲AC

Storage Conditions: Store at –20°C.

NdeII

Product	Size	Conc.	Cat.#
NdeII	200 u	10u/ µl	R7291
	1,000 u	10u/ µl	R7295

For Laboratory Use.

Description: ▼GATC
CTAG▲

Storage Conditions: Store at –20°C.

NheI

Product	Size	Conc.	Cat.#
NheI	250 u	10 u/µl	R6501
	1,250 u	10 u/µl	R6505

For Laboratory Use.

Description: G▼CTAG C
C GATC▲G

Storage Conditions: Store at –20°C.

NotI

Product	Size	Conc.	Cat.#
NotI	200 u	10 u/µl	R6431
	1,000 u	10 u/µl	R6435
NotI (HC)	1,000 u	40–80 u/µl	R4434

For Laboratory Use.

Description: GC▼GGCC GC
CG CCGG▲CG

Storage Conditions: Store at –20°C.

NruI

Product	Size	Conc.	Cat.#
NruI	200 u	10 u/µl	R7091

For Laboratory Use.

Description: TCG▼CGA
AGC▲GCT

Storage Conditions: Store at –20°C.

NsiI

37 D

Product	Size	Conc.	Cat.#
NsiI	250 u	10 u/µl	R6531

For Laboratory Use.

Description: A TGCA▼T
T▲ACGT A
Storage Conditions: Store at –20°C.

PstI

37 H

Product	Size	Conc.	Cat.#
PstI	3,000 u	10 u/µl	R6111
	15,000 u	10 u/µl	R6115
PstI (HC)	15,000 u	40–80 u/µl	R4114
	50,000 u	40–80 u/µl	R4117

For Laboratory Use.

Description: C TGCA▼G
G▲ACGT C
Storage Conditions: Store at –20°C.

PvuI

37 D

Product	Size	Conc.	Cat.#
PvuI	100 u	2–10 u/µl	R6321
	500 u	2–10 u/µl	R6325

For Laboratory Use.

Description: CG AT▼CG
GC▲TA GC
Storage Conditions: Store at –20°C.

PvuII

37 B

Product	Size	Conc.	Cat.#
PvuII	1,000 u	8–12 u/µl	R6331
	5,000 u	8–12 u/µl	R6335

For Laboratory Use.

Description: CAG▼CTG
GTC▲GAC
Storage Conditions: Store at –20°C.

RsaI

37 C

Product	Size	Conc.	Cat.#
RsaI	1,000 u	10 u/µl	R6371
RsaI (HC)	5,000 u	40–80 u/µl	R4374

For Laboratory Use.

Description: GT▼AC
CA▲TG
Storage Conditions: Store at –20°C.

SacI

37 J

Product	Size	Conc.	Cat.#
SacI	1,000 u	10 u/µl	R6061
	5,000 u	10 u/µl	R6065
SacI (HC)	5,000 u	40–80 u/µl	R4064

For Laboratory Use.

Description: G AGCT▼C
C▲TCGA G
Storage Conditions: Store at –20°C.

SacII

37 C

Product	Size	Conc.	Cat.#
SacII	500 u	10 u/µl	R6221

For Laboratory Use.

Description: CC GC▼GG
GG▲CG CC
Storage Conditions: Store at –20°C.

SalI

37 D

Product	Size	Conc.	Cat.#
SalI	2,000 u	10 u/µl	R6051
	10,000 u	10 u/µl	R6055
SalI (HC)	10,000 u	40–80 u/µl	R4054

For Laboratory Use.

Description: G▼TCGA C
C AGCT▲G
Storage Conditions: Store at –20°C.

Sau3AI

37 B

Product	Size	Conc.	Cat.#
Sau3AI	100 u	3–10 u/µl	R6191
	500 u	3–10 u/µl	R6195

For Laboratory Use.

Description: ▼GATC
CTAG▲
Storage Conditions: Store at –20°C.

ScaI

37 K

Product	Size	Conc.	Cat.#
ScaI	1,000 u	8–12 u/µl	R6211
ScaI (HC)	5,000 u	40–80 u/µl	R4214

For Laboratory Use.

Description: AGT▼ACT
TCA▲TGA
Storage Conditions: Store at –20°C.

SfiI

50° B

Product	Size	Conc.	Cat.#
SfiI	250 u	10 u/µl	R6391
SfiI (HC)	1,250 u	40–80 u/µl	R4394

For Laboratory Use.

Description: GGCCN NNN▼NGGCC
CCGGN▲NNN NCCGG

Storage Conditions: Store at –20°C.

SgfI

37° C

Product	Size	Conc.	Cat.#
SgfI	250 u	8–12 u/µl	R7103
SgfI (HC)	1,250 u	40–80 u/µl	R5104

For Laboratory Use.

Description: GCG AT▼CGC
CGC▲TA GCG

Storage Conditions: Store at –20°C. **Do not freeze.**

SinI

37° A

Product	Size	Conc.	Cat.#
SinI (HC)	1,000 u	40–80 u/µl	R4144

For Laboratory Use.

Description: G▼G(A/T)C C
C C(T/A)G▲G

Storage Conditions: Store at –20°C.

SmaI

25° J

Product	Size	Conc.	Cat.#
SmaI	1,000 u	8–12 u/µl	R6121
	5,000 u	8–12 u/µl	R6125
SmaI (HC)	5,000 u	40–80 u/µl	R4124

For Laboratory Use.

Description: CCC▼GGG
GGG▲CCC

Storage Conditions: Store at –20°C.

SnaBI

37° B

Product	Size	Conc.	Cat.#
SnaBI	100 u	2–10 u/µl	R6791
	500 u	2–10 u/µl	R6795

For Laboratory Use.

Description: TAC▼GTA
ATG▲CAT

Storage Conditions: Store at –20°C.

SpeI

37° B

Product	Size	Conc.	Cat.#
SpeI	200 u	10 u/µl	R6591
	1,000 u	10 u/µl	R6595

For Laboratory Use.

Description: A▼CTAG T
T GATC▲A

Storage Conditions: Store at –20°C.

SphI

37° K

Product	Size	Conc.	Cat.#
SphI	200 u	10 u/µl	R6261
	1,000 u	10 u/µl	R6265

For Laboratory Use.

Description: G CATG▼C
C▲GTAC G

Storage Conditions: Store at –20°C.

SspI

37° E

Product	Size	Conc.	Cat.#
SspI	500 u	10 u/µl	R6601
SspI (HC)	2,500 u	40–80 u/µl	R4604

For Laboratory Use.

Description: AAT▼ATT
TTA▲TAA

Storage Conditions: Store at –20°C.

StuI

37° B

Product	Size	Conc.	Cat.#
StuI	400 u	10 u/µl	R6421

For Laboratory Use.

Description: AGG▼CCT
TCC▲GGA

Storage Conditions: Store at –20°C.

StyI

37° F

Product	Size	Conc.	Cat.#
StyI	2,000 u	10 u/µl	R6481

For Laboratory Use.

Description: C▼C(A/T)(T/A)G G
G G(T/A)(A/T)C▲C

Storage Conditions: Store at –20°C.

TaqI

65° E

Product	Size	Conc.	Cat.#
TaqI	1,000 u	10 u/µl	R6151
	10,000 u	10 u/µl	R6155
TaqI (HC)	5,000 u	40–80 u/µl	R4154

For Laboratory Use.

Description: T▼CG A
A GC▲T
Storage Conditions: Store at –20°C.

Tru9I

65° F

Product	Size	Conc.	Cat.#
Tru9I	200 u	8–12 u/µl	R7011

For Laboratory Use.

Description: T▼TA A
A AT▲T
Storage Conditions: Store at –20°C.

Tth111I

65° B

Product	Size	Conc.	Cat.#
Tth111I	500 u	8–12 u/µl	R6841

For Laboratory Use.

Description: GACN▼N NGTC
CTGN N▲NCAG
Storage Conditions: Store at –20°C.

VspI

37° D

Product	Size	Conc.	Cat.#
VspI	500 u	8–12 u/µl	R6851

For Laboratory Use.

Description: AT▼TA AT
TA AT▲TA
Storage Conditions: Store at –20°C.

XbaI

37° D

Product	Size	Conc.	Cat.#
XbaI	2,000 u	8–12 u/µl	R6181
	10,000 u	8–12 u/µl	R6185
XbaI (HC)	10,000 u	40–80 u/µl	R4184

For Laboratory Use.

Description: T▼CTAG A
A GATC▲T
Storage Conditions: Store at –20°C.

XhoI

37° D

Product	Size	Conc.	Cat.#
XhoI	3,000 u	10 u/µl	R6161
	10,000 u	10 u/µl	R6165
XhoI (HC)	15,000 u	40–80 u/µl	R4164

For Laboratory Use.

Description: C▼TCGA G
G AGCT▲C
Storage Conditions: Store at –20°C.

XhoII

37° C

Product	Size	Conc.	Cat.#
XhoII	100 u	5–10 u/µl	R6811
	500 u	5–10 u/µl	R6815

For Laboratory Use.

Description: (A/G)▼GATC (T/C)
(T/C) CTAG▲(A/G)
Storage Conditions: Store at –20°C.

XmaI

37° B

Product	Size	Conc.	Cat.#
XmaI	50 u	1–5 u/µl	R6491
	250 u	1–5 u/µl	R6495

For Laboratory Use.

Description: C▼CCGG G
G GGCC▲C
Storage Conditions: Store at –20°C.

XmnI

37° B

Product	Size	Conc.	Cat.#
XmnI	500 u	10 u/µl	R7271
	2,500 u		R7273

For Laboratory Use.

Description: GAANN▼NNTTC
CTTNN▲NNAAG
Storage Conditions: Store at –20°C.

MULTI-CORE™ Buffer Pack

Product	Size	Cat.#
MULTI-CORE™ Buffer Pack	3 × 1 ml	R9991
For Laboratory Use.		

Description: The MULTI-CORE™ Buffer Pack contains convenient aliquots of the Promega universal restriction enzyme 10X buffer. The MULTI-CORE™ Buffer is formulated to provide simple buffering conditions for performing multiple digestions. Many Promega restriction enzymes have between 50% and 100% activity in reactions using MULTI-CORE™ Buffer.

Features:

- **Convenient and Economical:** MULTI-CORE™ Buffer enables co-digestion of DNA with more than one enzyme in a single reaction. In most cases, only modest adjustments in the amount of enzyme used will ensure complete multiple digestions.

Storage Conditions: Store at –20°C.

4-CORE® Buffer Pack

Product	Size	Cat.#
4-CORE® Buffer Pack (Buffers A, B, C and D), 1ml each	4ml (4 × 1 ml)	R9921
For Laboratory Use.		

Description: The 4-CORE® Buffer Pack contains convenient aliquots of Promega Restriction Enzyme 10X Buffers A, B, C and D. The majority of Promega restriction enzymes have optimal activity in one of these four 10X reaction buffers.

Storage Conditions: Store at –20°C.

Promega Flipper® Racks

Product	Size	Cat.#
Promega Flipper® Rack, Blue	8 × 8 tubes	Y9341
Promega Flipper® Rack, Purple	8 × 12 tubes	Y9422

Description: The versatile Promega Flipper® Racks are ideal for storage and transport of all your small tubes. These polypropylene racks withstand extreme temperatures, making them an excellent choice for freezer storage. They may also be autoclaved for use in sterile environments. Each rack is two-sided; one side accommodates 0.5ml microcentrifuge tubes, the other 1.5ml tubes or 2ml cryogenic tubes. The Blue Flipper® Rack holds 64 tubes and the Purple Flipper® Rack holds 96 tubes. Clear lids permit easy viewing of rack contents.

Features:

- **Withstand Extreme Temperatures:** Blue Flipper® Racks may be stored at –90°C; Purple Flipper® Racks at –30°C. Both may be autoclaved.
- **Convenient:** Store 0.5ml, 1.5ml or 2ml tubes.

Storage Conditions: Minimum storage temperature: Blue, –90°C; Purple, –30°C. Maximum temperature: Autoclavable.

Promega Flipper® Rack. Purple (Cat.# Y9422).

Promega Flipper® Rack. Blue (Cat.# Y9341).

DNA Polymerase I

Product	Size Conc.	Cat.#
DNA Polymerase I	500 u 5–10 u/µl	M2051
	2,500 u 5–10 u/µl	M2055

For Laboratory Use.

Description: DNA Polymerase I catalyzes the template-directed polymerization of nucleotides into duplex DNA in a 5′→3′ direction. DNA Polymerase I possesses a 3′→5′ exonuclease activity or "proofreading" function, which lowers the error rate during DNA replication, and a 5′→3′ exonuclease activity, which enables the enzyme to replace nucleotides in the growing strand of DNA by nick translation. The enzyme, purified from recombinant *E. coli*, is capable of catalyzing de novo synthesis of synthetic homopolymers and provides a convenient method for the preparation of a variety of defined DNA substrates.

Features:

- **Flexible:** DNA Polymerase I may be used in a variety of molecular applications.
- **May Be Heat-Inactivated:** DNA Polymerase I is inactivated by heating at 68°C for 10 minutes.
- **Provided with 10X Reaction Buffer:** 500mM Tris-HCl (pH 7.2 at 25°C), 100mM $MgSO_4$, 1mM DTT.

Protocol	Part#
Promega Product Information	9PIM205

Storage Conditions: Store at –20°C.

DNA Polymerase I Large (Klenow) Fragment

Product	Size	Conc.	Cat.#
DNA Polymerase I Large (Klenow) Fragment	150 u	5 u/µl	M2201
	500 u	5 u/µl	M2206

For Laboratory Use.

Description: DNA Polymerase I Large (Klenow) Fragment is a DNA-dependent DNA polymerase that lacks the 5′→3′ exonuclease activity of intact *E. coli* DNA Polymerase I but retains its 5′→3′ polymerase, 3′→5′ exonuclease and strand displacement activities. The enzyme is a 68kDa C-terminal fragment of DNA Polymerase I. The 5′→3′ polymerase activity of Klenow Fragment can be used to fill in 5′-protruding ends with unlabeled or labeled dNTPs, to sequence single- or double-stranded DNA templates, for in vitro mutagenesis using synthetic oligonucleotides, for cDNA second-strand synthesis and to generate single-stranded DNA probes. The 3′→5′ exonuclease activity can be used to generate blunt ends from a 3′-overhang.

Features:

- **Flexible:** DNA Polymerase I Large (Klenow) Fragment may be used in a variety of molecular applications. It is also active in many Promega 1X restriction enzyme buffers.
- **May Be Heat-Inactivated:** DNA Polymerase I Large (Klenow) Fragment is inactivated by heating at 75°C for 10 minutes.
- **Provided with 10X Reaction Buffer:** 500mM Tris-HCl (pH 7.2 at 25°C), 100mM $MgSO_4$, 1mM DTT.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway/**

Protocol	Part#
Promega Product Information	9PIM220

Storage Conditions: Store at –20°C.

DNA Polymerase I Large (Klenow) Fragment, Exonuclease Minus

Product	Size	Conc.	Cat.#
Klenow Fragment, Exonuclease Minus	100 u	5–10 u/µl	M2181

For Laboratory Use.

Description: DNA Polymerase I Large (Klenow) Fragment, Exonuclease Minus, is a DNA-dependent DNA polymerase that lacks both the 5′→3′ and the 3′→5′ exonuclease activities present in intact *E. coli* DNA Polymerase I. It is used for random primer labeling and in strand displacement amplification. Klenow Fragment, Exonuclease Minus, will leave a single-base 3′ overhang on a significant proportion of DNA fragments during fill-in of 5′-overhangs. Therefore, this enzyme is not recommended for preparation of blunt-ended fragments for ligation.

Features:

- **Provided with 10X Reaction Buffer:** 500mM Tris-HCl (pH 7.2 at 25°C), 100mM $MgSO_4$, 1mM DTT.
- **May Be Heat-Inactivated:** DNA Polymerase I Large (Klenow) Fragment, Exonuclease Minus, is inactivated by heating at 75°C for 10 minutes

Protocol	Part#
Promega Product Information	9PIM218

Storage Conditions: Store at –20°C.

DNA Polymerase I Large (Klenow) Fragment Mini Kit

Product	Size	Cat.#
DNA Polymerase I Large (Klenow) Fragment Mini Kit	150 u	U1300

Available Separately	Size	Conc.	Cat.#
DNA Polymerase I Large (Klenow) Fragment	150 u	5 u/µl	M2201
Set of dATP, dCTP, dGTP, dTTP	10µmol each	100 mM	U1330

For Laboratory Use.

Description: The DNA Polymerase I Large (Klenow) Fragment Mini Kit provides a convenient combination of polymerase and dNTPs. The kit contains 5µmol each of dATP, dGTP, dTTP and dCTP (10mM in water) and DNA Polymerase I Large (Klenow) Fragment, ready for use in a variety of applications.

Features:

- **Convenient:** The kit provides DNA Polymerase I Large (Klenow) Fragment and dNTPs conveniently packaged and ready to use in your application.

Storage Conditions: Store at –20°C.

T4 DNA Polymerase

Product	Size	Conc.	Cat.#
T4 DNA Polymerase	100 u	5–10 u/µl	M4211
	500 u	5–10 u/µl	M4215

For Laboratory Use.

Description: T4 DNA Polymerase catalyzes the 5′→3′ synthesis of DNA from a primed single-stranded DNA template. Although possessing a potent 3′→5′ proofreading exonuclease, T4 DNA Polymerase contains no 5′→3′ exonuclease activity. T4 DNA Polymerase can be used to fill 5′ protruding ends with labeled or unlabeled dNTPs or for the generation of blunt ends from DNA molecules with 3′ overhangs.

Features:

- **High Fidelity:** T4 DNA Polymerase is the enzyme of choice for applications where misincorporation is a concern.
- **Flexible:** T4 DNA Polymerase may be used in a variety of molecular applications. Active in many Promega 1X restriction enzyme buffers.
- **May Be Heat-Inactivated:** T4 DNA Polymerase is inactivated by heating at 75°C for 10 minutes.
- **Provided with 10X Reaction Buffer:** 250mM Tris-acetate (pH 7.7), 1M potassium acetate, 100mM magnesium acetate and 10mM DTT.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Protocol	Part#
Promega Product Information	9PIM421

Storage Conditions: Store at –20°C.

Terminal Deoxynucleotidyl Transferase, Recombinant

Product	Size	Conc.	Cat.#
Terminal Deoxynucleotidyl Transferase, Recombinant	300 u	30 u/µl	M1871
	1,500 u	30 u/µl	M1875
Available Separately		**Size**	**Cat.#**
Terminal Transferase Buffer Pack		3 × 500 µl	M1893

For Laboratory Use.

Description: Terminal Deoxynucleotidyl Transferase, Recombinant, catalyzes the repetitive addition of mononucleotides to the terminal 3′-OH of a DNA initiator accompanied by the release of inorganic phosphate. Single-stranded DNA is preferred as an initiator. Polymerization is not template-dependent. The addition of 1mM Co^{2+} (as $CoCl_2$) in the reaction buffer allows the tailing of 3′-ends with varying degrees of efficiency.

Features:

- **Tails Any Type of 3′ End:** The presence of 1mM $CoCl_2$ in the reaction buffer allows the tailing of any type of 3′ end (3′ and 5′ overhangs or blunt ends).
- **Tested for Apoptotic DNA Labeling:** Each lot of enzyme is qualified for success in the procedure outlined in the *DeadEnd™ Fluorometric TUNEL System Technical Bulletin* #TB235.
- **Provided with 5X Reaction Buffer:** 500mM cacodylate buffer (pH 6.8 at 25°C), 5mM $CoCl_2$, 0.5mM DTT.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Protocol	Part#
Promega Product Information	9PIM187

Storage Conditions: Store at –20°C.

SP6 RNA Polymerase

Product	Size	Conc.	Cat.#
SP6 RNA Polymerase	1,000 u	10–20 u/µl	P1085
	5,000 u	10–20 u/µl	P1081
SP6 RNA Polymerase (HC)	2,500 u	80 u/µl	P4084

For Laboratory Use.

Description: SP6 RNA Polymerase is a DNA-dependent RNA polymerase that exhibits extremely high specificity for its cognate promoter sequences. Only SP6 DNA or DNA cloned downstream from an SP6 promoter can serve as a template for SP6 RNA Polymerase-directed RNA synthesis.

Features:

- **Specific:** SP6 RNA Polymerase exhibits extremely high affinity and specificity for SP6 promoter sequences.
- **Highly Pure:** SP6 RNA Polymerase is >90% pure as determined by SDS polyacrylamide gel electrophoresis. Free of detectable levels of contaminating RNase and DNase activity.
- **Flexible:** Will incorporate ^{32}P, ^{33}P, ^{3}H and ^{35}S nucleoside triphosphates.
- **Provided with 5X Reaction Buffer:** Provided with 100mM DTT and Transcription Optimized 5X Buffer: 200mM Tris-HCl (pH 7.9 at 25°C), 30mM $MgCl_2$, 10mM spermidine, 50mM NaCl.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Protocol	Part#
Promega Product Information	9PIP108

Storage Conditions: Store at –20°C.

T3 RNA Polymerase

Product	Size	Conc.	Cat.#
T3 RNA Polymerase	1,000 u	10–20 u/µl	P2083
T3 RNA Polymerase (HC)	2,500 u	80 u/µl	P4024

For Laboratory Use.

Description: T3 RNA Polymerase is a DNA-dependent RNA polymerase that exhibits extremely high specificity for its cognate promoter sequences. Only T3 DNA or DNA cloned downstream from a T3 promoter can serve as a template for T3 RNA Polymerase-directed RNA synthesis.

Features:

- **Specific:** T3 RNA Polymerase exhibits extremely high affinity and specificity for T3 promoter sequences.
- **Highly Pure:** T3 RNA Polymerase is >90% pure as determined by SDS polyacrylamide gel electrophoresis. Free of detectable levels of contaminating RNase and DNase activity
- **Flexible:** Will incorporate ^{32}P, ^{33}P, ^{3}H and ^{35}S nucleoside triphosphates.
- **Provided with 5X Reaction Buffer:** Provided with 100mM DTT and Transcription Optimized 5X Buffer: 200mM Tris-HCl (pH 7.9 at 25°C), 30mM $MgCl_2$, 10mM spermidine, 50mM NaCl.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Protocol	Part#
Promega Product Information	9PIP208

Storage Conditions: Store at –20°C.

T7 RNA Polymerase

Product	Size	Conc.	Cat.#
T7 RNA Polymerase	1,000 u	10–20 u/µl	P2075
	5,000 u	10–20 u/µl	P2077
T7 RNA Polymerase (HC)	10,000 u	80 u/µl	P4074

For Laboratory Use.

Description: T7 RNA Polymerase is a DNA-dependent RNA polymerase that exhibits extremely high specificity for its cognate promoter sequences. Only T7 DNA or DNA cloned downstream from a T7 promoter can serve as a template for T7 RNA Polymerase-directed RNA synthesis.

Features:

- **Specific:** T7 RNA Polymerase exhibits extremely high affinity and specificity for T7 promoter sequences.
- **Highly Pure:** T7 RNA Polymerase is judged to be greater than 90% pure as determined by SDS polyacrylamide gel electrophoresis. Free of detectable levels of contaminating RNase and DNase activity.
- **Flexible:** Will incorporate ^{32}P, ^{33}P, ^{3}H and ^{35}S nucleoside triphosphates.
- **Provided with 5X Reaction Buffer:** Provided with 100mM DTT and Transcription Optimized 5X Buffer: 200mM Tris-HCl (pH 7.9 at 25°C), 30mM $MgCl_2$, 10mM spermidine, 50mM NaCl.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Protocol	Part#
Promega Product Information	9PIP207

Storage Conditions: Store at –20°C.

GoScript™ Reverse Transcriptase

Product	Size	Cat.#
GoScript™ Reverse Transcriptase	100 reactions	A5003
	500 reactions	A5004
Available Separately	**Size**	**Cat.#**
GoScript™ Reverse Transcription System	50 reactions	A5000
	100 reactions	A5001

For Laboratory Use.

Description: GoScript™ Reverse Transcriptase utilizes M-MLV and state-of-the-art buffer technology designed for qPCR to deliver robust, reliable cDNA synthesis of a full range of rare and abundant transcripts, even in the presence of inhibitors. GoScript™ Reverse Transcriptase is qualified for use in qPCR, including GoTaq® qPCR and Plexor® qPCR systems for performing RT-qPCR.

Features:

- **Ultra-Active:** Save money on every reaction.
- **Sensitive:** Detect rare transcripts.
- **Processive:** Transcribe long messages.
- **Resilient:** Synthesize cDNA in the presence of strong inhibitors.
- Replaces Superscript® I, II, III and other RTs.

Protocol	Part#
Technical Manual	TM316

Storage Conditions: Store at –20°C.

AMV Reverse Transcriptase

Product	Size	Conc.	Cat.#
AMV Reverse Transcriptase	300 u	10 u/µl	M5101
	1,000 u	10 u/µl	M5108
AMV Reverse Transcriptase (HC)	600 u	20–25 u/µl	M9004
Available Separately	**Size**	**Conc.**	**Cat.#**
Reverse Transcription 10X Buffer	1.4 ml		A3561
Magnesium Chloride Solution	1.5 ml	25 mM	A3511
Set of dATP, dCTP, dGTP, dTTP	40µmol each	100 mM	U1240

For Laboratory Use.

Description: Avian Myeloblastosis Virus Reverse Transcriptase (AMV RT) catalyzes the polymerization of DNA using template DNA, RNA or RNA:DNA hybrids. It requires a primer (DNA primers are more efficient than RNA primers) as well as Mg^{2+} or Mn^{2+}. The enzyme possesses an intrinsic RNase H activity. Both nonionic detergents and sulfhydryl compounds stabilize the enzyme activity in vitro.

Features:

- **Available at High Concentration:** Cat.# M9004 contains 600 units of AMV RT at 20–25u/µl.
- **Provided with 5X Reaction Buffer:** 250mM Tris-HCl (pH 8.3 at 25°C), 250mM KCl, 50mM $MgCl_2$, 2.5mM spermidine, 50mM DTT.
- **Temperature Stability:** AMV RT is the preferred reverse transcriptase for templates with high secondary structure due to its stability at higher reaction temperatures (37–58°C).

Protocol	Part#
Promega Product Information	9PIM510

Storage Conditions: Store at –20°C.

ImProm-II™ Reverse Transcriptase

Product	Size	Cat.#
ImProm-II™ Reverse Transcriptase	10 reactions	A3801
	100 reactions	A3802
	500 reactions	A3803
Available Separately	**Size**	**Cat.#**
ImProm-II™ Reverse Transcription System	100 reactions	A3800

For Laboratory Use.

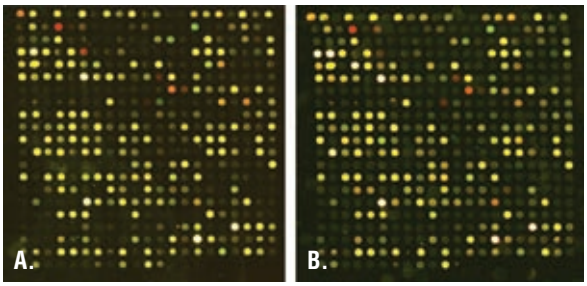
Description: ImProm-II™ Reverse Transcriptase enables robust, full-length cDNA synthesis for the reproducible analysis of rare or long messages. The ImProm-II™ Reverse Transcriptase can be used to reverse transcribe total RNA, poly(A)+ mRNA or synthetic transcript RNA templates.

Features:

- **Full-Length RT-PCR:** Reverse transcribe RNA templates up to 8.9kb.
- **Microarray-Compatible:** Incorporate regular and Cy®3- and Cy®5-modified nucleotides.
- **Scalable and Flexible:** Reaction volumes of 1–20µl can be used in subsequent PCR, and the optimized buffer also allows for coupled RT-PCR.
- **Provided with 5X Reaction Buffer:** 250mM Tris-HCl (pH 8.3 at 25°C), 375mM KCl and 50mM DTT. A 25mM $MgCl_2$ Solution is also included.
- **Versatile:** Use with your thermostable polymerase of choice for two-step RT-PCR.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Protocol	Part#
Technical Manual	TM236

Storage Conditions: Store at –20°C.



A comparison of typical 21-by-21 feature array blocks hybridized with ImProm-II™- or SuperScript®-II-generated fluorescent cDNA probes. **Panel A.** Array hybridized with ImProm-II™-generated fluorescent cDNA probes. **Panel B.** Array hybridized with SuperScript®-II-generated fluorescent cDNA probes. Image brightness was normalized to yield similar background intensities. See Kasler *et al. Promega Notes* **81**, 14–15, for experimental details.

M-MLV Reverse Transcriptase

Product	Size	Conc.	Cat.#
M-MLV Reverse Transcriptase	10,000 u	200 u/µl	M1701
	50,000 u	200 u/µl	M1705
M-MLV Reverse Transcriptase Buffer Pack	2 × 1 ml		M5313
Available Separately	**Size**	**Conc.**	**Cat.#**
Oligo(dT)$_{15}$ Primer	20 µg		C1101
Random Primers	20 µg		C1181
Set of dATP, dCTP, dGTP, dTTP	40µmol each	100 mM	U1240

Cat.# M1701, M1705, M5313, C1181, U1240 For Laboratory Use. Product may not be available in all countries. Please contact your local representative for more information.

Description: Moloney Murine Leukemia Virus Reverse Transcriptase (M-MLV RT) is an RNA-dependent DNA polymerase that can be used in cDNA synthesis with long messenger RNA templates (>5kb). The enzyme is a product of the *pol* gene of M-MLV and consists of a single subunit with a molecular weight of 71kDa. The RNase H activity of M-MLV RT is weaker than the commonly used Avian Myeloblastosis Virus (AMV) reverse transcriptase.

Features:

- **Provided with 5X Reaction Buffer:** 250mM Tris-HCl (pH 8.3 at 25°C), 375mM KCl, 15mM $MgCl_2$, 50mM DTT.
- **May Be Heat-Inactivated:** M-MLV RT is inactivated by heating at 70°C for 10 minutes.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Protocol	Part#
Promega Product Information	9PIM170

Storage Conditions: Store at –20°C.

M-MLV Reverse Transcriptase, RNase H Minus

Product	Size	Conc.	Cat.#
M-MLV Reverse Transcriptase, RNase H Minus	10,000 u	100–200 u/µl	M5301

Product may not be available in all countries. Please contact your local representative for more information.

Description: Moloney Murine Leukemia Virus Reverse Transcriptase, RNase H Minus (M-MLV RT [H–]), is an RNA-dependent DNA polymerase that can be used in cDNA synthesis with long messenger RNA templates (>5kb). This is a form of M-MLV Reverse Transcriptase that has been genetically altered to remove the associated RNase H activity. Although many researchers are successful in using M-MLV RT (H+) for analytical and some preparative cDNA applications, reverse transcriptases lacking RNase H activity provide another option for the preparation of long cDNAs and libraries containing a high percentage of full-length cDNA.

Features:

- **RNase H Minus:** Provides optimal conditions for the preparation of full-length cDNA from long RNA templates.
- **Provided with 5X Reaction Buffer:** 250mM Tris-HCl (pH 8.3 at 25°C), 375mM KCl, 15mM $MgCl_2$, 50mM DTT.
- **May Be Heat-Inactivated:** M-MLV RT is inactivated by heating at 70°C for 10 minutes.

Protocol	Part#
Promega Product Information	9PIM530

Storage Conditions: Store at –20°C.

M-MLV Reverse Transcriptase, RNase H Minus, Point Mutant

Product	Size	Cat.#
M-MLV Reverse Transcriptase, RNase H Minus, Point Mutant	2,500 u	M3681
	10,000 u	M3682
	50,000 u	M3683

Products may not be available in all countries. Please contact your local representative for more information.

Description: Moloney Murine Leukemia Virus Reverse Transcriptase, RNase H Minus (M-MLV RT [H–]), Point Mutant, is an RNA-dependent DNA polymerase that can be used in cDNA synthesis with long RNA templates (>5kb). The lack of RNase H activity is beneficial for this application, as RNase H can start to degrade templates when incubation times are long. Although many researchers are successful in using M-MLV RT (H+) for analytical and some preparative cDNA applications, reverse transcriptases lacking RNase H activity provide another option for the preparation of long cDNAs and for libraries containing a high percentage of full-length cDNA.

Features:

- **RNase H Minus:** Provides optimal conditions for the preparation of full-length cDNA from long RNA templates.
- **Temperature Stability:** Thermostability of this point mutant prevents problems associated with secondary structure.
- **Increased Polymerase Activity:** M-MLV RT (H–), Point Mutant, gives higher yields of cDNA compared with the deletion mutant (Cat.# M5301).
- **Provided with 5X Reaction Buffer:** 250mM Tris-HCl (pH 8.3 at 25°C), 375mM KCl, 15mM $MgCl_2$, 50mM DTT.
- **Broad Working Range:** More tolerance to variations in enzyme and substrate concentration means improved consistency in performance.

Protocol	Part#
Promega Product Information	9PIM368

Storage Conditions: Store at –20°C.

T4 DNA Ligase

Product	Size	Conc.	Cat.#
T4 DNA Ligase	100 u (Weiss units)	1–3 u/µl	M1801
	500 u (Weiss units)	1–3 u/µl	M1804
T4 DNA Ligase (HC)	500 u (Weiss units)	10–20 u/µl	M1794
Available Separately		**Size**	**Cat.#**
T4 DNA Ligase Buffer Pack		1.5ml (3 × 500 µl)	C1263

For Laboratory Use.

Description: T4 DNA Ligase catalyzes the joining of two strands of DNA between the 5′-phosphate and the 3′-hydroxyl groups of adjacent nucleotides in either a cohesive-ended or blunt-ended configuration. The enzyme has also been shown to catalyze the joining of RNA to either a DNA or RNA strand in a duplex molecule but will not join single-stranded nucleic acids.

Features:

- **Available at High Concentration:** Cat.# M1794 contains 500 units of T4 DNA Ligase at 10–20u/µl.
- **Flexible:** Use with 5′, 3′ or blunt-ended DNA inserts.
- **Provided with 10X Reaction Buffer:** 300mM Tris-HCl (pH 7.8 at 25°C), 100mM $MgCl_2$, 100mM DTT and 10mM ATP.
- **Blue/White Cloning Qualified:** Promega's blue/white cloning assay provides a higher level of quality control for enzymes used in cloning applications.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Protocol	Part#
Promega Product Information	9PIM180

Storage Conditions: Store at –20°C.

LigaFast™ Rapid DNA Ligation System

Product	Size	Cat.#
LigaFast™ Rapid DNA Ligation System	30 reactions	M8221
	150 reactions	M8225
Available Separately	**Size**	**Cat.#**
2X Rapid Ligation Buffer	1.5ml (3 × 500 µl)	C6711

For Laboratory Use.

Description: The LigaFast™ Rapid DNA Ligation System is designed for the efficient ligation of sticky-ended DNA inserts into plasmid vectors in just 5 minutes (blunt-ended inserts in as little as 15 minutes). Rapid ligation is based on the combination of T4 DNA Ligase with a unique 2X Rapid Ligation Buffer. The LigaFast™ System is designed to eliminate any further purification prior to transformation of ligated DNA. The specially formulated 2X Rapid Ligation Buffer requires no additional ATP or Mg^{2+} addition prior to use.

Features:

- **Flexible:** Use with 5′, 3′ or blunt-ended DNA inserts.
- **Fast:** Ligation of cohesive ends in 5 minutes, blunt ends in 15 minutes at room temperature.
- **Convenient:** No requirement to purify ligated DNA prior to heat-shock transformation in *E. coli*. Ligations conducted at room temperature.
- **Ready-To-Use:** No additional buffer modifications required prior to use.
- **Efficient:** Ligations performed using the LigaFast™ System are comparable to standard overnight ligations.
- **Blue/White Cloning Qualified:** Promega's blue/white cloning assay provides a higher level of quality control for enzymes used in cloning applications.

Protocol	Part#
Promega Product Information	9PIM822

Storage Conditions: Store at –20°C.

T4 RNA Ligase

Product	Size	Conc.	Cat.#
T4 RNA Ligase	500 u	10 u/µl	M1051

Description: T4 RNA Ligase catalyzes the ATP-dependent ligation of single-stranded RNA or DNA onto the 5′-phosphoryl termini of single-stranded RNA or DNA. The enzyme, purified from recombinant *E. coli* CA4 (RNase I-deficient), has an apparent molecular weight of 43.5kDa. T4 RNA Ligase also catalyzes the addition of [5′-^{32}P] nucleoside 3′,5′-bis (phosphate) onto single-stranded RNA.

Features:

- **May Be Heat-Inactivated:** T4 RNA Ligase may be inactivated by heating at 65°C for 15 minutes.
- **Provided with 10X Reaction Buffer:** 500mM Tris-HCl (pH 7.8 at 25°C), 100mM $MgCl_2$, 50mM DTT, 10mM ATP.

Protocol	Part#
Promega Product Information	9PIM105

Storage Conditions: Store at –20°C.

T4 Polynucleotide Kinase

Product	Size	Conc.	Cat.#
T4 Polynucleotide Kinase	100 u	5–10 u/µl	M4101
	1,000 u	5–10 u/µl	M4103
T4 PNK Buffer Pack	1.5ml (3 × 500 µl)		C1313

For Laboratory Use.

Description: T4 Polynucleotide Kinase catalyzes the transfer of the γ-phosphate from ATP to the 5′-terminus of polynucleotides or to mononucleotides bearing a 5′-hydroxyl group. The enzyme, purified from recombinant *E. coli*, may be used to phosphorylate RNA, DNA and synthetic oligonucleotides prior to subsequent manipulations such as ligation.

Features:

- **May Be Heat-Inactivated:** T4 Polynucleotide Kinase may be inactivated by heating at 68°C for 10 minutes.
- **Provided with 10X Reaction Buffer:** 700mM Tris-HCl (pH 7.6 at 25°C), 100mM $MgCl_2$, 50mM DTT.
- **Blue/White Cloning Qualified:** Promega's blue/white cloning assay provides a higher level of quality control for enzymes used in cloning applications.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Protocol	Part#
Technical Bulletin	TB519

Storage Conditions: Store at –20°C.

TSAP Thermosensitive Alkaline Phosphatase

Product	Size	Cat.#
TSAP Thermosensitive Alkaline Phosphatase	100 units	M9910

For Laboratory Use. Units listed are MBU (molecular biology units).

Description: TSAP Thermosensitive Alkaline Phosphatase catalyzes the removal of 5′ phosphate groups from DNA, preventing the recircularization and religation of linearized cloning vector DNA during ligation. It is effective on 3′ overhangs, 5′ overhangs and blunt ends. It is also useful for preparing DNA for 5′ end-labeling by removing existing phosphate groups from the 5′ end.

Features:

- **Easy To Use:** TSAP is active in all Promega restriction enzyme buffers, eliminating any cleanup steps or buffer swaps.
- **Heat-Inactivated:** TSAP is irreversibly inactivated by heating at 74°C for 15 minutes. This allows streamlining of the restriction enzyme digestion, dephosphorylation and ligation procedure by eliminating the need for cleanup after alkaline phosphatase treatment.
- **Blue/White Cloning-Qualified:** Promega's blue/white cloning assay provides a higher level of quality control for enzymes used in cloning applications.
- **Provided with Promega MULTI-CORE™ Buffer.**

Protocol	Part#
Promega Product Information	9PIM991

Storage Conditions: Store at –20°C.

Comparison of Alkaline Phosphatases.

	TSAP	CIAP*
Heat Inactivated	Yes	No
Inactivation Temperature	74	N/A
Incubation Time	15 min	2 × 30 min
Special Buffer Required/Recommended	No	Yes
Active in all Promega Restriction Enzyme Buffers	Yes	No
Units required in different RE Buffers	1–2	N/A
Blue/White Cloning-Qualified	Yes	Yes

Only TSAP does not require a special buffer and is active in all Promega restriction enzyme buffers, making it the most convenient and cost-effective choice.

CIAP = Calf Intestinal Alkaline Phosphatase

9493LA

Alkaline Phosphatase, Calf Intestinal (CIAP)

Product	Size	Conc.	Cat.#
Alkaline Phosphatase, Calf Intestinal	1,000 u	1 u/µl	M1821
Alkaline Phosphatase, Calf Intestinal (HC)	1,000 u	20 u/µl	M2825
Available Separately	**Size**		**Cat.#**
CIAP Buffer Pack	1.5ml (3 × 500 µl)		M1833

For Laboratory Use.

Description: Alkaline Phosphatase, Calf Intestinal (CIAP), catalyzes the hydrolysis of 5′-phosphate groups from DNA, RNA, and ribo- and deoxyribonucleoside triphosphates. This enzyme is used to prevent recircularization and religation of linearized cloning vector DNA by removing phosphate groups from both 5′-termini and may also be used for the dephosphorylation of 5′ phosphorylated ends of DNA or RNA for subsequent labeling with [^{32}P]ATP and T4 Polynucleotide Kinase. CIAP is active on 5′ overhangs, 5′ recessed and blunt ends.

Features:

- **Available at High Concentration:** Cat.# M2825 contains 1,000 units of CIAP at 20u/µl.
- **Blue/White Cloning Qualified:** Promega's blue/white cloning assay provides a higher level of quality control for enzymes used in cloning applications.
- **Provided with 10X Reaction Buffer:** 0.5M Tris-HCl (pH 9.3 at 25°C), 10mM $MgCl_2$, 1mM $ZnCl_2$, 10mM spermidine.

Protocol	Part#
Promega Product Information	9PIM182

Storage Conditions: Store at –20°C.

Exonuclease III

Product	Size	Conc.	Cat.#
Exonuclease III	5,000 u	150–200 u/µl	M1811
	25,000 u	150–200 u/µl	M1815
For Laboratory Use.			

Description: Exonuclease III is a 3′→5′ exonuclease specific for double-stranded DNA. The enzyme catalyzes the stepwise removal of mononucleotides starting from a 3′-OH at nicks, blunt ends, recessed ends and 3′-overhangs of less than 4 bases, yielding nucleoside 5′-phosphates. Exonuclease III will also degrade DNA from 3′-phosphate ends due to its intrinsic 3′-phosphatase activity. In addition, the enzyme has apurinic endonuclease activity and ribonuclease H activity. Exonuclease III is used in conjunction with S1 nuclease for unidirectional deletion of sequences from the termini of DNA fragments.

Features:

- **Flexible:** Control deletion rate by varying incubation temperature.
- **May Be Heat-Inactivated:** Exonuclease III may be inactivated by heating to 75°C for 10 minutes.
- **Provided with 10X Reaction Buffer:** 660mM Tris-HCl (pH 8.0 at 25°C), 6.6mM $MgCl_2$.

Protocol	Part#
Promega Product Information	9PIM181

Storage Conditions: Store at –20°C.

Mung Bean Nuclease

Product	Size	Conc.	Cat.#
Mung Bean Nuclease	2,000 u	50–100 u/µl	M4311

Description: Mung Bean Nuclease catalyzes the degradation of single-stranded DNA and RNA endonucleolytically to yield 5′-phosphoryl-terminated products. While the nuclease prefers ssDNA over dsDNA by 30,000-fold, at very high concentrations the enzyme degrades double-stranded DNA from both ends. Mung Bean Nuclease has been used for transcript mapping studies, for flushing staggered ends and for the separation of cDNA strands after synthesis with reverse transcriptase and DNA Polymerase I.

Features:

- **Provided with 10X Reaction Buffer:** 300mM sodium acetate (pH 5.0 at 15°C), 500mM NaCl, 10mM $ZnCl_2$.

Protocol	Part#
Promega Product Information	9PIM431

Storage Conditions: Store at –20°C.

S1 Nuclease

Product	Size	Conc.	Cat.#
S1 Nuclease	10,000 u	20–100 u/µl	M5761

Description: S1 Nuclease degrades single-stranded DNA and RNA endonucleolytically to yield 5′-phosphoryl-terminated products. Double-stranded nucleic acids (DNA:DNA, DNA:RNA or RNA:RNA) are resistant to degradation except with extremely high concentrations of enzyme. The enzyme is used to remove single-stranded termini from double-stranded DNA, for selective cleavage of single-stranded DNA and for mapping RNA transcripts.

Features:

- **Provided with 10X Reaction Buffer:** 0.5M sodium acetate (pH 4.5 at 25°C), 2.8M NaCl, 45mM $ZnSO_4$.

Storage Conditions: Store at –20°C.

RQ1 RNase-Free DNase

Product	Size	Conc.	Cat.#
RQ1 RNase-Free DNase	1,000 u	1 u/µl	M6101
For Laboratory Use.			

Description: RQ1 RNase-Free DNase is a preparation of deoxyribonuclease I that degrades single-stranded or double-stranded DNA to produce 3′-hydroxyl oligonucleotides. This preparation is qualified for use in applications where maintaining the integrity of RNA is critical.

Features:

- **Convenient:** 10X Reaction Buffer (400mM Tris-HCl [pH 8.0 at 25°C], 100mM $MgSO_4$, 10mM $CaCl_2$) and Stop Buffer (20mM EGTA [pH 8.0 at 25°C]) are provided.

Protocol	Part#
Promega Product Information	9PIM610

Storage Conditions: Store at –20°C.

Ribonuclease H

Product	Size	Conc.	Cat.#
Ribonuclease H	50 u	0.5–2 u/µl	M4281
	250 u	0.5–2 u/µl	M4285
For Laboratory Use.			

Description: Ribonuclease H (RNase H) is an endonuclease that specifically hydrolyzes the phosphodiester bonds of RNA hybridized to DNA to produce 3′-OH and 5′-P-terminated products. It will not degrade single-stranded nucleic acids, double-stranded DNA or double-stranded RNA.

Storage Conditions: Store at –20°C.

RNase ONE™ Ribonuclease

Product	Size	Conc.	Cat.#
RNase ONE™ Ribonuclease	1,000 u	5–10 u/µl	M4261
	5,000 u	5–10 u/µl	M4265

For Laboratory Use.

Description: RNase ONE™ Ribonuclease is a 27kDa periplasmic enzyme from *E. coli* that catalyzes the degradation of RNA to cyclic nucleotide monophosphate (NMP) intermediates. Slower hydrolysis further catalyzes the degradation of these intermediates to 3′-NMPs. RNase ONE™ Ribonuclease is one of the few known RNases that can cleave a phosphodiester bond between any two ribonucleotides. RNase ONE™ Ribonuclease may be used to remove RNA from DNA preparations, for RNase protection assays and for mapping or quantitation of RNA by selective cleavage of single-stranded regions.

Features:

- **Flexible:** RNase ONE™ Ribonuclease has the ability to cleave phosphodiester bonds between any two ribonucleotides.
- **Provided with 10X Reaction Buffer:** 100mM Tris-HCl (pH 7.5 at 25°C), 50mM EDTA, 2M sodium acetate.

Storage Conditions: Store at –20°C. **Do not freeze at –70°C. Do not store on dry ice.**

Agar*ACE*™ Enzyme

Product	Size	Conc.	Cat.#
Agar*ACE*™ Enzyme	25 u	0.15–0.30 u/µl	M1741
	500 u	0.15–0.30 u/µl	M1743

For Laboratory Use.

Description: Agar*ACE*™ Enzyme is a unique agarose-digesting enzyme developed by Promega for simple and quantitative recovery of intact DNA or RNA from agarose gels. The enzyme is sufficiently thermostable that low-melting-point (LMP) agarose melted at 65–75°C does not have to be equilibrated to the reaction temperature before hydrolysis. Simply place the tube containing the molten agarose at 42–47°C and add Agar*ACE*™ Enzyme. With slight modifications to the protocol, Agar*ACE*™ Enzyme can also effectively hydrolyze non-LMP agaroses, although the higher temperatures required to melt them may be deleterious to the DNA or RNA. Finally, Agar*ACE*™ Enzyme does not require buffer exchange in order to exhibit high agarose hydrolytic activity; its activity does not appreciably change in TAE or TBE buffers used in normal electrophoretic procedures.

Features:

- **Fast:** Hydrolyzes 200mg of 1% LMP agarose in 15 minutes.
- **Convenient:** No buffer equilibration step required.
- **Performance Tested:** Nucleic acids recovered using Agar*ACE*™ Enzyme are ready for direct use in a number of procedures such as ligation, random primer labeling, PCR and sequencing.

Protocol	Part#
Technical Bulletin	TB228

Storage Conditions: Store at –20°C.

Single-Stranded DNA Binding Protein

Product	Size	Conc.	Cat.#
Single-Stranded DNA Binding Protein	100 µg	1–5 µg/µl	M3011

Description: *E. coli* Single-Stranded DNA Binding Protein (SSB) consists of four identical 18.9kDa subunits. It binds with high affinity in a cooperative manner to single-stranded DNA but does not bind well to double-stranded DNA. It is involved in DNA replication and in recombination in vivo.

Storage Conditions: Store at –20°C.

Topoisomerase I

Product	Size	Conc.	Cat.#
Topoisomerase I	200 u	2–10 u/µl	M2851

Description: Topoisomerase I, isolated from wheat germ, is an enzyme capable of removing negative supercoils from covalently closed circular DNA.

Storage Conditions: Store at –70°C.

RNasin® Plus RNase Inhibitor

Product	Size	Conc.	Cat.#
RNasin® Plus RNase Inhibitor	2,500 u	40 u/µl	N2611
	10,000 u	40 u/µl	N2615

For Laboratory Use.

Description: RNasin® Plus RNase Inhibitor is a recombinant mammalian RNase inhibitor that is expressed as a soluble protein in *E. coli*, allowing easy purification through a combination of ion exchange and hydrophobic interaction chromatography. The protein is capable of inhibiting eukaryotic RNases (e.g., RNase A and RNase B) similarly to human placental RNase inhibitor. RNasin® Plus RNase Inhibitor is tested in RT-PCR and is compatible with enzymes such as AMV, M-MLV and ImProm-II™ Reverse Transcriptases or *Taq* and *Tfl* DNA Polymerases. RNasin® Plus RNase Inhibitor also is tested and compatible with quantitative, real-time RT-PCR in a TaqMan® assay.

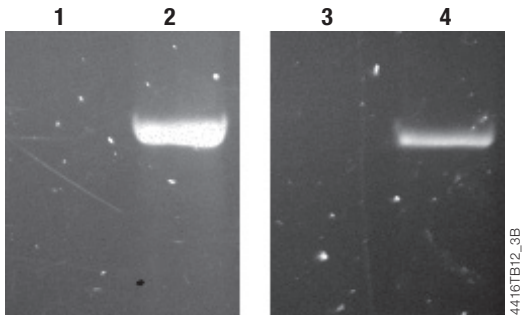
The inhibitor offers increased resistance to oxidation over the human version of the protein. Two cysteines in the human protein have been identified as especially sensitive to oxidation and react by forming a disulfide bond that can block the active site of the inhibitor. RNasin® Plus, through natural amino acid diversity, lacks the ability to form this site-blocking disulfide. In addition, the new protein has characteristics never before realized, including continued inhibition of RNases above 50°C. Heating solutions of RNasin® Plus and RNase followed by cooling does not result in the reappearance of RNase activity—even when the solution is heated above the denaturation temperature of the RNasin® Plus protein alone. This allows RNasin® Plus to protect RNA species prior to, during and after heating, even at temperatures normally used during first-strand DNA synthesis in RT-PCR. We have taken solutions up to 70°C for 15 minutes and did not see RNase reactivation.

Features:

- **Improved Resistance to Oxidation:** Due to natural amino acid diversity, RNasin® Plus lacks the capability to form the active site-blocking disulfide bond that can form in the human protein under oxidative conditions.
- **Improved Purification:** RNasin® Plus is expressed by *E. coli* as a soluble protein, allowing easy purification by a combination of ion exchange and hydrophobic interaction chromatography. No direct affinity chromatography required. The new process yields a >90% pure protein with no *E. coli* RNase carryover.
- **Proven Compatibility with RT-PCR Systems:** RNasin® Plus has proven compatible with the Access and AccessQuick™ RT-PCR Systems, ImProm-II™ Reverse Transcription System and the Reverse Transcription System. Also proven compatible with TaqMan®-based RT-PCR Systems.
- **Protection During RNA Template Denaturation:** Heating mixtures of RNasin® Plus and RNase does not lead to reactivation of the RNase at temperatures even as high as 70°C for 15 minutes. Many RT-PCR protocols call for RNA template denaturation (e.g., 65–70°C for 5–10 minutes) in the presence of the RT primers prior to full RT reaction assembly for maximum sensitivity. You can now include RNasin® Plus at this step.
- **Protection During Higher Temperature RT Reactions:** Add RNasin® Plus during RT reaction assembly and take the reaction to temperatures above 50°C with enzymes like the ImProm-II™ and AMV Reverse Transcriptases. RNases that may be present will not be reactivated at the higher temperature.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Protocol	Part#
Promega Product Information	9PIN261

Storage Conditions: Store at –20°C.



Protection from RNase at 70°C. Separate tubes of RNasin® Plus and RNase (lanes 1 and 3) were heated to 70°C for 15 minutes. RNasin® Plus and RNase were combined and then heated to 70°C for 15 minutes (lanes 2 and 4). To each set of reactions, either 100ng (lanes 1 and 2) or 10ng (lanes 3 and 4) of Luciferase Control RNA (Cat.# L4561) were added. The reactions were held at 37°C for 1 hour, then used in an RT-PCR to amplify the entire 1.8kb transcript. The gel shows the amplified product from the RT-PCR. All lanes used 400u of RNasin® Plus and 1.25µg of a rat liver protein extract (abundant source of RNase; Sigma Cat.# L-1380) dissolved in water to 0.5µg/µl.

RNasin® Ribonuclease Inhibitor

Product	Size	Conc.	Cat.#
Recombinant RNasin® Ribonuclease Inhibitor	2,500 u	20–40 u/µl	N2511
	10,000 u	20–40 u/µl	N2515
RNasin® Ribonuclease Inhibitor	2,500 u	20–40 u/µl	N2111
	10,000 u	20–40 u/µl	N2115

Cat.# N2511, N2515 For Laboratory Use.

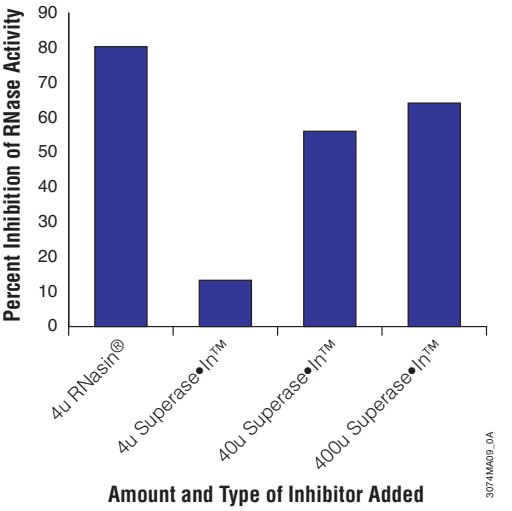
Description: Natural and Recombinant RNasin® Ribonuclease Inhibitors have broad-spectrum RNase inhibitory properties, including the inhibition of eukaryotic RNases of the neutral type. The 50kDa protein exerts its inhibitory effect by noncovalently binding to RNases in a 1:1 ratio. The K_i value for the binding of RNasin® Ribonuclease Inhibitor to RNase (e.g., RNase A) is approximately 10^{-14}M. Promega offers two different preparations: Recombinant RNasin® Ribonuclease Inhibitor and Natural RNasin® Ribonuclease Inhibitor. Both products are purified using a combination of ion exchange and affinity chromatography.

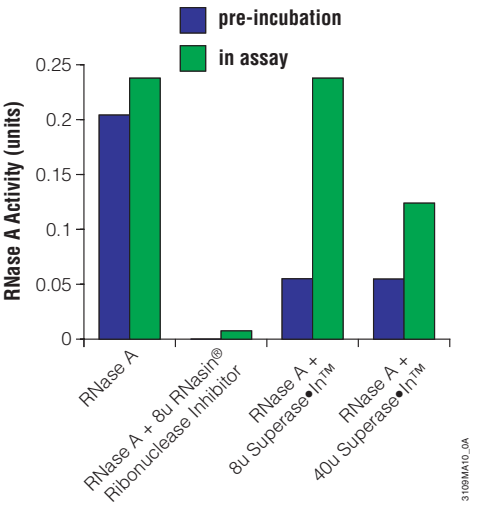
Features:

- **Inhibits Common Eukaryotic RNases:** RNasin® Ribonuclease Inhibitor has broad-spectrum RNase inhibitory properties, including RNase A, RNase B, RNase C and human placental RNase. Does not inhibit RNase T1, S1 nuclease, RNase from *Aspergillus*, RNase H, RNase ONE™ Ribonuclease.
- **Compatibility:** RNasin® Ribonuclease Inhibitor does not inhibit SP6, T7 or T3 RNA Polymerase; ImProm-II™, AMV or M-MLV Reverse Transcriptase; or *Taq* DNA polymerase.
- **Broad pH Range:** Active over a broad pH range (pH 5–8).
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Protocol	Part#
Recombinant RNasin® Product Information	9PIN251
RNasin® Product Information	9PIN211

Storage Conditions: Store at –20°C.





Comparison of RNasin® Ribonuclease Inhibitor and Superase•In™ inhibition of RNase A activity. Panel A. Total yeast RNA assay. Total yeast RNA was incubated in the presence of 5ng RNase A for 5 minutes at 37°C in 0.5ml of reaction mix containing 50mM MOPS and 5mM $MgCl_2$ (pH 6.5). The indicated amounts of inhibitor (RNasin® or Superase•In™) were mixed with the RNA prior to RNase addition. After incubation, 0.5ml 10% TCA was added to stop the reaction and to precipitate the large RNA molecules. An OD_{280} measurement was taken of the TCA-soluble material. **Panel B**. "Pre-incubation" and "in assay" conditions. The total yeast RNA assay was performed as described in Panel A along with an experimental modification of "pre-incubation." For the pre-incubation assay, the ribonuclease inhibitors were mixed with RNase and incubated for 15 minutes at 22°C. The pre-incubation mix was then added to the RNA.

Sequencing, Mutagenesis and Labeling

DNA Sequencing Systems	**264**
Sequencing Primers	**264**
Mutagenesis Systems	**265**
Nested Deletion Mutagenesis Systems	**267**
Nucleic Acid Labeling Systems	**267**

Automatic Processor Compatible (APC) Film

Product	Size	Cat.#
Automatic Processor Compatible (APC) Film	25 sheets	Q4411
Automatic Processor Compatible (APC) Film, Sample Size	6 sheets	Q4412

Description: The SILVER SEQUENCE™ Automatic Processor Compatible (APC) Film provides the means to capture enhanced images and permanent copies of SILVER SEQUENCE™ results. The film is exposed using fluorescent light from a standard light box. Films are easily developed using typical darkroom reagents; development may be performed manually or by using an automatic film processor. Size of the film is 30 × 40cm.

Protocol	Part#
Technical Manual	TM023

RNA Polymerase Promoter Sequencing Primers

Product	Size	Conc.	Cat.#
SP6 Promoter Primer	2 µg	10 µg/ml	Q5011
T7 Promoter Primer	2 µg	10 µg/ml	Q5021
T7 EEV Promoter Primer	2 µg	10 µg/ml	Q6700

Description: The SP6 and T7 Promoter Primers are designed for sequencing inserts cloned into the pGEM® Vectors. The SP6 Promoter Primer is designed for sequencing inserts cloned into the pALTER®-MAX and pCI-neo Vectors. The primers are designed to be annealed to single-stranded DNA or, after alkaline denaturation, to double-stranded DNA. The promoter primers are purified by gel electrophoresis or HPLC. The T7 EEV Promoter Primer is suitable for sequencing the pALTER®-MAX, pCMVTNT™ , pTNT™ and phMGFP Vectors, and the pCI/pSI series of mammalian expression vectors.

Primer Sequences
SP6: 5′-d(TATTTAGGTGACACTATAG)-3′
T7: 5′-d(TAATACGACTCACTATAGGG)-3′
T7 EEV: 5′-d(AAGGCTAGAGTACTTAATACGA)-3′

Storage Conditions: Store at –20°C.

pUC/M13 Sequencing Primers

Product	Size	Conc.	Cat.#
pUC/M13 Primer, Forward (17mer)	2 µg	10 µg/ml	Q5391
pUC/M13 Primer, Reverse (17mer)	2 µg	10 µg/ml	Q5401
pUC/M13 Primer, Reverse (22mer)	2 µg	10 µg/ml	Q5421
pUC/M13 Primer, Forward (24mer)	2 µg	10 µg/ml	Q5601

Description: The pUC/M13 Primers are designed for sequencing inserts cloned into the M13 vectors and pUC plasmids developed by Messing. These primers also can be used for sequencing other *lacZ*-containing plasmids such as the pGEM®-Z and pGEM®-Zf Vectors. The primers are purified by gel electrophoresis or HPLC.

Primer Sequences
Forward (17mer): 5′-d(GTTTTCCCAGTCACGAC)-3′
Reverse (17mer): 5′-d(CAGGAAACAGCTATGAC)-3′
Reverse (22mer): 5′-d(TCACACAGGAAACAGCTATGAC)-3′
Forward (24mer): 5′-d(CGCCAGGGTTTTCCCAGTCACGAC)-3′

Storage Conditions: Store at –20°C. The primers are supplied in sterile water.

pTARGET™ Sequencing Primer

Product	Size	Cat.#
pTARGET™ Sequencing Primer	2 µg	Q4461

Description: The pTARGET™ Sequencing Primer is designed for sequencing inserts cloned into the pTARGET™ Mammalian Expression Vector (Cat.# A1410). This sequencing primer hybridizes to the region of the *lacZ* gene from nucleotides 1367–1344 on the pTARGET™ Vector.

This primer can be used **only** for sequencing inserts in the pTARGET™ Vector. The primer sequence is **not** a binding site for any RNA polymerases and **cannot** be used to generate in vitro transcripts.

The sequence of the pTARGET™ Sequencing Primer is 5′-d(TTACGCCAAGTTATTTAGGTGACA)-3′. It is supplied at a concentration of 10ng/µl (1.25pmol/µl) in sterile water.

Storage Conditions: Store at –20°C.

PinPoint™ Vector Sequencing Primer

Product	Size	Cat.#
PinPoint™ Vector Sequencing Primer	2 µg	V4211
	2 µg	V4211

Description: The PinPoint™ Vector Sequencing Primer is designed for sequencing inserts cloned into the PinPoint™ Xa Vectors (components of Cat.# V2020). The primer hybridizes upstream of the Factor Xa site at nucleotides 325–343, approximately 40–50 base pairs upstream of the multiple cloning region and can be used to determine if an insert is cloned in-frame with the biotinylation purification tag of the PinPoint™ Xa Vectors. The sequence of the PinPoint™ Vector Sequencing Primer is 5′-d(CGTGACGCGGTGCAGGGCG)-3′. It is supplied dried.

Features:

- **Performance Tested:** The PinPoint™ Vector Sequencing Primer is tested in double-stranded sequencing reactions with circular PinPoint™ Vectors.

Storage Conditions: Store at –20°C.

GeneEditor™ in vitro Site-Directed Mutagenesis System

Product	Size	Cat.#
GeneEditor™ in vitro Site-Directed Mutagenesis System	30 reactions	Q9280

Description: The GeneEditor™ in vitro Site-Directed Mutagenesis System is a high-efficiency system for the generation and selection of oligonucleotide-directed mutations. This system uses antibiotic selection to obtain a high frequency of mutants. Selection Oligonucleotides provided with the GeneEditor™ System encode mutations that alter the ampicillin resistance gene, creating a new additional resistance to the GeneEditor™ Antibiotic Selection Mix. In the GeneEditor™ System protocol, the Selection Oligonucleotide is annealed to a single- or double-stranded DNA template at the same time as a mutagenic oligonucleotide. Subsequent synthesis and ligation of the mutant strand links the two oligonucleotides. The resistance to the GeneEditor™ Antibiotic Selection Mix encoded by this mutant DNA strand facilitates selection of the desired mutation. The system will work with any cloning vector that contains ampicillin resistance as a selectable marker. Mutants generated using this system retain ampicillin resistance and gain resistance to the GeneEditor™ Antibiotic Selection Mix.

Features:

- **Efficient:** High-efficiency mutagenesis. 700bp deletions have been made with over 60% efficiency.
- **Convenient:** All reagents required are provided including competent cells. In addition, no single-stranded DNA production is required.
- **High Fidelity:** T4 DNA Polymerase is used rather than a lower fidelity thermostable DNA polymerase for less risk of secondary undesirable mutations.

Protocol	Part#
Technical Manual	TM047

Storage Conditions: Store competent cells at –70°C; store other components at –20°C.

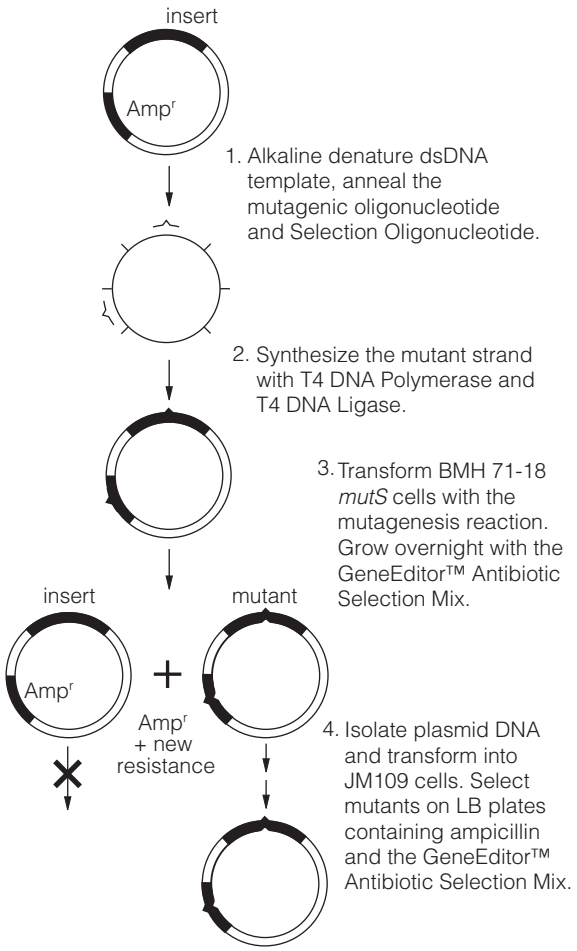
GeneEditor™ Antibiotic Selection Mix

Product	Size	Cat.#
GeneEditor™ Antibiotic Selection Mix	20 ml	Q9291

Description: The GeneEditor™ Antibiotic Selection Mix is used for positive selection of mutants in conjunction with the GeneEditor™ in vitro Site-Directed Mutagenesis System. The mix contains a formulation of selected antibiotics. Only mutants created with the GeneEditor™ System will grow in the presence of the Selection Mix.

Protocol	Part#
Technical Manual	TM047

Storage Conditions: Store at –20°C.



Schematic diagram of the GeneEditor™ in vitro Site-Directed Mutagenesis procedure.

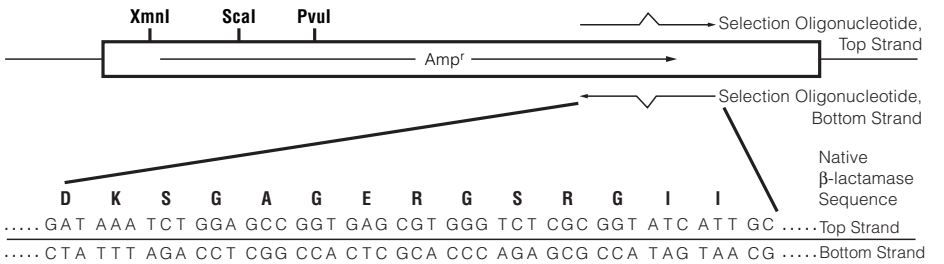
GeneEditor™ Selection Oligonucleotides

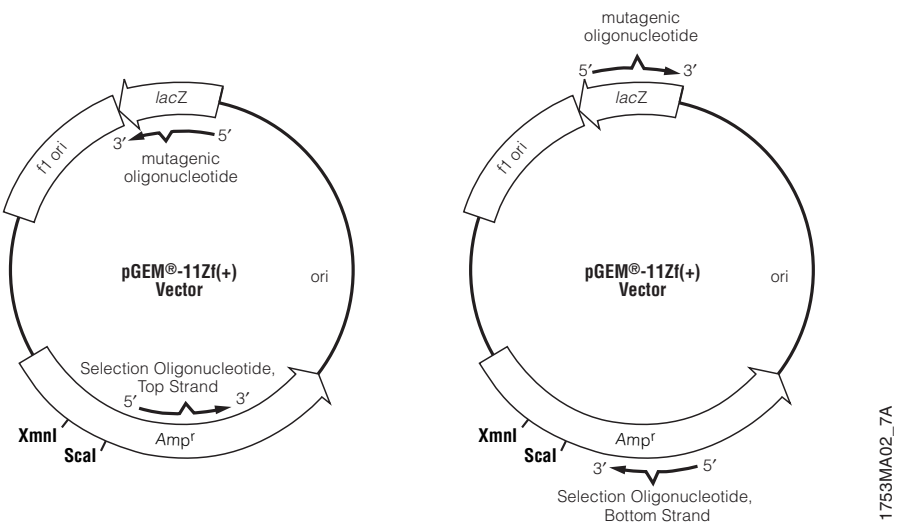
Product	Size	Cat.#
Bottom Strand Selection Oligonucleotide	35 µl	Q9301
Top Strand Selection Oligonucleotide	35 µl	Q9321

Protocol	Part#
Technical Manual	TM047

Storage Conditions: Store at –20°C.

Description: The **Top** or **Bottom** Strand Selection Oligonucleotide (35mer) is used in conjunction with the GeneEditor™ in vitro Site-Directed Mutagenesis System to change the resistance of the ampicillin gene to resistance to the GeneEditor™ Antibiotic Selection Mix. The GeneEditor™ in vitro Site-Directed Mutagenesis System is designed for use with vectors containing ampicillin resistance as a selectable marker.





Hybridization location of selection oligonucleotides. The location and orientation of the Top and Bottom Strand Selection Oligonucleotides is shown relative to the ampicillin resistance gene in the pGEM®-11Zf(+) Vector. The direction of the ampicillin resistance gene in any vector can be determined by the location of the unique restriction sites XmnI, ScaI and PvuI at the 5'-end. The Top Strand Selection Oligonucleotide hybridizes to the bottom strand, covering the sequence indicated. To determine which Selection Oligonucleotide to use, determine which strand is encoded by your mutagenic oligonucleotide and use the corresponding Selection Oligonucleotide. It is essential that both selection and mutagenic oligonucleotides hybridize to the same strand. The example shown indicates the proper oligonucleotide orientation for introducing a mutation into the *lac*Z α-peptide of the pGEM®-11Zf(+) Vector provided with the system.

Erase-a-Base® System

Product	Size	Cat.#
Erase-a-Base® System (minus vectors & bacterial strain)	1 system	E5750

Description: The Erase-a-Base® System is designed for the rapid construction of plasmid or M13 subclones containing progressive unidirectional deletions of any inserted DNA. The system is based on the procedure developed by Henikoff, in which exonuclease III (Exo III) is used to specifically digest insert DNA from a 5′ protruding or blunt end restriction site. The adjacent sequencing primer binding site is protected from digestion by a 4-base 3′ overhang restriction site or by an α-phosphorothioate-filled end.

Features:

- **Rapid:** Construction of nested deletions from plasmid or M13 clones are rapid. Construction is complete in a few hours.
- **Efficient:** Produce deletion sets spanning several kilobases.

Protocol	Part#
Technical Manual	TM006

Storage Conditions: Store at –20°C.

DNA 5′ End-Labeling System

Product	Size	Cat.#
DNA 5′ End-Labeling System	10 reactions	U2010
For Laboratory Use.		

Description: The DNA 5′ End-Labeling System is a complete system for phosphorylating both double- and single-stranded DNA and RNA with T4 Polynucleotide Kinase and [γ-^{32}P]ATP. The system includes enzymes, buffers and control DNA standards to measure reaction efficiencies. Calf Intestinal Alkaline Phosphatase is included for removal of the 5′-phosphate prior to labeling with T4 Polynucleotide Kinase.

Features:

- **Convenient:** Can use to label both single-stranded and double-stranded DNA and RNA.
- **Complete:** System includes enzymes, buffers and control DNA standards for measuring reaction efficiencies (except radionucleotides).
- **Flexible:** Works with [γ-^{32}P]ATP, [γ-^{33}P]ATP or [γ-^{35}S]ATP.

Protocol	Part#
Technical Bulletin	TB096

Storage Conditions: Store at –20°C.

Prime-a-Gene® Labeling System

Product	Size	Cat.#
Prime-a-Gene® Labeling System	30 reactions	U1100
Available Separately	**Size**	**Cat.#**
Nuclease-Free Water	150 ml	P1195
Labeling 5X Buffer	300 µl	U1151
For Laboratory Use.		

Description: The Prime-a-Gene® Labeling System provides a complete set of complementary reagents, including Labeling 5X Buffer that contains random synthetic hexadeoxynucleotide primers for random-primed labeling of linear template DNA with radionucleotides. As little as 25ng of input DNA can be used to generate probes with specific activities >1 × 10^{9}cpm/µg.

Features:

- **Ready to Use:** Includes reagents needed for random-primed labeling of linear DNA, including random synthetic hexadeoxynucleotide primers (excluding radionucleotides).
- **High Specific Activity:** Probes with specific activities >1 × 10^{9}cpm/µg can be generated.

Protocol	Part#
Technical Bulletin	TB049

Storage Conditions: Store at –20°C.

Vectors, Competent Cells and Cloning Tools

Nucleic Acids	**270**
Cloning and Transcription Vectors	**271**
Competent Cells	**276**
Bacterial Strains	**277**
Helper Phage	**278**

Herring Sperm DNA

Product	Size	Conc.	Cat.#
Herring Sperm DNA	10 mg	10 µg/µl	D1811
	100 mg	10 µg/µl	D1815
	500 mg	10 µg/µl	D1816

For Laboratory Use.

Description: Herring Sperm DNA is tested and certified to be free of any DNase or RNase activity. It is useful as a blocking agent in nucleic acid hybridization experiments.

Features:

- **Quality Tested:** Certified to be free of any DNase or RNase activity.
- **Multiple Applications:** Use as a blocking agent in hybridizations or as carrier DNA.
- **Ready to Use:** Provided as a 10mg/ml solution.
- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Storage Conditions: Store at –20°C.
Note: Product may be viscous at 4°C. Prior to use, ensure product is at room temperature (it may be briefly warmed at 37°C) and mixed thoroughly to ensure homogeneity.

Lambda DNA

Product	Size	Cat.#
Lambda DNA	250 µg	D1501

For Laboratory Use.

Description: λ DNA *c*I857 *Sam7* is isolated from infected *E. coli* strain W3350. Restriction enzyme-digested λ DNA (48,502bp) may be used as a molecular weight size marker in gel analysis of nucleic acids. λ DNA is also a commonly used substrate in restriction enzyme activity assays. The nucleotide sequence has been determined.

Features:

- **Choose Your Configuration:** Learn more about our custom options for this product at: **www.promega.com/myway**

Storage Conditions: Store at –20°C.

Unmethylated Lambda DNA

Product	Size	Cat.#
Unmethylated Lambda DNA	250 µg	D1521

For Laboratory Use.

Description: Unmethylated *c*I857 *Sam7* Lambda DNA (48,502bp) is isolated from infected GM119, an *E. coli* strain lacking both the *dam* and *dcm* methylase activities. Unmethylated Lambda DNA is used as a substrate for restriction enzymes sensitive to DNA methylation.

Features:

- **Unmethylated Substrate:** Use as a substrate for methylation-sensitive restriction enzymes.

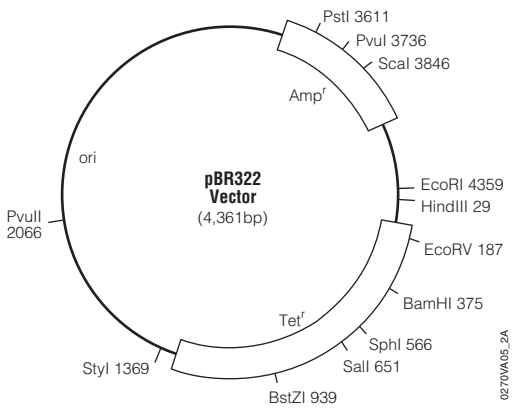
Storage Conditions: Store at –20°C.

pBR322 Vector

Product	Size	Conc.	Cat.#
pBR322 Vector	10 µg	1 µg/µl	D1511

Description: The plasmid pBR322 Vector (4,361bp) carries the genes for tetracycline and ampicillin resistance. pBR322 DNA digests typically are used as molecular weight size markers in gel analysis of nucleic acids.

Storage Conditions: Store at –20°C.



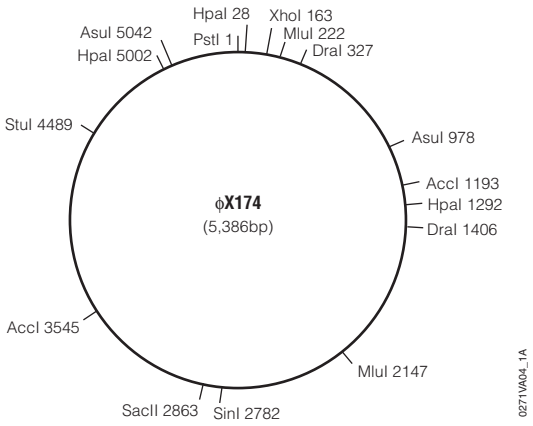
ΦX174, RF DNA

Product	Size	Conc.	Cat.#
ΦX174, RF DNA	50 µg	1 µg/µl	D1531

For Laboratory Use.

Description: The icosahedral bacteriophage ΦX174 replicative form (RF) is a double-stranded circular DNA molecule of 5,386 bases. Restriction enzyme-digested ΦX174 DNA generates molecular weight size markers used in gel analysis of nucleic acids. ΦX174 DNA is often used in the assays of restriction enzymes for the presence of nickase activity.

Storage Conditions: Store at –20°C.



pGEM®-3Z Vector

Product	Size	Cat.#
pGEM®-3Z Vector	20 µg	P2151

Description: The pGEM®-3Z Vector is intended for use as a standard cloning vector, as well as for the highly efficient synthesis of RNA in vitro. The vector carries the *lacZ* α-peptide and the multiple cloning region arrangement from pUC18 allowing selection of recombinants by blue/white screening. In addition, the vector contains both the SP6 and T7 RNA polymerase promoters flanking the multiple cloning region.

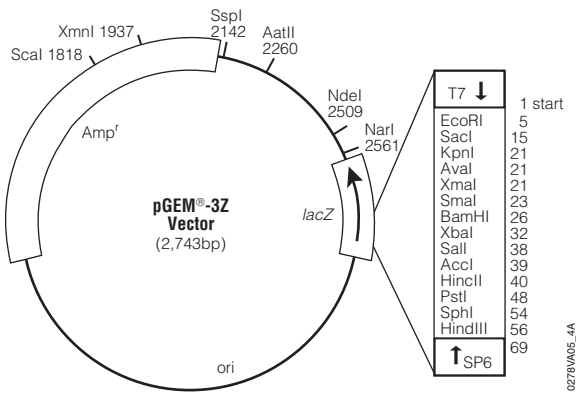
The pGEM®-3Z and pGEM®-4Z Vectors are essentially identical except for the orientation of the SP6 and T7 promoters.

Features:

- **Blue/White Screening:** Allows the easy identification of recombinant clones.
- **Versatile:** This vector can be used for standard cloning and in vitro transcription from SP6 and T7 RNA polymerase promoters flanking the multiple cloning region.
- **Convenient:** Multiple cloning site provides a selection of restriction sites for cloning.

Protocol	Part#
Technical Bulletin	TB033

Storage Conditions: Store vector at –20°C.



pGEM®-4Z Vector

Product	Size	Cat.#
pGEM®-4Z Vector	20 µg	P2161

Description: The pGEM®-4Z Vector is intended for use as a standard cloning vector, as well as for the highly efficient synthesis of RNA in vitro. The vector carries the *lacZ* α-peptide and the multiple cloning region arrangement from pUC18 allowing selection of recombinants by blue/white screening. In addition, the vector contains both the SP6 and T7 RNA polymerase promoters flanking the multiple cloning region.

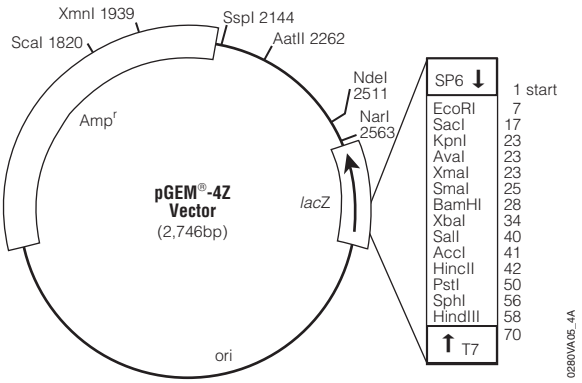
The pGEM®-3Z and pGEM®-4Z Vectors are essentially identical except for the orientation of the SP6 and T7 promoters.

Features:

- **Blue/White Screening:** Easily identify recombinant clones.
- **Versatile:** Use this vector for standard cloning and in vitro transcription from SP6 and T7 RNA polymerase promoters flanking the multiple cloning region.
- **Convenient:** Multiple cloning site provides a selection of restriction sites for cloning.

Protocol	Part#
Technical Bulletin	TB036

Storage Conditions: Store vector at –20°C and bacterial strain at –70°C.



pGEM®-3Zf(+/–) Vectors

Product	Size	Cat.#
pGEM®-3Zf(+) Vector	20 µg	P2271
pGEM®-3Zf(–) Vector	20 µg	P2261

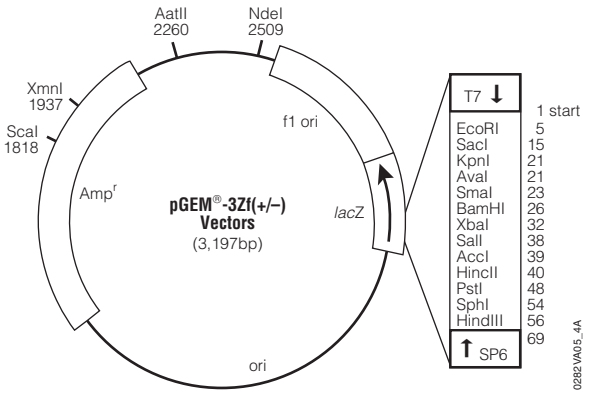
Description: The pGEM®-3Zf(+) and pGEM®-3Zf(–) Vectors are derived from the pGEM®-3Z Vector and contain the origin of replication of the filamentous phage f1. These plasmids contain T7 and SP6 RNA polymerase promoters flanking a multiple cloning region within the α-peptide coding region of β-galactosidase. Insertional inactivation of the α-peptide allows recombinant clones to be identified directly by color screening on indicator plates when using appropriate *E. coli* strains (e.g., JM109). The multiple cloning region contains unique restriction sites for EcoRI, SacI, KpnI, AvaI, SmaI, BamHI, XbaI, SalI, AccI, HincII, PstI, SphI and HindIII. The pGEM®-3Zf(+) and -3Zf(–) Vectors are identical except for the orientation of the f1 origin and can be used as standard cloning vectors, as templates for in vitro transcription and for the production of circular ssDNA.

Features:

- **Blue/White Screening:** Allows the easy identification of recombinant clones.
- **Versatile:** These vectors can be used for standard cloning, single-stranded DNA production and in vitro transcription from SP6 and T7 RNA polymerase promoters flanking the multiple cloning region.
- **Convenient:** Multiple cloning site provides a selection of restriction sites for cloning.

Protocol	Part#
pGEM®-3Zf(+) Vector Technical Bulletin	TB086
pGEM®-3Zf(–) Vector Technical Bulletin	TB045

Storage Conditions: Store vector at –20°C and bacterial strain at –70°C.



pGEM®-5Zf(+) Vector

Product	Size	Cat.#
pGEM®-5Zf(+) Vector	20 µg	P2241

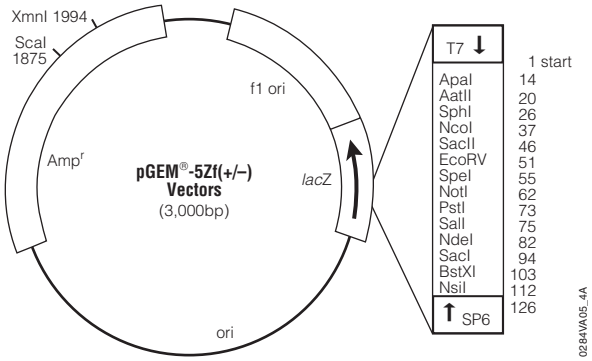
Description: The pGEM®-5Zf(+) Vector is derived from the pGEM®-3Zf(+) Vector and contains the origin of replication of the filamentous phage f1. This plasmid serves as a standard cloning vector, as a template for in vitro transcription and can be used for the production of circular ssDNA. This vector contains T7 and SP6 RNA polymerase promoters flanking a multiple cloning region within the α-peptide coding region of β-galactosidase. Insertional inactivation of the α-peptide allows recombinant clones to be identified directly by color screening on indicator plates when using appropriate *E. coli* strains (e.g., JM109). The multiple cloning region contains unique restriction sites for ApaI, AatII, SphI, NcoI, SacII, EcoRV, SpeI, NotI, PstI, SalI, NdeI, SacI, BstXI and NsiI. This arrangement is designed specifically for generating unidirectional deletions with the Erase-a-Base® System.

Features:

- **Blue/White Screening:** Allows the easy identification of recombinant clones.
- **Versatile:** This vector can be used for standard cloning, single-stranded DNA production and in vitro transcription from SP6 and T7 RNA polymerase promoters flanking the multiple cloning region.
- **Convenient:** Multiple cloning site provides a selection of restriction sites for cloning.
- **Unidirectional Deletions:** Restriction sites are positioned conveniently for use with the Erase-a-Base® System.

Protocol	Part#
Technical Bulletin	TB047

Storage Conditions: Store vector at –20°C and bacterial strain at –70°C.



pGEM®-7Zf(+/–) Vectors

Product	Size	Cat.#
pGEM®-7Zf(+) Vector	20 µg	P2251
pGEM®-7Zf(–) Vector	20 µg	P2371

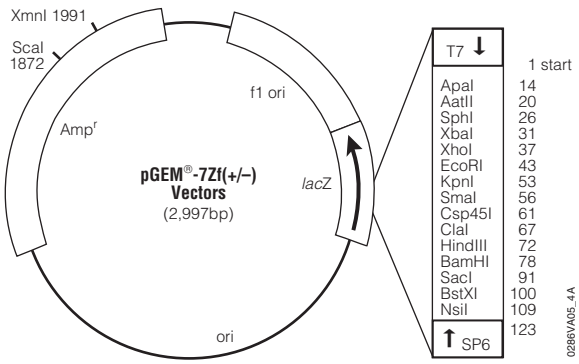
Description: The pGEM®-7Zf(+) and pGEM®-7Zf(–) Vectors are derivatives of the pGEM®-3Zf(+) Vector and contain the origin of replication of the filamentous phage f1. These plasmids serve as standard cloning vectors, as templates for in vitro transcription and can be used for the production of circular ssDNA. These plasmids contain SP6 and T7 RNA polymerase promoters flanking a region of multiple cloning sites within the α-peptide coding region of β-galactosidase. Insertional inactivation of the α-peptide allows recombinant clones to be identified directly by color screening on indicator plates when using appropriate *E. coli* strains (e.g., JM109). The multiple cloning region is unique and includes restriction sites for ApaI, AatII, SphI, XbaI, XhoI, EcoRI, KpnI, SmaI, Csp45I, ClaI, HindIII, BamHI, SacI, BstXI and NsiI. This arrangement is designed specifically for generating unidirectional deletions with the Erase-a-Base® System. pGEM®-7Zf(+) and pGEM®-7Zf(–) Vectors are identical except for the orientation of the f1 origin.

Features:

- **Blue/White Screening:** Allows the easy identification of recombinant clones.
- **Versatile:** These standard cloning vectors are equipped for single-stranded DNA production and in vitro transcription from SP6 and T7 RNA polymerase promoters flanking the multiple cloning region.
- **Convenient:** Multiple cloning site provides a selection of restriction sites for cloning.
- **Unidirectional Deletions:** Restriction sites are positioned conveniently for use with the Erase-a-Base® System.

Protocol	Part#
pGEM®-7Zf(+) Vector Technical Bulletin	TB048
pGEM®-7Zf(–) Vector Technical Bulletin	TB069

Storage Conditions: Store vector at –20°C and bacterial strain at –70°C.



pGEM®-9Zf(–) Vector

Product	Size	Cat.#
pGEM®-9Zf(–) Vector	20 µg	P2391

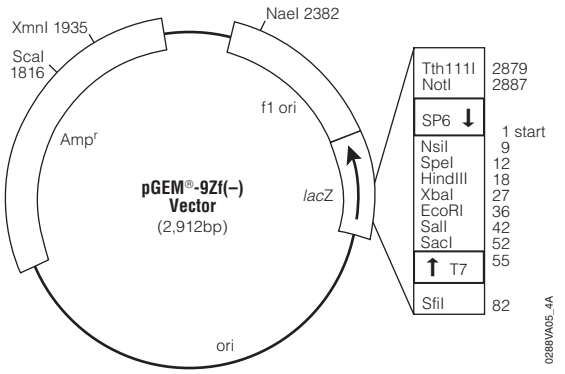
Description: The pGEM®-9Zf(–) Vector is a recombinant plasmid designed to provide a versatile range of cloning strategies, efficient synthesis of RNA in vitro and the production of single-stranded DNA. The plasmid contains T7 and SP6 RNA polymerase promoters flanking a multiple cloning region within the α-peptide coding region of β-galactosidase. Insertional inactivation of the α-peptide allows recombinant clones to be identified directly by color screening on indicator plates when using appropriate *E. coli* strains (e.g., JM109). The multiple cloning region is unique and includes restriction sites for NsiI, SpeI, HindIII, XbaI, EcoRI, SalI and SacI.

Features:

- **Excisable SP6/T7 Insert:** This vector allows the excision of an insert containing the SP6 and T7 RNA polymerase promoters.
- **Blue/White Screening:** Allows the easy identification of recombinant clones.
- **Versatile:** This vector can be used for standard cloning, single-stranded DNA production and in vitro transcription from SP6 and T7 RNA polymerase promoters flanking the multiple cloning region.
- **Convenient:** Multiple cloning site provides a selection of restriction sites for cloning.

Protocol	Part#
Technical Bulletin	TB070

Storage Conditions: Store vector at –20°C and bacterial strain at –70°C.



pGEM®-11Zf(+/–) Vectors

Product	Size	Cat.#
pGEM®-11Zf(+) Vector	20 µg	P2411
pGEM®-11Zf(–) Vector	20 µg	P2421

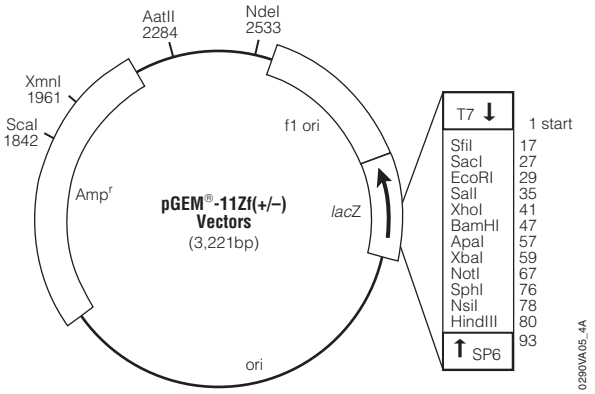
Description: The pGEM®-11Zf(+) and pGEM®-11Zf(–) Vectors can be used as standard cloning vectors, as templates for in vitro transcription and for the production of ssDNA. These plasmids contain T7 and SP6 RNA polymerase promoters flanking a multiple cloning region within the α-peptide coding region of β-galactosidase. Insertional inactivation of the α-peptide allows recombinant clones to be identified directly by color screening on indicator plates when using appropriate *E. coli* strains (e.g., JM109). The multiple cloning region contains unique restriction sites for SfiI, SacI, EcoRI, SalI, XhoI, BamHI, ApaI, XbaI, NotI, SphI, NsiI and HindIII. The pGEM®-11Zf(–) and pGEM®-11Zf(+) Vectors are identical except for the orientation of the f1 origin.

Features:

- **Blue/White Screening:** Easily identify recombinant clones.
- **Versatile:** These vectors can be used for standard cloning, single-stranded DNA production and in vitro transcription from SP6 and T7 RNA polymerase promoters flanking the multiple cloning region.
- **Convenient:** Multiple cloning site provides a selection of restriction sites for cloning.

Protocol	Part#
pGEM®-11Zf(+) Vector Technical Bulletin	TB075
pGEM®-11Zf(–) Vector Technical Bulletin	TB074

Storage Conditions: Store vector at –20°C and bacterial strain at –70°C.



pSP72 Vector

Product	Size	Cat.#
pSP72 Vector	20 µg	P2191

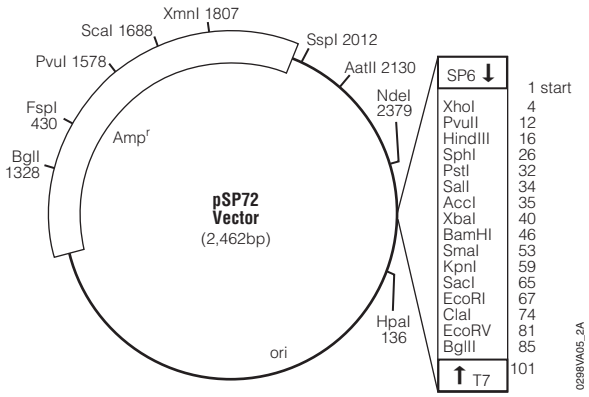
Description: The pSP72 Vector can be used as a standard cloning vector and also can be used for transcription of RNA in vitro. The pSP72 Vector contains the SP6 and T7 RNA polymerase promoters flanking a unique multiple cloning region, which includes restriction sites for XhoI, PvuII, HindIII, SphI, PstI, SalI, AccI, XbaI, BamHI, SmaI, KpnI, SacI, EcoRI, ClaI, EcoRV and BglII. The pSP72 and pSP73 Vectors are essentially identical except for the orientation of the multiple cloning site region.

Features:

- **Versatile:** This vector can be used for standard cloning and in vitro transcription from SP6 and T7 RNA polymerase promoters flanking the multiple cloning region.
- **Convenient:** Multiple cloning site provides a selection of restriction sites for cloning.

Protocol	Part#
Technical Bulletin	TB040

Storage Conditions: Store vector at –20°C.



pSP73 Vector

Product	Size	Cat.#
pSP73 Vector	20 µg	P2221

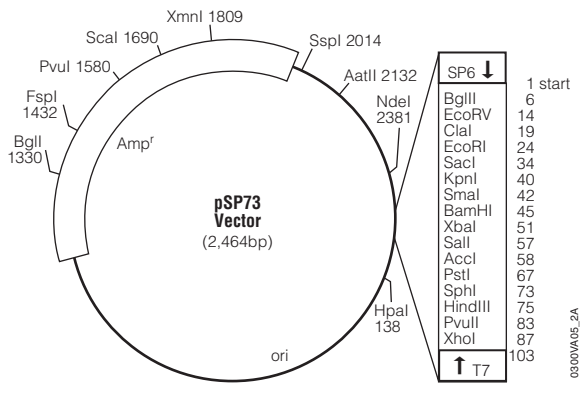
Description: The pSP73 Vector offers a wide range of restriction sites, providing greater versatility in cloning and transcription of RNA in vitro. The pSP73 Vector contains the SP6 and T7 RNA polymerase promoters and a unique multiple cloning region, which includes restriction sites for BglII, EcoRV, ClaI, EcoRI, SacI, KpnI, SmaI, BamHI, XbaI, SalI, AccI, PstI, SphI, HindIII, PvuII and XhoI. The pSP72 and pSP73 Vectors are essentially identical except for the orientation of the multiple cloning region.

Features:

- **Versatile:** This vector can be used for standard cloning and in vitro transcription from SP6 and T7 RNA polymerase promoters flanking the multiple cloning region.
- **Convenient:** Multiple cloning site provides a selection of restriction sites for cloning.

Protocol	Part#
Technical Bulletin	TB041

Storage Conditions: Store vector at –20°C.



pSP64 Poly(A) Vector

Product	Size	Cat.#
pSP64 Poly(A) Vector	20 µg	P1241

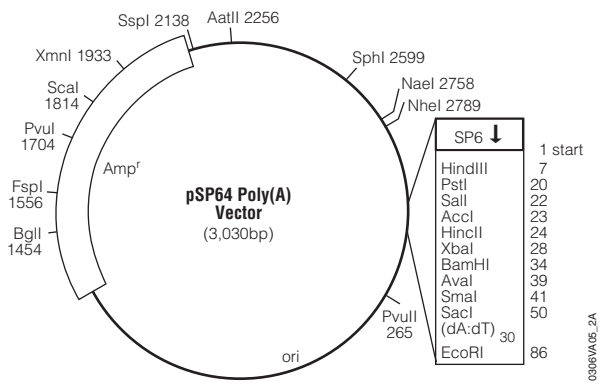
Description: The pSP64 Poly(A) Vector can be used as a standard cloning vector and for in vitro transcription from the SP6 promoter. The pSP64 Poly(A) Vector also can be used to generate poly(A)+ transcripts in vitro. The vector has a stretch of 30 dA:dT residues inserted between the SacI and EcoRI sites. Therefore, when foreign DNA is cloned into any polylinker site other than EcoRI (HindIII, PstI, SalI, AccI, HincII, XbaI, BamHI, AvaI, SmaI or SacI), linearization of the recombinant plasmid with EcoRI allows the use of SP6 RNA polymerase in vitro to prepare RNA copies of the inserted sequences that contain a synthetic 3´ "poly(A)" tail of 30 residues.

Features:

- **In Vitro Transcription:** The SP6 promoter is next to the polylinker.
- **Generates Poly(A)+ Transcripts In Vitro:** A stretch of 30 dA:dT residues are inserted between the SacI and EcoRI sites in the polylinker. Poly(A) tails can stabilize RNAs and lead to greater yields for in vitro translation reactions.
- **Convenient:** Multiple cloning region provides a selection of restriction sites for cloning.

Protocol	Part#
Technical Bulletin	TB052

Storage Conditions: Store vector at –20°C.



JM109 and HB101 Competent Cells

Product	Size	Cat.#
JM109 Competent Cells, >10^8cfu/µg	5 × 200 µl	L2001
JM109 Competent Cells, >10^7cfu/µg	5 × 200 µl	L1001
HB101 Competent Cells, >10^8cfu/µg	5 × 200 µl	L2011

Cat.# L2001 For Laboratory Use.

Description: JM109 and HB101 Competent Cells are prepared according to a modified procedure of Hanahan. JM109 Competent Cells are available for convenient transformation in two efficiencies: High Efficiency at greater than 10^8cfu/µg and Subcloning Efficiency at greater than 10^7cfu/µg. JM109 cells are an ideal host for many molecular biology applications. HB101 cells are useful for cloning in vectors that do not require α-complementation for blue/white screening. pGEM®-3Z Vector is provided as a positive control.

JM109 Genotype: *end*A1, *rec*A1, *gyr*A96, *thi, hsd*R17 (r_K^-, m_K^+), *rel*A1, *sup*E44, Δ(*lac-pro*AB), [F´ *tra*D36, *pro*AB, *laq*I^qZΔM15].

HB101 Genotype: F–, *thi*-1, *hsd*S20 (r_B^-, m_B^-), *sup*E44, *rec*A13, *ara*-14, *leu*B6, *pro*A2, *lac*Y1, *gal*K2, *rps*L20 (str^r), *xyl*-5, *mtl*-1.

Features:

- **Convenient:** Ready to use; no preparation time necessary.
- **Reliable:** Transformation efficiencies guaranteed; positive control included.
- **Versatile:** Use JM109 for subcloning, T-vector cloning, blue/white screening, production of single-stranded DNA from phagemids (F´ episome).
- **High Transformation Efficiency:** Greater than 10^8cfu/µg (Cat.# L2001 and L2011).
- **Safe:** The *rec*A– mutation prevents undesirable recombination events, and the *end*A– mutation in JM109 cells prevents carryover nuclease in miniprep DNA.

Protocol	Part#
Technical Bulletin	TB095

Storage Conditions: Store at –70°C.

BMH 71-18 *mut*S Competent Cells

Product	Size	Cat.#
BMH 71-18 *mut*S Competent Cells, >10^7cfu/µg	5 × 200 µl	L1201

Description: BMH 71-18 *mut*S is a mismatch repair minus strain of *E. coli*. Use of this strain prevents repair of the newly synthesized unmethylated strand leading to high mutation efficiencies. Use these cells in conjunction with any mutagenesis system that requires the use of a repair minus (*mut*S) cell line including the GeneEditor™ in vitro Site-Directed Mutagenesis System, restriction site elimination mutagenesis systems, and hemimethylation/DpnI-based systems. BMH 71-18 *mut*S is *rec*A+, and as a result, inserts containing highly repetitive sequences may be unstable. BMH 71-18 *mut*S is tetracycline-resistant due to the presence of Tn10. BMH 71-18 *mut*S is also restriction (+). Isolate template DNA from a modification (+) K12 strain or it will be restricted. For example, DNA isolated from HB101 or NM522 (modification minus strains) or BL21 (*E. coli* B strain) cells should not be used. BMH 71-18 *mut*S Competent Cells have a transformation efficiency of greater than 10^7cfu/µg. One milliliter of cells is sufficient for ten transformations.

Genotype: *thi, sup*E, Δ(*lac-pro*AB), [*mut*S::Tn10], [F′, *pro*AB, *lacI*^{q}ZΔM15].

Features:

- **Convenient:** A time-saving alternative to making competent cells in your laboratory.
- **Useful for Mutagenesis:** Compatible with most mutagenesis systems requiring a repair minus (*mut*S) strain.

Protocol	Part#
Technical Bulletin	TB095

Storage Conditions: Store at –70°C.

Bacterial Strains BMH 71-18 *mut*S and ES1301 *mut*S

Product	Size	Cat.#
Bacterial Strain ES1301 *mut*S, Glycerol Stock (noncompetent)	200 µl	Q6131
Bacterial Strain BMH 71-18 *mut*S, Glycerol Stock (noncompetent)	500 µl	Q6321

Description: BMH 71-18 *mut*S and ES1301 *mut*S are mismatch repair minus strains of *E. coli*. Use of these strains prevents repair of the newly synthesized unmethylated strand, leading to high mutation efficiencies and making them helpful in such systems as the GeneEditor™ Mutagenesis System. Both ES1301 and BMH 71-18 *mut*S are *rec*A+, therefore inserts containing highly repetitive sequences may be unstable. ES1301 is Tet^s, which allows for cycling between antibiotic resistances. BMH 71-18 is Tet^r; therefore, it can only be used to select Amp^r or CAT^r mutants or in the GeneEditor™ System.

ES1301 *mut*S Genotype: *lac*Z53, *mut*S201::Tn5, *thy*A36, *rha*-5, *met*B1, *deo*C, IN(*rrn*D–*rrn*E).

BMH 71-18 *mut*S Genotype: *thi, sup*E, Δ(*lac-pro*AB), [*mut*S::Tn10], [F′, *pro*AB, *lacI*^{q}ZΔM15].

Storage Conditions: Store at –70°C.

Bacterial Strain JM109

Product	Size	Cat.#
Bacterial Strain JM109, Glycerol Stock	500 µl	P9751

Description: Bacterial Strain JM109 is a useful host for transformation of pGEM® Vectors and for production of single-stranded DNA from M13 or phagemid vectors. The strain grows well and is transformed efficiently by a variety of methods. Because JM109 is *rec*A– and lacks the *E. coli* K restriction system, undesirable restriction of cloned DNA and recombination with host chromosomal DNA are prevented. The endonuclease A– mutation leads to an improved yield and quality of isolated plasmid DNA.

JM109 is deficient in β-galactosidase activity due to deletions in both genomic and episomal copies of the *lac*Z gene. The deletion in the episomal (F′ factor) copy of the *lac*Z gene (*lac*ZΔM15) can be complemented by addition of a functional α-peptide encoded by a pGEM®-Z or pGEM®-Zf Vector. If complementation does not occur, bacterial colonies are white. To maintain the F′, JM109 should be grown on minimal (M-9) media supplemented with 1mM thiamine.

Genotype: *end*A1, *rec*A1, *gyr*A96, *thi, hsd*R17 (r_k^-, m_k^+), *rel*A1, *sup*E44, Δ(*lac-pro*AB), [F′ *tra*D36, *pro*AB, *lacI*^{q}ZΔM15].

Features:

- **Reliable:** Grows well and is transformed efficiently.
- **Versatile:** Useful for cloning, single-stranded DNA production, and blue/white screening.
- **High Yields of Plasmid DNA:** The endonuclease A– mutation improves yield and quality of isolated plasmid DNA.

Storage Conditions: Store at –70°C.

Bacterial Strain JM109(DE3)

Product	Size	Cat.#
Bacterial Strain JM109(DE3), Glycerol Stock	500 µl	P9801

Description: Bacterial Strain JM109(DE3), derived from JM109, contains a chromosomal copy of the gene for T7 RNA polymerase. JM109(DE3) is used for the high-level expression of genes cloned into vectors for expression of sequence downstream from the T7 promoter, provided that the cloned sequence contains a ribosomal binding site. **Please note:** JM109(DE3) cannot be used for blue/white selection.

Genotype: *end*A1, *rec*A1, *gyr*A96, *thi, hsd*R17 (r_k^-, m_k^+), *rel*A1, *sup*E44, λ–, Δ(*lac-pro*AB), [F′, *tra*D36, *pro*AB, *lacI*^{q}ZΔM15], IDE3.

Features:

- **T7 Promoter Expression:** Contains an IPTG-inducible gene for T7 RNA polymerase.

Storage Conditions: Store at –70°C.

Bacterial Strain LE392

Product	Size	Cat.#
Bacterial Strain LE392, Glycerol Stock	500 µl	K9981

Description: Bacterial Strain LE392 is used for both genomic and cDNA cloning. LE392 lacks the *E. coli* K restriction system but is *rec*+. It is a permissive host, allowing both recombinant and parental phage to grow. Unlike Y1090, LE392 does not allow color selection of recombinants or IPTG induction of expression. Also, since LE392 contains *lon* protease activity, fusion proteins are often less stable in this host than in Y1090. When using this strain, phage titers tend to be higher than with Y1090 host cells. Therefore, we recommend (if color selection is not required) that LE392 be the primary strain for amplification of recombinant phage and for screening a cDNA library with nucleic acid probes.

Genotype: *hsd*R514(r_k^-, m_k^+), *gln*V(*sup*E44), *try*T (*sup*F58), *lac*Y1 or Δ(*lac*IZY)6, *gal*K2, *gal*T22, *met*B1, *trp*R55.

Storage Conditions: Store at –70°C.

Bacterial Strain NM522

Product	Size	Cat.#
Bacterial Strain NM522, Glycerol Stock	500 µl	P2301

Description: NM522 contains an F′ episome, which is required for production of ssDNA and for blue/white color selection. To maintain the F′, NM522 should be grown on minimal (M-9) medium.

Genotype: *sup*E, *thi*, Δ(*lac-pro*AB), *hsd*5 (r^-, m^-), [F′, *pro*AB, *lacI*^{q}ZΔM15].

Storage Conditions: Store at –70°C.

Helper Phage

Product	Size	Cat.#
R408 Helper Phage DNA	10 µg	P2341

Description: The pGEM®-Zf Vectors require the presence of a helper phage in order to be propagated, packaged and secreted as single-stranded DNA (ssDNA) viral particles. R408 helper phage allows the preferential secretion of single-stranded plasmid DNA over the phage ssDNA, thereby increasing the absolute yield of plasmid ssDNA. Differences in the yields and absolute amounts of plasmid and phage ssDNA produced have been observed to be dependent on the particular host and vector combination.

Concentration: Minimum specification is ≥5×10^{10}pfu/ml.

Features:

- **Increased Single-Stranded DNA Yield:** R408 Helper Phage allows the preferential secretion of single-stranded plasmid DNA over the phage ssDNA.

Storage Conditions: Store at –20°C.

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A1120	Wizard® Genomic DNA Purification Kit	100 isolations × 300 µl	102
A1125	Wizard® Genomic DNA Purification Kit	500 isolations × 300 µl	102
A1222	PureYield™ Plasmid Miniprep System	250 preps	89
A1223	PureYield™ Plasmid Miniprep System	100 preps	89
A1250	Access RT-PCR System	100 reactions	14
A1260	Access RT-PCR Introductory System	20 reactions	14
A1280	Access RT-PCR System	500 reactions	14
A1311	Wizard® SV Wash Solution	185 ml	95
A1311	Wizard® SV Wash Solution	185 ml	102
A1311	Wizard® SV Wash Solution	185 ml	103
A1318	Wizard® SV 96 Wash Solution	370 ml	95
A1330	Wizard® *Plus* SV Minipreps DNA Purification System	50 preps	89
A1331	Vacuum Adapters	20 each	97
A1331	Vacuum Adapters	20 each	107
A1340	Wizard® *Plus* SV Minipreps DNA Purification System + Vacuum Adapters	50 preps	89
A1360	pGEM®-T Easy Vector System I	20 reactions	19
A1380	pGEM®-T Easy Vector System II	20 reactions	19
A1410	pTARGET™ Mammalian Expression Vector System	20 reactions	20
A1441	Alkaline Protease Solution	3 ml	89
A1441	Alkaline Protease Solution	3 ml	95
A1460	Wizard® *Plus* SV Minipreps DNA Purification System	250 preps	89
A1465	Wizard® *Plus* SV Minipreps DNA Purification System	1,000 preps	89
A1470	Wizard® *Plus* SV Minipreps DNA Purification System + Vacuum Adapters	250 preps	89
A1481	Wizard® SV 96 Neutralization Solution	500 ml	95
A1485	Neutralization Solution (NSB)	500 ml	90
A1488	Wizard® SV 96 Neutralization Solution	950 ml	95
A1491	Promega 10 Barrier Tips, 960/pk	0.5–10 µl	20
A1501	Promega 10E Barrier Tips, 960/pk	0.5–10 µl	20
A1511	Promega 10F Barrier Tips, 960/pk	0.5–10 µl	20
A1521	Promega 20 Barrier Tips, 960/pk	2–20 µl	20
A1531	Promega GEL Barrier Tips, 768/pk	0.5–30 µl	20
A1541	Promega 100 Barrier Tips, 960/pk	10–100 µl	20
A1551	Promega 200 Barrier Tips, 960/pk	50–200 µl	20
A1561	Promega 1000 Barrier Tips, 480/pk	100–1,000 µl	20
A1620	Wizard® Genomic DNA Purification Kit	100 isolations × 10 ml	102
A1630	Wizard® MagneSil® Plasmid Purification System	4 × 96 preps	96
A1631	Wizard® MagneSil® Plasmid Purification System	8 × 96 preps	96
A1635	Wizard® MagneSil® Plasmid Purification System, HTP1	100 × 96 preps	96
A1641	MagneSil® RED	100 ml	96
A1641	MagneSil® RED	100 ml	108
A1655	Elution Buffer	500 ml	96
A1701	AccessQuick™ RT-PCR System	20 reactions	14
A1702	AccessQuick™ RT-PCR System	100 reactions	14
A1703	AccessQuick™ RT-PCR System	500 reactions	14
A1712	ReliaPrep™ LV 32 HSM Instrument Cover	1 each	101
A1713	ReliaPrep™ Tube Rack	1 each	101
A1714	ReliaPrep™ Tube Rack Stand	1 each	101
A1715	ReliaPrep™ LV 32 HSM Instrument	1 each	101
A1751	ReliaPrep™ Large Volume HT gDNA Isolation System	96 × 10ml preps	101
A1831	Wizard® MagneSil® Sequencing Reaction Clean-Up System	4 × 96 preps	100
A1832	Wizard® MagneSil® Sequencing Reaction Clean-Up System	8 × 96 preps	100
A1835	Wizard® MagneSil® Sequencing Reaction Clean-Up System, HTP1	100 × 96 preps	100
A2180	Wizard® PCR Preps DNA Purification System	250 preps	98
A2191	Endotoxin Removal Resin	100 ml	94
A2201	MagneSil® BLUE	100 ml	96
A2201	MagneSil® BLUE	100 ml	108
A2221	4/40 Wash Solution	115 ml	94
A2241	Wizard® SV 96 Lysate Clearing Plates	10 pack	95
A2241	Wizard® SV 96 Lysate Clearing Plates	10 pack	107

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A2248	Wizard® SV 96 Lysate Clearing Plates	100 pack	107
A2250	Wizard® SV 96 Plasmid DNA Purification System	1 × 96 preps	95
A2255	Wizard® SV 96 Plasmid DNA Purification System	5 × 96 preps	95
A2258	Wizard® SV 9600 Plasmid DNA Purification System	100 × 96 preps	95
A2271	Wizard® SV 96 Binding Plates	10 pack	95
A2271	Wizard® SV 96 Binding Plates	10 pack	108
A2278	Wizard® SV 96 Binding Plates	100 pack	95
A2278	Wizard® SV 96 Binding Plates	100 pack	108
A2291	Vac-Man® 96 Vacuum Manifold	1 each	107
A2311	Collar for Vac-Man® 96 Vacuum Manifold	1 each	107
A2360	Wizard® SV Genomic DNA Purification System	50 preps	102
A2361	Wizard® SV Genomic DNA Purification System	250 preps	102
A2370	Wizard® SV 96 Genomic DNA Purification System	1 × 96 preps	103
A2371	Wizard® SV 96 Genomic DNA Purification System	4 × 96 preps	103
A2380	Wizard MagneSil Tfx™ System	4 × 96 preps	94
A2392	PureYield™ Plasmid Maxiprep System	10 preps	91
A2393	PureYield™ Plasmid Maxiprep System	25 preps	91
A2492	PureYield™ Plasmid Midiprep System	25 preps	90
A2495	PureYield™ Plasmid Midiprep System	100 preps	90
A2496	PureYield™ Plasmid Midiprep System	300 preps	90
A3500	Reverse Transcription System	100 reactions	13
A3511	Magnesium Chloride Solution	1.5 ml	5
A3511	Magnesium Chloride Solution	1.5 ml	13
A3511	Magnesium Chloride Solution	1.5 ml	255
A3513	Magnesium Chloride Solution	25 ml	5
A3561	Reverse Transcription 10X Buffer	1.4 ml	13
A3561	Reverse Transcription 10X Buffer	1.4 ml	255
A3600	pGEM®-T Vector System I	20 reactions	18
A3610	pGEM®-T Vector System II	20 reactions	18
A3800	ImProm-II™ Reverse Transcription System	100 reactions	12
A3800	ImProm-II™ Reverse Transcription System	100 reactions	256
A3801	ImProm-II™ Reverse Transcriptase	10 reactions	12
A3801	ImProm-II™ Reverse Transcriptase	10 reactions	256
A3802	ImProm-II™ Reverse Transcriptase	100 reactions	12
A3802	ImProm-II™ Reverse Transcriptase	100 reactions	256
A3803	ImProm-II™ Reverse Transcriptase	500 reactions	12
A3803	ImProm-II™ Reverse Transcriptase	500 reactions	256
A3811	Wash Buffer, Plant	40 ml	106
A4011	Plexor® qPCR System	200 reactions	8
A4021	Plexor® One-Step qRT-PCR System	200 reactions	8
A4051	Plexor® Two-Step qRT-PCR System	200 reactions	8
A4080	MagneSil® Genomic, Large Volume System	8 preps	105
A4082	MagneSil® Genomic, Large Volume System	48 preps	105
A4085	MagneSil® Genomic, Large Volume System	96 preps	105
A4091	eLysis Buffer, Large Volume System	1 L	105
A5000	GoScript™ Reverse Transcription System	50 reactions	12
A5000	GoScript™ Reverse Transcription System	50 reactions	255
A5001	GoScript™ Reverse Transcription System	100 reactions	12
A5001	GoScript™ Reverse Transcription System	100 reactions	255
A5003	GoScript™ Reverse Transcriptase	100 reactions	12
A5003	GoScript™ Reverse Transcriptase	100 reactions	255
A5004	GoScript™ Reverse Transcriptase	500 reactions	12
A5004	GoScript™ Reverse Transcriptase	500 reactions	255
A5081	ReliaPrep™ Blood gDNA Miniprep System	100 preps	101
A5082	ReliaPrep™ Blood gDNA Miniprep System	250 preps	101
A6001	GoTaq® qPCR Master Mix	200 reactions	7
A6002	GoTaq® qPCR Master Mix	1,000 reactions	7
A7100	Wizard® *Plus* Minipreps DNA Purification System	50 preps	92
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A7112	Cell Resuspension Solution (CRA)	150 ml	92

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A7112	Cell Resuspension Solution (CRA)	150 ml	93
A7113	Wizard® SV 96 Cell Resuspension Solution	500 ml	95
A7114	Cell Resuspension Solution	500 ml	96
A7115	Cell Resuspension Solution (CRA)	315 ml	90
A7118	Wizard® SV 96 Cell Resuspension Solution	800 ml	95
A7122	Cell Lysis Solution (CLA)	150 ml	92
A7122	Cell Lysis Solution (CLA)	150 ml	92
A7122	Cell Lysis Solution (CLA)	150 ml	93
A7122	Cell Lysis Solution (CLA)	150 ml	93
A7123	Wizard® SV 96 Cell Lysis Solution	500 ml	95
A7124	Cell Lysis Solution	500 ml	96
A7125	Cell Lysis Solution (CLA)	315 ml	90
A7128	Wizard® SV 96 Cell Lysis Solution	800 ml	95
A7131	Neutralization Solution (NSA)	150 ml	92
A7131	Neutralization Solution (NSA)	150 ml	92
A7131	Neutralization Solution (NSA)	150 ml	93
A7131	Neutralization Solution (NSA)	150 ml	93
A7132	Neutralization Solution	500 ml	96
A7141	Wizard® Minipreps DNA Purification Resin	250 ml	92
A7170	Wizard® PCR Preps DNA Purification System	50 preps	98
A7181	Wizard® PCR Preps DNA Purification Resin	250 ml	98
A7211	Wizard® Minicolumns	250 each	92
A7211	Wizard® Minicolumns	250 each	98
A7231	Vac-Man® Laboratory Vacuum Manifold, 20-sample capacity	1 each	107
A7241	Direct Purification Buffer	25 ml	98
A7261	One-Way Luer-Lok® Stopcocks	10 each	107
A7270	Wizard® *Plus* Maxipreps DNA Purification System	10 preps	93
A7280	Wizard® DNA Clean-Up System	100 preps	98
A7300	Wizard® *Plus* Megapreps DNA Purification System	5 preps	93
A7361	Wizard® Megapreps DNA Purification Resin	1,000 ml	93
A7401	Wizard® Maxipreps DNA Purification Resin	500 ml	93
A7421	Wizard® Maxi/Megapreps Filtering System	50 each	93
A7421	Wizard® Maxi/Megapreps Filtering System	50 each	93
A7500	Wizard® *Plus* Minipreps DNA Purification System	100 preps	92
A7510	Wizard® *Plus* Minipreps DNA Purification System	250 preps	92
A7640	Wizard® *Plus* Midipreps DNA Purification System	25 preps	92
A7651	Wizard® Midicolumns	100 each	92
A7660	Vac-Man® Jr. Laboratory Vacuum Manifold, 2-sample capacity	1 each	107
A7701	Wizard® Midipreps DNA Purification Resin	1,000 ml	92
A7710	ReadyAmp™ Genomic DNA Purification System	100 reactions	105
A7933	Cell Lysis Solution (Genomic Purification)	1 liter	102
A7941	Nuclei Lysis Solution	50 ml	102
A7941	Nuclei Lysis Solution	50 ml	102
A7941	Nuclei Lysis Solution	50 ml	103
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A7951	Protein Precipitation Solution	25 ml	102
A7953	Protein Precipitation Solution	350 ml	102
A7963	DNA Rehydration Solution	50 ml	102
A7973	RNase A Solution, 4mg/ml	1 ml	102
A7973	RNase A Solution, 4mg/ml	1 ml	102
A7973	RNase A Solution, 4mg/ml	1 ml	103
A8102	Column Wash Solution (CWB)	125 ml	92
A8102	Column Wash Solution (CWB)	125 ml	92
A8102	Column Wash Solution (CWB)	125 ml	93
A8102	Column Wash Solution (CWB)	125 ml	93
A8191	Lysis Buffer A, Food	100 ml	106
A8231	MagneSil® GREEN	100 ml	100
A8231	MagneSil® GREEN	100 ml	108
A8251	DNA IQ™ Resin	50 ml	119
A8251	DNA IQ™ Resin	50 ml	120
A8261	Lysis Buffer	150 ml	119
A8261	Lysis Buffer	150 ml	120
A8271	2X Wash Buffer	70 ml	119
A8271	2X Wash Buffer	70 ml	120
A8281	Elution Buffer	50 ml	119
A8281	Elution Buffer	50 ml	120
A8501	Differex™ Digestion Buffer	150 ml	120
A8511	Differex™ Separation Solution	40 ml	120
A9161	Collection Plates (4-pack)	1 each	96
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A9301	Membrane Binding Solution	20 ml	97
A9301	Membrane Binding Solution	20 ml	99
A9340	Wizard® SV 96 PCR Clean-Up System	1 × 96 preps	99
A9341	Wizard® SV 96 PCR Clean-Up System	4 × 96 preps	99
A9342	Wizard® SV 96 PCR Clean-Up System	8 × 96 preps	99
A9345	Wizard® SV 96 PCR Clean-Up System	100 × 96 preps	99
AS1010	Maxwell® 16 Blood DNA Purification Kit	48 preps	85
AS1010	Maxwell® 16 Blood DNA Purification Kit	48 preps	87
AS1020	Maxwell® 16 Cell DNA Purification Kit	48 preps	85
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AS1030	Maxwell® 16 Tissue DNA Purification Kit	48 preps	85
AS1030	Maxwell® 16 Tissue DNA Purification Kit	48 preps	87
AS1040	DNA IQ™ Reference Sample Kit for Maxwell® 16	48 preps	118
AS1050	Maxwell® 16 Total RNA Purification Kit	48 preps	86
AS1050	Maxwell® 16 Total RNA Purification Kit	48 preps	87
AS1060	Maxwell® 16 Polyhistidine Protein Purification Kit	48 preps	226
AS1120	Maxwell® 16 Mouse Tail DNA Purification Kit	48 preps	85
AS1130	Maxwell® 16 FFPE Tissue LEV DNA Purification Kit	48 preps	85
AS1130	Maxwell® 16 FFPE Tissue LEV DNA Purification Kit	48 preps	87
AS1140	Maxwell® 16 Cell LEV DNA Purification Kit	48 preps	85
AS1140	Maxwell® 16 Cell LEV DNA Purification Kit	48 preps	87
AS1150	Maxwell® 16 Viral Total Nucleic Acid Purification Kit	48 preps	87
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AS1201	Maxwell® 16 Cartridge Rack	1 each	84
AS1201	Maxwell® 16 Cartridge Rack	1 each	87
AS1202	Maxwell® 16 Magnetic Elution Rack	1 each	84
AS1220	Maxwell® 16 Tissue LEV Total RNA Purification Kit	48 preps	86
AS1220	Maxwell® 16 Tissue LEV Total RNA Purification Kit	48 preps	87
AS1225	Maxwell® 16 Cell LEV Total RNA Purification Kit	48 preps	86
AS1225	Maxwell® 16 Cell LEV Total RNA Purification Kit	48 preps	87
AS1240	DNA IQ™ Casework Pro Kit for Maxwell® 16	48 preps	118
AS1250	Maxwell® 16 LEV Hardware Kit	1 each	84
AS1250	Maxwell® 16 LEV Hardware Kit	1 each	87
AS1251	Maxwell® 16 LEV Cartridge Rack	1 each	84
AS1251	Maxwell® 16 LEV Cartridge Rack	1 each	87
AS1261	Maxwell® 16 LEV Magnet	1 each	84
AS1261	Maxwell® 16 LEV Magnet	1 each	85
AS1261	Maxwell® 16 LEV Magnet	1 each	87
AS1290	Maxwell® 16 LEV Blood DNA Kit	48 preps	85
AS1290	Maxwell® 16 LEV Blood DNA Kit	48 preps	87
AS2000	Maxwell® 16 Instrument	1 each	84
AS2000	Maxwell® 16 Instrument	1 each	85
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AS3000	Maxwell® 16 MDx Instrument	1 each	85
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AS3060	Maxwell® 16 Forensic Instrument	1 each	118
AS3060	Maxwell® 16 Forensic Instrument	1 each	118
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B1001	StemElite™ Gene Expression System	100 qPCR reactions	9
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B1002	StemElite™ Gene Expression System Plus	100 qPCR reactions + 50 RT reactions	10
B1002	StemElite™ Gene Expression System Plus	100 qPCR reactions + 50 RT reactions	10
B1002	StemElite™ Gene Expression System Plus	100 qPCR reactions + 50 RT reactions	11
B1002	StemElite™ Gene Expression System Plus	100 qPCR reactions + 50 RT reactions	11
B1011	StemElite™ NANOG/GAPDH Primer Pair (20X)	100 µl	9
B1021	StemElite™ SOX2/GAPDH Primer Pair (20X)	100 µl	9
B1031	StemElite™ POU5F1/GAPDH Primer Pair (20X)	100 µl	9
B1041	StemElite™ LIN28/GAPDH Primer Pair (20X)	100 µl	9
B1051	StemElite™ KLF4/GAPDH Primer Pair (20X)	100 µl	9
B1061	StemElite™ MYC/GAPDH Primer Pair (20X)	100 µl	9
B1071	StemElite™ NPPA/GAPDH Primer Pair (20X)	100 µl	10
B1081	StemElite™ MYL7/GAPDH Primer Pair (20X)	100 µl	10
B1091	StemElite™ MYL2/GAPDH Primer Pair (20X)	100 µl	10
B1101	StemElite™ MYH6/GAPDH Primer Pair (20X)	100 µl	10
B1111	StemElite™ MYH7/GAPDH Primer Pair (20X)	100 µl	10
B1121	StemElite™ NKX2-5/GAPDH Primer Pair (20X)	100 µl	10
B1131	StemElite™ TNNT2/GAPDH Primer Pair (20X)	100 µl	10
B1141	StemElite™ TNNI3/GAPDH Primer Pair (20X)	100 µl	10
B1151	StemElite™ MEF2C/GAPDH Primer Pair (20X)	100 µl	10
B1161	StemElite™ PLN/GAPDH Primer Pair (20X)	100 µl	10
B1171	StemElite™ GATA4/GAPDH Primer Pair (20X)	100 µl	10
B1301	StemElite™ HNF4A/GAPDH Primer Pair (20X)	100 µl	10
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B1321	StemElite™ PDX1/GAPDH Primer Pair (20X)	100 µl	10
B1331	StemElite™ INS/GAPDH Primer Pair (20X)	100 µl	10
B1341	StemElite™ FOXA2/GAPDH Primer Pair (20X)	100 µl	11
B1351	StemElite™ SOX17/GAPDH Primer Pair (20X)	100 µl	11
B1361	StemElite™ GATA6/GAPDH Primer Pair (20X)	100 µl	11
B1371	StemElite™ Mus-Nanog/Actb Primer Pair (20X)	100 µl	11
B1381	StemElite™ Mus-Sox2/Actb Primer Pair (20X)	100 µl	11
B1391	StemElite™ Mus-Pou5f1/Actb Primer Pair (20X)	100 µl	11
B1401	StemElite™ Mus-Lin28/Actb Primer Pair (20X)	100 µl	11
B1411	StemElite™ Mus-Klf4/Actb Primer Pair (20X)	100 µl	11
B1421	StemElite™ Mus-Myc/Actb Primer Pair (20X)	100 µl	11
C1101	Oligo(dT)$_{15}$ Primer	20 µg	16
C1101	Oligo(dT)$_{15}$ Primer	20 µg	16
C1101	Oligo(dT)$_{15}$ Primer	20 µg	256
C1141	PCR Nucleotide Mix	200 µl	16
C1145	PCR Nucleotide Mix	1,000 µl	16
C1181	Random Primers	20 µg	16
C1181	Random Primers	20 µg	16
C1181	Random Primers	20 µg	256
C1263	T4 DNA Ligase Buffer Pack	1.5ml (3 × 500 µl)	257
C1281	Spin Columns	10 each	16
C1291	EcoRI Adaptors	150 pmol	16
C1291	EcoRI Adaptors	150 pmol	16
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C4360	Universal RiboClone® cDNA Synthesis System	1 system	16
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C8790	GeneClip™ U1 Hairpin Cloning System—hMGFP	1 system	56
C9331	pFN10A (ACT) Flexi® Vector	20 µg	240
C9341	pFN11A (BIND) Flexi® Vector	20 µg	240
C9351	pGL4.31[*luc2P/GAL4*UAS/Hygro] Vector	20 µg	147
C9351	pGL4.31[*luc2P/GAL4*UAS/Hygro] Vector	20 µg	240
C9360	CheckMate™/Flexi® Vector Mammalian Two-Hybrid System	1 each	240
C9370	CheckMate™ Positive Control Vectors	1 set	240
C9380	CheckMate™ Negative Control Vectors	1 set	240
C9421	pReg neo Vector	20 µg	224
C9431	pF12A RM Flexi® Vector	20 µg	224
C9441	pF12K RM Flexi® Vector	20 µg	224
C9451	Coumermycin A1	5 mg	224
C9461	Novobiocin Sodium Salt	1 g	224
C9470	Regulated Mammalian Expression System	1 system	224
CD4002	20-Position Microcentrifuge Tube Magnetic Separator	1 each	115
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D1511	pBR322 Vector	10 µg	270
D1521	Unmethylated Lambda DNA	250 µg	270
D1531	ΦX174, RF DNA	50 µg	271
D1811	Herring Sperm DNA	10 mg	270
D1815	Herring Sperm DNA	100 mg	270
D1816	Herring Sperm DNA	500 mg	270
DC1000	Plexor® HY System	800 reactions	121
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DC6301	CSF1PO, TPOX, TH01, vWA Multiplex (Fluorescein)	100 reactions	132
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DC6450	*GenePrint*® SilverSTR® III System (D7S820, D13S317, D16S539)	400 reactions	133
DC6451	*GenePrint*® SilverSTR® III System (D7S820, D13S317, D16S539)	100 reactions	133
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DC6501	PowerPlex® 1.1 and 2.1 Systems	100 reactions	129
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DC6531	PowerPlex® 16 System	100 reactions	123
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DD2011	K562 DNA High Molecular Weight	30 µg	130
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DG2101	CTT Allelic Ladder Mix	150 µl	133
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DG2131	FFFL Allelic Ladder Mix (Fluorescein)	150 µl	132
DG2141	FFv Allelic Ladder Mix	150 µl	133
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DM2411	Gold ST★R 10X Buffer	1.2 ml	134
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DV4351	Blue Dextran Loading Solution	3 × 1 ml	123
DV4351	Blue Dextran Loading Solution	3 × 1 ml	125
DV4351	Blue Dextran Loading Solution	3 × 1 ml	134
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E1051	Chloramphenicol Acetyltransferase	100 u	161
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E1171	pGloSensor™-20F cAMP Plasmid	20 µg	39
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E1310	pGL4.50[*luc2*/CMV/Hygro] Vector	20 µg	147
E1320	pGL4.51[*luc2*/CMV/Neo] Vector	20 µg	145
E1320	pGL4.51[*luc2*/CMV/Neo] Vector	20 µg	147
E1330	pmirGLO Dual-Luciferase miRNA Target Expression Vector	20 µg	148
E1340	pGL4.33[*luc2P*/SRE/Hygro] Vector	20 µg	147
E1350	pGL4.34[*luc2P*/SRF-RE/Hygro] Vector	20 µg	147
E1360	pGL4.36[*luc2P*/MMTV/Hygro] Vector	20 µg	147
E1360	pGL4.36[*luc2P*/MMTV/Hygro] Vector	20 µg	148
E1370	pGL4.35[*luc2P*/9X*GAL4*UAS/Hygro] Vector	20 µg	147
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E1380	pFN26A (BIND) *hRluc*-neo Flexi® Vector	20 µg	148
E1390	pBIND-ERα Vector	20 µg	148
E1411	pCBR-Basic Vector	20 µg	149
E1421	pCBR-Control Vector	20 µg	149
E1431	pCBG68-Basic Vector	20 µg	149
E1441	pCBG68-Control Vector	20 µg	149
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E1605	Beetle Luciferin, Potassium Salt	1 g	142
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E1621	pGL2-Enhancer Vector	20 µg	151
E1631	pGL2-Promoter Vector	20 µg	151
E1641	pGL2-Basic Vector	20 µg	151
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E1702	QuantiLum® Recombinant Luciferase	5 mg	142
E1741	pGL3-Control Vector	20 µg	150
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E1851	pCAT®3-Control Vector	20 µg	161
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E1881	pCAT®3-Enhancer Vector	20 µg	161
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E1960	Dual-Luciferase® Reporter Assay System 10-Pack	1,000 assays	137
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E2241	pRL-TK Vector	20 µg	150
E2261	pRL-CMV Vector	20 µg	150
E2271	pRL-null Vector	20 µg	150
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E2510	Steady-Glo® Luciferase Assay System	10 ml	141
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E3211	AP2 Consensus Oligonucleotide	175 pmol	189
E3212	AP2 Consensus Oligonucleotide	35 pmol	189
E3221	TFIID Consensus Oligonucleotide	175 pmol	189
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E3241	OCT1 Consensus Oligonucleotide	175 pmol	189
E3242	OCT1 Consensus Oligonucleotide	35 pmol	189
E3281	CREB Consensus Oligonucleotide	175 pmol	189
E3282	CREB Consensus Oligonucleotide	35 pmol	189
E3291	NF-κB Consensus Oligonucleotide	175 pmol	189
E3292	NF-κB Consensus Oligonucleotide	35 pmol	189
E3300	Gel Shift Assay System	100 reactions	190
E3511	ΦX174 DNA/HinfI Dephosphorylated Markers	2.5 µg	208
E3521	HeLaScribe® Nuclear Extract, Gel Shift Assay Grade	3 × 40 µl	190
E3581	Gel Shift Binding 5X Buffer	5 × 200 µl	190
E3621	HeLaScribe® Nuclear Extract Positive Control DNA	300 ng	189
E3770	rhNF-κB (p50)	50 gsu	188
E3790	rhTFIIB	50 gsu	188
E3971	Reporter Lysis 5X Buffer	30 ml	142
E3971	Reporter Lysis 5X Buffer	30 ml	159
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E4030	Luciferase Assay System with Reporter Lysis Buffer	100 assays	142
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E4780	Beta-Glo® Assay System	10 × 100 ml	160
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E5401	GloMax® Injector Tips Replacement	1 each	157
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E6011	rATP, 100mM	400 µl	189
E6021	rUTP, 100mM	400 µl	189
E6031	rGTP, 100mM	400 µl	189
E6041	rCTP, 100mM	400 µl	189
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E6090	QuantiFluor™-ST Handheld Fluorometer with UV/Blue Channels	1 each	158
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E6091	Minicell Borosilicate Glass Cuvettes	400 each	158
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E6093	10 × 10mm Square Methacrylate Cuvette (3.5ml capacity)	100 each	156
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E6111	QuantiFluor™-P Minicell Adapter Kit (for measuring 75–250µl of sample)	400 each	158
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E6113	QuantiFluor™-ST Solid Standard	1 each	158
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E6482	EnduRen™ Live Cell Substrate	3.4 mg	143
E6485	EnduRen™ Live Cell Substrate	34 mg	143
E6491	ViviRen™ Live Cell Substrate	0.37 mg	144
E6492	ViviRen™ Live Cell Substrate	3.7 mg	144
E6495	ViviRen™ Live Cell Substrate	37 mg	144
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E6541	GloMax® 96 Tubing Replacement Kit for Injectors	1 each	157
E6551	Luciferin-EF™	25 mg	27
E6552	Luciferin-EF™	250 mg	27
E6651	pGL4.10[*luc2*] Vector	20 µg	146
E6661	pGL4.11[*luc2P*] Vector	20 µg	146
E6671	pGL4.12[*luc2CP*] Vector	20 µg	146
E6681	pGL4.13[*luc2*/SV40] Vector	20 µg	146
E6691	pGL4.14[*luc2*/Hygro] Vector	20 µg	146
E6701	pGL4.15[*luc2P*/Hygro] Vector	20 µg	146
E6711	pGL4.16[*luc2CP*/Hygro] Vector	20 µg	146
E6721	pGL4.17[*luc2*/Neo] Vector	20 µg	146
E6731	pGL4.18[*luc2P*/Neo] Vector	20 µg	146
E6741	pGL4.19[*luc2CP*/Neo] Vector	20 µg	146
E6751	pGL4.20[*luc2*/Puro] Vector	20 µg	146
E6761	pGL4.21[*luc2P*/Puro] Vector	20 µg	146
E6771	pGL4.22[*luc2CP*/Puro] Vector	20 µg	146
E6881	pGL4.70[*hRluc*] Vector	20 µg	147
E6891	pGL4.71[*hRlucP*] Vector	20 µg	147
E6901	pGL4.72[*hRlucCP*] Vector	20 µg	147
E6911	pGL4.73[*hRluc*/SV40] Vector	20 µg	147
E6921	pGL4.74[*hRluc*/TK] Vector	20 µg	147
E6931	pGL4.75[*hRluc*/CMV] Vector	20 µg	147
E6941	pGL4.76[*hRluc*/Hygro] Vector	20 µg	147
E6951	pGL4.77[*hRlucP*/Hygro] Vector	20 µg	147
E6961	pGL4.78[*hRlucCP*/Hygro] Vector	20 µg	147
E6971	pGL4.79[*hRluc*/Neo] Vector	20 µg	147
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E7071	Single Injector System for GloMax®-Multi Detection System	1 each	154
E7071	Single Injector System for GloMax®-Multi Detection System	1 each	155
E7081	Dual Injector System for GloMax®-Multi Detection System	1 each	154
E7081	Dual Injector System for GloMax®-Multi Detection System	1 each	155
E7501	pGL4.82[*hRluc*/Puro] Vector	20 µg	147
E7511	pGL4.83[*hRlucP*/Puro] Vector	20 µg	147
E7521	pGL4.84[*hRlucCP*/Puro] Vector	20 µg	147
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E8051	GloMax®-Multi+ Fluorescence Module	1 each	154
E8061	GloMax®-Multi+ Visible Absorbance Module	1 each	154
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E8081	DB-15 Communication Cable	1 each	154
E8411	pGL4.23[*luc2*/minP] Vector	20 µg	147
E8421	pGL4.24[*luc2P*/minP] Vector	20 µg	147
E8431	pGL4.25[*luc2CP*/minP] Vector	20 µg	147
E8441	pGL4.26[*luc2*/minP/Hygro] Vector	20 µg	147
E8451	pGL4.27[*luc2P*/minP/Hygro] Vector	20 µg	147
E8461	pGL4.28[*luc2CP*/minP/Hygro] Vector	20 µg	147
E8471	pGL4.29[*luc2P*/CRE/Hygro] Vector	20 µg	147
E8481	pGL4.30[*luc2P*/NFAT-RE/Hygro] Vector	20 µg	147
E8491	pGL4.32[*luc2P*/NF-κB-RE/Hygro] Vector	20 µg	147
E8500	GloResponse™ CRE-*luc2P* HEK293 Cell Line	2 vials	139
E8510	GloResponse™ NFAT-RE-*luc2P* HEK293 Cell Line	2 vials	139
E8520	GloResponse™ NF-κB-RE-*luc2P* HEK293 Cell Line	2 vials	139
E8530	GloResponse™ 9X*GAL4*UAS-*luc2P* HEK293 Cell Line	2 vials	139
E8530	GloResponse™ 9X*GAL4*UAS-*luc2P* HEK293 Cell Line	2 vials	148
E8916	GloMax®-Multi Detection System External PC Connect Kit	1 each	155
E8917	GloMax®-Multi Optical Kit AFC	1 each	154
E8917	GloMax®-Multi Optical Kit AFC	1 each	155
E8921	GloMax®-Multi Optical Kit Blue	1 each	154
E8921	GloMax®-Multi Optical Kit Blue	1 each	155
E8922	GloMax®-Multi Optical Kit UV	1 each	154
E8922	GloMax®-Multi Optical Kit UV	1 each	155
E8923	GloMax®-Multi Optical Kit Green	1 each	154
E8923	GloMax®-Multi Optical Kit Green	1 each	155
E8924	GloMax®-Multi Optical Kit Red	1 each	154
E8924	GloMax®-Multi Optical Kit Red	1 each	155
E8925	Injector Inlet Tubing Assembly	1 set	154
E8925	Injector Inlet Tubing Assembly	1 set	155
E8926	Injector Outlet Tubing Assembly for Single-Injector System	1 each	154
E8926	Injector Outlet Tubing Assembly for Single-Injector System	1 each	155
E8927	Injector Outlet Tubing Assembly for Dual-Injector System	1 each	154
E8927	Injector Outlet Tubing Assembly for Dual-Injector System	1 each	155
E8928	Waste Collection Tray	1 each	154
E8928	Waste Collection Tray	1 each	155
E8929	GloMax®-Multi Detection System 490nm Absorbance Filter Set	1 each	154
E8929	GloMax®-Multi Detection System 490nm Absorbance Filter Set	1 each	155
E8935	USB Flash Drive, 2.0, 2GB	1 each	154
E8935	USB Flash Drive, 2.0, 2GB	1 each	155
E8942	GloMax®-Multi+ Detection System Power Supply—24V, 150W	1 each	154
E8943	GloMax®-Multi+ Detection System 6-384 Well Plate Adapter	1 each	154
E8944	GloMax®-Multi+ Detection System 96 Well Optical Crosstalk Mask	1 each	154
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FF3750	Wizard® Magnetic DNA Purification System for Food	200 preps	106
FF3751	Wizard® Magnetic DNA Purification System for Food	400 preps	106
FF3760	Wizard® Magnetic 96 DNA Plant System	2 × 96 preps	106
FF3761	Wizard® Magnetic 96 DNA Plant System	4 × 96 preps	106
G0940	Caspase-Glo® 2 Assay	10 ml	69
G0941	Caspase-Glo® 2 Assay	50 ml	69
G0970	Caspase-Glo® 6 Assay	10 ml	70
G0971	Caspase-Glo® 6 Assay	50 ml	70
G1111	CellTiter 96® AQ_{ueous} MTS Reagent Powder	1 g	66
G1112	CellTiter 96® AQ_{ueous} MTS Reagent Powder	250 mg	66
G1131	Anti-NGF mAb	100 µg	177
G1132	Anti-NGF mAb	20 µg	177
G1161	Chicken IgY, Control Immunoglobulin	1 mg	181
G1180	Proteasome-Glo™ 3-Substrate Cell-Based Assay System	10 ml	77
G1200	Proteasome-Glo™ 3-Substrate Cell-Based Assay System	50 ml	77
G1221	Anti-TGFβ_1 pAb	100 µg	179
G1291	TGFβ Sample 10X Buffer	20 ml	169
G1321	pFC17K HaloTag® CMV*d3* Flexi® Vector	20 µg	166
G1351	Anti-Chicken IgY, HRP Conjugate	300 µl	181
G1471	Human Genomic DNA: Male	100 µg	210
G1491	rhBDNF	5 µg	54
G1521	Human Genomic DNA: Female	100 µg	210
G1531	EGGstract® IgY Purification System	25 isolations	182
G1551	pFC17A HaloTag® CMV*d3* Flexi® Vector	20 µg	166
G1561	Anti-TrkB In pAb	100 µg	179
G1571	pFC16K HaloTag® CMV*d2* Flexi® Vector	20 µg	166
G1591	pFC16A HaloTag® CMV*d2* Flexi® Vector	20 µg	166
G1601	pFC15K HaloTag® CMV*d1* Flexi® Vector	20 µg	166
G1611	pFC15A HaloTag® CMV*d1* Flexi® Vector	20 µg	166
G1631	Anti-Rat CNTF pAb	200 µg	175
G1641	Anti-Human BDNF pAb	200 µg	174
G1651	Anti-Human NT-3 pAb	200 µg	177
G1681	pFC20A HaloTag® T7 SP6 Flexi® Vector	20 µg	166
G1691	pFC20K HaloTag® T7 SP6 Flexi® Vector	20 µg	166
G1711	Lambda DNA/HindIII Markers	100µg (200 lanes)	207
G1721	Lambda DNA/EcoRI Markers	100µg (200 lanes)	207
G1731	Lambda DNA/EcoRI + HindIII Markers	100µg (200 lanes)	207
G1741	pGEM® DNA Markers	50µg (50 lanes)	207
G1751	ΦX174 DNA/HinfI Markers	50µg (50 lanes)	207
G1761	ΦX174 DNA/HaeIII Markers	50µg (50 lanes)	207
G1780	CytoTox 96® Non-Radioactive Cytotoxicity Assay	1,000 assays	68
G1841	pFN19K HaloTag® T7 SP6 Flexi® Vector	20 µg	166
G1881	Blue/Orange Loading Dye, 6X	3ml (3 × 1 ml)	210
G1891	pFN19A HaloTag® T7 SP6 Flexi® Vector	20 µg	166
G1911	HaloLink™ Resin	2ml (0.5ml settled resin)	238
G1912	HaloLink™ Resin	5ml (1.25ml settled resin)	238
G1914	HaloLink™ Resin	10ml (settled resin)	238
G2101	100bp DNA Ladder	250µl (50 lanes)	206
G2610	EGGstract® IgY Purification System	6 isolations	182
G2681	pFN18K HaloTag® T7 Flexi® Vector	20 µg	166
G2751	pFN18A HaloTag® T7 Flexi® Vector	20 µg	166
G2781	rhGDNF	5 µg	54
G2791	Anti-Human GDNF pAb	200 µg	176
G2821	pFN21A HaloTag® CMV Flexi® Vector	20 µg	166
G2831	pFN21K HaloTag® CMV Flexi® Vector	20 µg	166
G2841	pFN22A HaloTag® CMV*d1* Flexi® Vector	20 µg	166
G2851	pFN22K HaloTag® CMV*d1* Flexi® Vector	20 µg	166
G2861	pFN23A HaloTag® CMV*d2* Flexi® Vector	20 µg	166
G2871	pFN23K HaloTag® CMV*d2* Flexi® Vector	20 µg	166

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G2881	pFN24A HaloTag® CMV*d3* Flexi® Vector	20 µg	166
G2891	Anti-Chicken IgY, Biotin Conjugate	500 µg	181
G2930	Griess Reagent System	1,000 assays	55
G2981	pFN24K HaloTag® CMV*d3* Flexi® Vector	20 µg	166
G3011	ProMega-Markers® Lambda Ladders	40–60 lanes	209
G3041	Human Genomic DNA	100 µg	210
G3091	Mouse Genomic DNA	100 µg	210
G3161	PCR Markers	250µl (50 lanes)	206
G3191	RNA Markers	50 µl	209
G3231	Anti-Human p75 pAb	200 µg	178
G3250	DeadEnd™ Fluorometric TUNEL System	60 reactions	74
G3311	Block & Sample 5X Buffer	54 ml	168
G3580	CellTiter 96® AQ_{ueous} One Solution Cell Proliferation Assay	1,000 assays	65
G3581	CellTiter 96® AQ_{ueous} One Solution Cell Proliferation Assay	5,000 assays	65
G3582	CellTiter 96® AQ_{ueous} One Solution Cell Proliferation Assay	200 assays	65
G3780	HaloTag® Flexi® Vectors—CMV Deletion Series Sample Pack	9 × 2 µg	166
G4000	CellTiter 96® Non-Radioactive Cell Proliferation Assay	1,000 assays	66
G4100	CellTiter 96® Non-Radioactive Cell Proliferation Assay	5,000 assays	66
G4471	10bp DNA Step Ladder	32.5µg (50 lanes)	205
G4481	Anti-VAChT pAb	100 µg	180
G4491	HaloTag® Standard Protein	30 µg	234
G4511	25bp DNA Step Ladder	100µg (55 lanes)	205
G4521	50bp DNA Step Ladder	90µg (52 lanes)	205
G5021	rhEGF	100 µg	54
G5071	rhFGF, Basic	25 µg	54
G5111	rhIGF-I	25 µg	54
G5141	mNGF, 2.5S	100 µg	54
G5241	rhTNFα	10 µg	54
G5381	Vitronectin, Human	100 µg	54
G5421	CellTiter 96® AQ_{ueous} Non-Radioactive Cell Proliferation Assay	1,000 assays	66
G5430	CellTiter 96® AQ_{ueous} Non-Radioactive Cell Proliferation Assay	5,000 assays	66
G5440	CellTiter 96® AQ_{ueous} Non-Radioactive Cell Proliferation Assay	50,000 assays	66
G5601	Anti-GFAP pAb	100 µg	176
G5631	rhLung β Tryptase	100 µg	79
G5711	1kb DNA Ladder	500µl (100 lanes)	206
G5740	β-Actin Primer Pair	20 reactions	15
G5770	CNTF Primer Pair	20 reactions	15
G5961	Caspase Inhibitor Ac-DEVD-CHO	100 µl	74
G6050	HaloTag® Cloning Starter System	1 each	164
G6080	CellTiter-Fluor™ Cell Viability Assay	10 ml	62
G6081	CellTiter-Fluor™ Cell Viability Assay	5 × 10 ml	62
G6082	CellTiter-Fluor™ Cell Viability Assay	2 × 50 ml	62
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G6261	DUB-Glo™ Protease Assay (DUB/SENP/NEDP)	50 ml	75
G6270	HaloTag® Protein Purification System Sample Pack	1 each	225
G6280	HaloTag® Protein Purification System	1 each	225
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G6320	ApoTox-Glo™ Triplex Assay	10 ml	59
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G6410	ApoLive-Glo™ Multiplex Assay	10 ml	59
G6411	ApoLive-Glo™ Multiplex Assay	5 × 10 ml	59
G6500	HaloTag® Mammalian Pull-Down and Labeling System	24 reactions	239
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G6521	Protease Inhibitor Cocktail, 50X	1 ml	239
G6591	HaloTag® Control Vector	20 µg	239
G6801	NT-3 Primer Pair	20 reactions	15
G6861	p75 Primer Pair	20 reactions	15
G6941	1kb DNA Step Ladder	90µg (300 lanes)	205
G6951	100bp DNA Step Ladder	100µg (100 lanes)	205
G6961	200bp DNA Step Ladder	100µg (100 lanes)	205
G7061	rhSkin β Tryptase	100 µg	79
G7121	Anti-βIII Tubulin mAb	100 µg	179

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G7232	Caspase Inhibitor Z-VAD-FMK, 20mM	125 µl	74
G7341	Anti-PARP p85 Fragment pAb	50 µl	178
G7351	CaspACE™ Assay System, Colorimetric	50 assays	73
G7360	DeadEnd™ Colorimetric TUNEL System	20 reactions	73
G7431	TMB One Solution	100 ml	182
G7441	Anti-pS^{473} Akt pAb	40 µl	169
G7451	Anti-Luciferase pAb	200 µg	176
G7461	CaspACE™ FITC-VAD-FMK In Situ Marker	50 µl	73
G7462	CaspACE™ FITC-VAD-FMK In Situ Marker	125 µl	73
G7481	Anti-ACTIVE® Caspase-3 pAb	50 µl	174
G7511	BenchTop ΦX174 DNA/HaeIII Markers	250µl (50 lanes)	204
G7521	BenchTop pGEM® DNA Markers	250µl (50 lanes)	204
G7531	BenchTop PCR Markers	300µl (50 lanes)	204
G7541	BenchTop 1kb DNA Ladder	600µl (100 lanes)	204
G7570	CellTiter-Glo® Luminescent Cell Viability Assay	10 ml	63
G7571	CellTiter-Glo® Luminescent Cell Viability Assay	10 × 10 ml	63
G7572	CellTiter-Glo® Luminescent Cell Viability Assay	100 ml	63
G7573	CellTiter-Glo® Luminescent Cell Viability Assay	10 × 100 ml	63
G7590	TGFβ$_1$ E$_{max}$® ImmunoAssay System	2 × 96 wells	168
G7591	TGFβ$_1$ E$_{max}$® ImmunoAssay System	5 × 96 wells	168
G7600	TGFβ$_2$ E$_{max}$® ImmunoAssay System	5 × 96 wells	169
G7610	BDNF E$_{max}$® ImmunoAssay System	2 × 96 wells	167
G7611	BDNF E$_{max}$® ImmunoAssay System	5 × 96 wells	167
G7620	GDNF E$_{max}$® ImmunoAssay System	2 × 96 wells	167
G7621	GDNF E$_{max}$® ImmunoAssay System	5 × 96 wells	167
G7630	NGF E$_{max}$® ImmunoAssay System	2 × 96 wells	168
G7631	NGF E$_{max}$® ImmunoAssay System	5 × 96 wells	168
G7640	NT-3 E$_{max}$® ImmunoAssay System	2 × 96 wells	168
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G7791	Apo-ONE® Homogeneous Caspase-3/7 Assay	100 ml	72
G7792	Apo-ONE® Homogeneous Caspase-3/7 Assay	1 ml	72
G7890	CytoTox-ONE™ Homogeneous Membrane Integrity Assay	200–800 assays	67
G7891	CytoTox-ONE™ Homogeneous Membrane Integrity Assay	1,000–4,000 assays	67
G7892	CytoTox-ONE™ Homogeneous Membrane Integrity Assay, HTP	1,000–4,000 assays	67
G8080	CellTiter-Blue® Cell Viability Assay	20 ml	67
G8081	CellTiter-Blue® Cell Viability Assay	100 ml	67
G8082	CellTiter-Blue® Cell Viability Assay	10 × 100 ml	67
G8090	Caspase-Glo® 3/7 Assay	2.5 ml	70
G8091	Caspase-Glo® 3/7 Assay	10 ml	70
G8092	Caspase-Glo® 3/7 Assay	100 ml	70
G8093	Caspase-Glo® 3/7 Assay	10 × 10 ml	70
G8200	Caspase-Glo® 8 Assay	2.5 ml	71
G8201	Caspase-Glo® 8 Assay	10 ml	71
G8202	Caspase-Glo® 8 Assay	100 ml	71
G8210	Caspase-Glo® 9 Assay	2.5 ml	71
G8211	Caspase-Glo® 9 Assay	10 ml	71
G8212	Caspase-Glo® 9 Assay	100 ml	71
G8230	BacTiter-Glo™ Microbial Cell Viability Assay	10 ml	64
G8231	BacTiter-Glo™ Microbial Cell Viability Assay	10 × 10 ml	64
G8232	BacTiter-Glo™ Microbial Cell Viability Assay	100 ml	64
G8233	BacTiter-Glo™ Microbial Cell Viability Assay	10 × 100 ml	64
G8252	HaloTag® TMR Ligand	15 µl	234
G8252	HaloTag® TMR Ligand	15 µl	238
G8291	BenchTop 100bp DNA Ladder	300µl (50 lanes)	204
G8350	DPPIV-Glo™ Protease Assay	10 ml	76
G8351	DPPIV-Glo™ Protease Assay	50 ml	76
G8501	Calpain-Glo™ Protease Assay	10 ml	78
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G8622	Proteasome-Glo™ Chymotrypsin-Like Assay	50 ml	76
G8631	Proteasome-Glo™ Trypsin-Like Assay	10 ml	76
G8632	Proteasome-Glo™ Trypsin-Like Assay	50 ml	76
G8641	Proteasome-Glo™ Caspase-Like Assay	10 ml	76
G8642	Proteasome-Glo™ Caspase-Like Assay	50 ml	76
G8660	Proteasome-Glo™ Chymotrypsin-Like Cell-Based Assay	10 ml	77
G8661	Proteasome-Glo™ Chymotrypsin-Like Cell-Based Assay	5 × 10 ml	77
G8662	Proteasome-Glo™ Chymotrypsin-Like Cell-Based Assay	2 × 50 ml	77
G8760	Proteasome-Glo™ Trypsin-Like Cell-Based Assay	10 ml	77
G8761	Proteasome-Glo™ Trypsin-Like Cell-Based Assay	5 × 10 ml	77
G8860	Proteasome-Glo™ Caspase-Like Cell-Based Assay	10 ml	77
G8861	Proteasome-Glo™ Caspase-Like Cell-Based Assay	5 × 10 ml	77
G9200	MultiTox-Fluor Multiplex Cytotoxicity Assay	10 ml	60
G9201	MultiTox-Fluor Multiplex Cytotoxicity Assay	5 × 10 ml	60
G9202	MultiTox-Fluor Multiplex Cytotoxicity Assay	2 × 50 ml	60
G9260	CytoTox-Fluor™ Cytotoxicity Assay	10 ml	62
G9261	CytoTox-Fluor™ Cytotoxicity Assay	5 × 10 ml	62
G9262	CytoTox-Fluor™ Cytotoxicity Assay	2 × 50 ml	62
G9270	MultiTox-Glo Multiplex Cytotoxicity Assay	10 ml	59
G9271	MultiTox-Glo Multiplex Cytotoxicity Assay	5 × 10 ml	59
G9272	MultiTox-Glo Multiplex Cytotoxicity Assay	2 × 50 ml	59
G9281	Anti-HaloTag® pAb	200 µg	225
G9290	CytoTox-Glo™ Cytotoxicity Assay	10 ml	61
G9291	CytoTox-Glo™ Cytotoxicity Assay	5 × 10 ml	61
G9292	CytoTox-Glo™ Cytotoxicity Assay	2 × 50 ml	61
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G9381	Mammalian Lysis Buffer	40 ml	239
G9410	HaloCHIP™ System	20 reactions	236
G9451	Protease-Glo™ Assay	1 each	75
G9461	pGloSensor™-10F Linear Vector	1 µg	75
G9500	Cell ID™ System	50 reactions	35
G9530	StemElite™ ID System	50 reactions	34
G9651	pFC14A HaloTag® CMV Flexi® Vector	20 µg	166
G9661	pFC14K HaloTag® CMV Flexi® Vector	20 µg	166
H5001	Boric Acid, Molecular Biology Grade	500 g	24
H5003	Boric Acid, Molecular Biology Grade	1 kg	24
H5031	EDTA, Disodium Salt, Molecular Biology Grade	100 g	25
H5032	EDTA, Disodium Salt, Molecular Biology Grade	500 g	25
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H5051	Formamide, Molecular Grade	100 ml	26
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H5114	Sodium Dodecyl Sulphate, Molecular Biology Grade (SDS)	500 g	29
H5115	Sodium Dodecyl Sulphate, Molecular Biology Grade (SDS)	1 kg	29
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H5273	Sodium Chloride, Molecular Biology Grade	1 kg	29
H5302	HEPES, Molecular Biology Grade (free acid)	100 g	27
H5303	HEPES, Molecular Biology Grade (free acid)	500 g	27
H5381	Guanidine-HCl, Molecular Biology Grade	100 g	27
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H5433	Glycerol, Molecular Biology Grade	1,000 ml	26
K9981	Bacterial Strain LE392, Glycerol Stock	500 µl	278
L1001	JM109 Competent Cells, $>10^7$cfu/µg	5 × 200 µl	240
L1001	JM109 Competent Cells, $>10^7$cfu/µg	5 × 200 µl	276
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L1030	*E. coli* S30 Extract System for Linear Templates	30 reactions	222
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L2011	HB101 Competent Cells, $>10^8$cfu/µg	5 × 200 µl	276
L2081	TNT® SP6 Quick Coupled Transcription/Translation System, Trial Size	5 reactions	217
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L3251	Wheat Germ Extract Plus	10 reactions	220
L4120	TNT® T3 Coupled Wheat Germ Extract System	40 reactions	218
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L4330	Rabbit Reticulocyte Lysate/ Wheat Germ Extract Combination System	24 reactions	220
L4380	Wheat Germ Extract	5 × 200 µl	220
L4461	Amino Acid Mixture, Complete	175 µl	223
L4471	Amino Acid Mixture Minus Cysteine	175 µl	223
L4540	Flexi® Rabbit Reticulocyte Lysate System	30 reactions	219
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L4600	TNT® SP6 Coupled Reticulocyte Lysate System	40 reactions	218
L4601	TNT® SP6 Coupled Reticulocyte Lysate System, Trial Size	8 reactions	218
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L4611	TNT® T7 Coupled Reticulocyte Lysate System, Trial Size	8 reactions	218
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L4741	Luciferase SP6 Control DNA	20 µg	219
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L4950	TNT® T3 Coupled Reticulocyte Lysate System	40 reactions	218
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L5001	FluoroTect™ $Green_{Lys}$ in vitro Translation Labeling System	40 reactions	230
L5010	TNT® T7/T3 Coupled Reticulocyte Lysate System	40 reactions	218
L5020	TNT® T7/SP6 Coupled Reticulocyte Lysate System	40 reactions	218
L5030	TNT® T7/SP6 Coupled Wheat Germ Extract System	40 reactions	218
L5040	TNT® T7/T3 Coupled Wheat Germ Extract System	40 reactions	218
L5061	Transcend™ tRNA	30 µl	229
L5070	Transcend™ Colorimetric Non-Radioactive Translation Detection System	30 reactions	229
L5080	Transcend™ Chemiluminescent Non-Radioactive Translation Detection System	30 reactions	229
L5511	Amino Acid Mixture Minus Methionine and Cysteine	175 µl	223
L5540	TNT® T7 Quick for PCR DNA	40 reactions	219
L5671	pF3A WG (BYDV) Flexi® Vector	20 µg	220
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M1705	M-MLV Reverse Transcriptase	50,000 u	256
M1741	Agar*ACE*™ Enzyme	25 u	260
M1743	Agar*ACE*™ Enzyme	500 u	260
M1794	T4 DNA Ligase (HC)	500 u (Weiss units)	257
M1801	T4 DNA Ligase	100 u (Weiss units)	257
M1804	T4 DNA Ligase	500 u (Weiss units)	257
M1811	Exonuclease III	5,000 u	259
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M1833	CIAP Buffer Pack	1.5ml (3 × 500 µl)	258
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M1893	Terminal Transferase Buffer Pack	3 × 500 µl	254
M1941	*Tfl* DNA Polymerase	100 u	6
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M2055	DNA Polymerase I	2,500 u	253
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M2105	*Tth* DNA Polymerase	500 u	15
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M2201	DNA Polymerase I Large (Klenow) Fragment	150 u	253
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M2825	Alkaline Phosphatase, Calf Intestinal (HC)	1,000 u	258
M2851	Topoisomerase I	200 u	260
M3001	GoTaq® DNA Polymerase	100 u	5
M3005	GoTaq® DNA Polymerase	500 u	5
M3008	GoTaq® DNA Polymerase	2,500 u	5
M3011	Single-Stranded DNA Binding Protein	100 µg	260
M3681	M-MLV Reverse Transcriptase, RNase H Minus, Point Mutant	2,500 u	257
M3682	M-MLV Reverse Transcriptase, RNase H Minus, Point Mutant	10,000 u	257
M3683	M-MLV Reverse Transcriptase, RNase H Minus, Point Mutant	50,000 u	257
M4101	T4 Polynucleotide Kinase	100 u	258
M4103	T4 Polynucleotide Kinase	1,000 u	258
M4211	T4 DNA Polymerase	100 u	254
M4215	T4 DNA Polymerase	500 u	254
M4261	RNase ONE™ Ribonuclease	1,000 u	260
M4265	RNase ONE™ Ribonuclease	5,000 u	260
M4281	Ribonuclease H	50 u	259
M4285	Ribonuclease H	250 u	259
M4311	Mung Bean Nuclease	2,000 u	259
M5001	GoTaq® Hot Start Polymerase	100 u	4
M5005	GoTaq® Hot Start Polymerase	500 u	4
M5006	GoTaq® Hot Start Polymerase	2,500 u	4
M5008	GoTaq® Hot Start Polymerase	10,000 u	4
M5101	AMV Reverse Transcriptase	300 u	255
M5108	AMV Reverse Transcriptase	1,000 u	255
M5122	GoTaq® Hot Start Green Master Mix	100 reactions	4
M5123	GoTaq® Hot Start Green Master Mix	1,000 reactions	4
M5132	GoTaq® Hot Start Colorless Master Mix	100 reactions	4
M5133	GoTaq® Hot Start Colorless Master Mix	1,000 reactions	4
M5301	M-MLV Reverse Transcriptase, RNase H Minus	10,000 u	256
M5313	M-MLV Reverse Transcriptase Buffer Pack	2 × 1 ml	256
M5661	*Taq*Bead™ Hot Start Polymerase, 1.25u/bead, Nonbarrier	100 reactions	4
M5761	S1 Nuclease	10,000 u	259
M6101	RQ1 RNase-Free DNase	1,000 u	187
M6101	RQ1 RNase-Free DNase	1,000 u	259
M7122	GoTaq® Green Master Mix	100 reactions	5
M7123	GoTaq® Green Master Mix	1,000 reactions	5
M7132	GoTaq® Colorless Master Mix	100 reactions	5
M7133	GoTaq® Colorless Master Mix	1,000 reactions	5
M7501	PCR Master Mix	10 reactions	6
M7502	PCR Master Mix	100 reactions	6
M7505	PCR Master Mix	1,000 reactions	6
M7660	GoTaq® PCR Core System I	200 reactions	6
M7665	GoTaq® PCR Core System II	200 reactions	6
M7741	*Pfu* DNA Polymerase	100 u	7
M7745	*Pfu* DNA Polymerase	500 u	7
M7911	5X Green GoTaq® Reaction Buffer	20 ml	5

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Cat.#	Product	Size	Page
M7921	5X Colorless GoTaq® Reaction Buffer	20 ml	5
M8221	LigaFast™ Rapid DNA Ligation System	30 reactions	257
M8225	LigaFast™ Rapid DNA Ligation System	150 reactions	257
M8291	GoTaq® Flexi DNA Polymerase	100 u	5
M8295	GoTaq® Flexi DNA Polymerase	500 u	5
M8296	GoTaq® Flexi DNA Polymerase	2,500u (5 × 500 u)	5
M8297	GoTaq® Flexi DNA Polymerase	5,000u (10 × 500 u)	5
M8298	GoTaq® Flexi DNA Polymerase	10,000u (20 × 500 u)	5
M8901	5X Colorless GoTaq® Flexi Reaction Buffer	20 ml	5
M8911	5X Green GoTaq® Flexi Reaction Buffer	20 ml	5
M9004	AMV Reverse Transcriptase (HC)	600 u	255
M9910	TSAP Thermosensitive Alkaline Phosphatase	100 units	258
MA1010	ISOQUANT® Isoaspartate Detection Kit	100 assays	184
MB1001	MagaZorb® DNA Mini-Prep Kit	50 preps	106
MB1004	MagaZorb® DNA Mini-Prep Kit	200 preps	106
MB1008	MagaZorb® DNA Mini-Prep Kit	800 preps	106
MB1101	MagaZorb® DNA Mini-Prep 96-Well Kit	200 preps	106
MB2001	MagaZorb® Total RNA Mini-Prep Kit	50 preps	113
MB2004	MagaZorb® Total RNA Mini-Prep Kit	200 preps	113
MC5008	Proteinase K (PK) Solution	16 ml	106
MD1360	MagneSil® Blood Genomic, Max Yield System	1 × 96 preps	104
MD1370	MagneSil® ONE, Fixed Yield Blood Genomic System	1 × 96 preps	103
MD1392	Lysis Buffer, Blood	160 ml	103
MD1401	Salt Wash, Blood	90 ml	104
MD1411	Alcohol Wash, Blood	70 ml	104
MD1412	Alcohol Wash, Blood	120 ml	103
MD1421	Elution Buffer, Blood	45 ml	103
MD1421	Elution Buffer, Blood	45 ml	104
MD1431	Anti-Foam Reagent	300 µl	103
MD1431	Anti-Foam Reagent	300 µl	104
MD1441	MagneSil® Paramagnetic Particles	25 ml	104
MD1451	MagneSil® PMPs—Fixed Yield	25 ml	103
MD1460	MagneSil® KF, Genomic System	200 preps	105
MD1471	MagneSil® KF, Paramagnetic Particles	40 ml	105
MD1490	MagneSil® Genomic, Fixed Tissue System	100 samples	104
MD1521	Lysis Buffer, KF	160 ml	105
MD1531	Y Chromosome Deletion Detection System, Version 2.0	25 reactions	197
MD1641	MSI Analysis System, Version 1.2	100 reactions (50 reaction pairs)	196
N2111	RNasin® Ribonuclease Inhibitor	2,500 u	262
N2115	RNasin® Ribonuclease Inhibitor	10,000 u	262
N2511	Recombinant RNasin® Ribonuclease Inhibitor	2,500 u	262
N2515	Recombinant RNasin® Ribonuclease Inhibitor	10,000 u	262
N2611	RNasin® Plus RNase Inhibitor	2,500 u	261
N2615	RNasin® Plus RNase Inhibitor	10,000 u	261
P1041	VivoGlo™ Luciferin, In Vivo Grade	50 mg	145
P1042	VivoGlo™ Luciferin, In Vivo Grade	250 mg	145
P1043	VivoGlo™ Luciferin, In Vivo Grade	1 g	145
P1061	VivoGlo™ Luciferin-β-Galactoside Substrate (6-O-β-galactopyranosyl luciferin)	50 mg	145
P1062	VivoGlo™ Luciferin-β-Galactoside Substrate (6-O-β-galactopyranosyl luciferin)	250 mg	145
P1081	SP6 RNA Polymerase	5,000 u	254
P1085	SP6 RNA Polymerase	1,000 u	254
P1111	EnduRen™ In Vivo *Renilla* Luciferase Substrate	0.34 mg	145
P1112	EnduRen™ In Vivo *Renilla* Luciferase Substrate	3.4 mg	145
P1121	Riboprobe® System Buffers	1 system	187
P1132	rATP, 10mM	0.5 ml	187
P1142	rCTP, 10mM	0.5 ml	187
P1152	rGTP, 10mM	0.5 ml	187
P1162	rUTP, 10mM	0.5 ml	187
P1171	DTT, Molecular Grade	100 µl	25
P1171	DTT, Molecular Grade	100 µl	187
P1181	Transcription Optimized 5X Buffer	200 µl	187
P1193	Nuclease-Free Water	50ml (2 × 25 ml)	27
P1193	Nuclease-Free Water	50ml (2 × 25 ml)	187
P1195	Nuclease-Free Water	150 ml	27
P1195	Nuclease-Free Water	150 ml	95
P1195	Nuclease-Free Water	150 ml	112

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Cat.#	Product	Size	Page
P1195	Nuclease-Free Water	150 ml	267
P1221	rATP, rCTP, rGTP, rUTP, each at 10mM in separate tubes	0.5 ml	187
P1231	ViviRen™ In Vivo *Renilla* Luciferase Substrate	0.37 mg	145
P1232	ViviRen™ In Vivo *Renilla* Luciferase Substrate	3.7 mg	145
P1241	pSP64 Poly(A) Vector	20 µg	276
P1280	RiboMAX™ Large Scale RNA Production System—SP6	1 system	186
P1300	RiboMAX™ Large Scale RNA Production System—T7	1 system	186
P1320	T7 RiboMAX™ Express Large Scale RNA Production System	1 system	186
P1420	Riboprobe® System—SP6	1 system	186
P1430	Riboprobe® System—T3	1 system	186
P1440	Riboprobe® System—T7	1 system	186
P1450	Riboprobe® Combination System—T3/T7 RNA Polymerase	1 system	187
P1460	Riboprobe® Combination System—SP6/T7 RNA Polymerase	1 system	187
P1621	Luciferin-4A	3 mg	80
P1651	Luciferin-4F2/3	3 mg	80
P1661	Luciferin-4F12	3 mg	80
P1671	Luciferin-2J2/4F12 (ester)	3 mg	80
P1700	T7 RiboMAX™ Express RNAi System	50 × 20µl reactions	56
P1711	Ribo m^7G Cap Analog	10 A_{254} units	188
P1712	Ribo m^7G Cap Analog	25 A_{254} units	188
P1721	Luciferin-NAT2	3 mg	80
P1731	Luciferin-MultiCYP (ester)	3 mg	80
P1741	Luciferin-3A7	3 mg	80
P1781	VivoGlo™ Caspase-3/7 Substrate (Z-DEVD-Aminoluciferin, Sodium Salt)	50 mg	145
P1782	VivoGlo™ Caspase-3/7 Substrate (Z-DEVD-Aminoluciferin, Sodium Salt)	5 × 50 mg	145
P2075	T7 RNA Polymerase	1,000 u	255
P2077	T7 RNA Polymerase	5,000 u	255
P2083	T3 RNA Polymerase	1,000 u	254
P2151	pGEM®-3Z Vector	20 µg	271
P2161	pGEM®-4Z Vector	20 µg	272
P2191	pSP72 Vector	20 µg	275
P2221	pSP73 Vector	20 µg	275
P2241	pGEM®-5Zf(+) Vector	20 µg	273
P2251	pGEM®-7Zf(+) Vector	20 µg	273
P2261	pGEM®-3Zf(–) Vector	20 µg	272
P2271	pGEM®-3Zf(+) Vector	20 µg	272
P2301	Bacterial Strain NM522, Glycerol Stock	500 µl	278
P2341	R408 Helper Phage DNA	10 µg	278
P2371	pGEM®-7Zf(–) Vector	20 µg	273
P2391	pGEM®-9Zf(–) Vector	20 µg	274
P2411	pGEM®-11Zf(+) Vector	20 µg	274
P2421	pGEM®-11Zf(–) Vector	20 µg	274
P2561	pGEM® Express Positive Control Template	10µg (2 × 5 µg)	188
P4024	T3 RNA Polymerase (HC)	2,500 u	254
P4074	T7 RNA Polymerase (HC)	10,000 u	255
P4084	SP6 RNA Polymerase (HC)	2,500 u	254
P9751	Bacterial Strain JM109, Glycerol Stock	500 µl	277
P9801	Bacterial Strain JM109(DE3), Glycerol Stock	500 µl	277
Q4132	SILVER SEQUENCE™ Staining Reagents	10 gels	134
Q4411	Automatic Processor Compatible (APC) Film	25 sheets	264
Q4412	Automatic Processor Compatible (APC) Film, Sample Size	6 sheets	264
Q4461	pTARGET™ Sequencing Primer	2 µg	264
Q5011	SP6 Promoter Primer	2 µg	264
Q5021	T7 Promoter Primer	2 µg	264
Q5391	pUC/M13 Primer, Forward (17mer)	2 µg	264
Q5401	pUC/M13 Primer, Reverse (17mer)	2 µg	264
Q5421	pUC/M13 Primer, Reverse (22mer)	2 µg	264
Q5601	pUC/M13 Primer, Forward (24mer)	2 µg	264
Q6131	Bacterial Strain ES1301 *mutS*, Glycerol Stock (noncompetent)	200 µl	277
Q6321	Bacterial Strain BMH 71-18 *mutS*, Glycerol Stock (noncompetent)	500 µl	277
Q6700	T7 EEV Promoter Primer	2 µg	264
Q9280	GeneEditor™ in vitro Site-Directed Mutagenesis System	30 reactions	265
Q9291	GeneEditor™ Antibiotic Selection Mix	20 ml	265
Q9301	Bottom Strand Selection Oligonucleotide	35 µl	266
Q9321	Top Strand Selection Oligonucleotide	35 µl	266
R3961	Bovine Serum Albumin, Acetylated	1 ml	24
R4014	EcoRI (HC)	25,000 u	246
R4017	EcoRI (HC)	50,000 u	246

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R4024	BamHI (HC)	12,500 u	244
R4027	BamHI (HC)	50,000 u	244
R4044	HindIII (HC)	25,000 u	247
R4047	HindIII (HC)	50,000 u	247
R4054	SalI (HC)	10,000 u	249
R4064	SacI (HC)	5,000 u	249
R4074	BglI (HC)	5,000 u	244
R4114	PstI (HC)	15,000 u	249
R4117	PstI (HC)	50,000 u	249
R4124	SmaI (HC)	5,000 u	250
R4144	SinI (HC)	1,000 u	250
R4154	TaqI (HC)	5,000 u	251
R4164	XhoI (HC)	15,000 u	251
R4174	HaeIII (HC)	12,500 u	246
R4184	XbaI (HC)	10,000 u	251
R4204	HinfI (HC)	5,000 u	247
R4214	ScaI (HC)	5,000 u	249
R4344	KpnI (HC)	12,500 u	247
R4354	EcoRV (HC)	10,000 u	246
R4364	ApaI (HC)	25,000 u	243
R4374	RsaI (HC)	5,000 u	249
R4394	SfiI (HC)	1,250 u	250
R4404	MspI (HC)	10,000 u	248
R4434	NotI (HC)	1,000 u	248
R4604	SspI (HC)	2,500 u	250
R4624	BbuI (HC)	1,000 u	244
R4954	EcoICRI (HC)	5,000 u	246
R5104	SgfI (HC)	1,250 u	250
R6011	EcoRI	5,000 u	246
R6017	EcoRI	15,000 u	246
R6021	BamHI	2,500 u	244
R6025	BamHI	12,500 u	244
R6031	HincII	200 u	246
R6035	HincII	1,000 u	246
R6037	HincII	5,000 u	246
R6041	HindIII	5,000 u	247
R6045	HindIII	15,000 u	247
R6051	SalI	2,000 u	249
R6055	SalI	10,000 u	249
R6061	SacI	1,000 u	249
R6065	SacI	5,000 u	249
R6071	BglI	1,000 u	244
R6077	BglI	5,000 u	244
R6081	BglII	500 u	244
R6085	BglII	2,500 u	244
R6087	BglII	10,000 u	244
R6091	AvaI	200 u	243
R6095	AvaI	1,000 u	243
R6111	PstI	3,000 u	249
R6115	PstI	15,000 u	249
R6121	SmaI	1,000 u	250
R6125	SmaI	5,000 u	250
R6131	AvaII	100 u	243
R6135	AvaII	1,000 u	243
R6151	TaqI	1,000 u	251
R6155	TaqI	10,000 u	251
R6161	XhoI	3,000 u	251
R6165	XhoI	10,000 u	251
R6171	HaeIII	2,500 u	246
R6175	HaeIII	10,000 u	246
R6181	XbaI	2,000 u	251
R6185	XbaI	10,000 u	251
R6191	Sau3AI	100 u	249
R6195	Sau3AI	500 u	249
R6201	HinfI	1,000 u	247
R6205	HinfI	5,000 u	247
R6211	ScaI	1,000 u	249
R6221	SacII	500 u	249
R6231	DpnI	200 u	246
R6241	CfoI	3,000 u	245
R6261	SphI	200 u	250
R6265	SphI	1,000 u	250
R6271	DraI	2,000 u	246
R6281	AluI	500 u	243
R6291	DdeI	200 u	245
R6295	DdeI	1,000 u	245
R6301	HpaI	100 u	247
R6305	HpaI	500 u	247
R6311	HpaII	1,000 u	247
R6315	HpaII	5,000 u	247
R6321	PvuI	100 u	249
R6325	PvuI	500 u	249
R6331	PvuII	1,000 u	249
R6335	PvuII	5,000 u	249
R6341	KpnI	2,500 u	247
R6345	KpnI	10,000 u	247
R6351	EcoRV	2,000 u	246
R6355	EcoRV	10,000 u	246
R6361	ApaI	5,000 u	243
R6371	RsaI	1,000 u	249
R6381	MluI	1,000 u	247
R6391	SfiI	250 u	250

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R6401	MspI	2,000 u	248
R6405	MspI	10,000 u	248
R6411	AccI	100 u	243
R6415	AccI	500 u	243
R6421	StuI	400 u	250
R6431	NotI	200 u	248
R6435	NotI	1,000 u	248
R6441	HhaI	1,000 u	246
R6471	BstXI	250 u	245
R6475	BstXI	1,000 u	245
R6481	StyI	2,000 u	250
R6491	XmaI	50 u	251
R6495	XmaI	250 u	251
R6501	NheI	250 u	248
R6505	NheI	1,250 u	248
R6513	NcoI	200 u	248
R6515	NcoI	1,000 u	248
R6531	NsiI	250 u	249
R6541	AatII	50 u	243
R6545	AatII	250 u	243
R6551	ClaI	500 u	245
R6555	ClaI	2,500 u	245
R6561	BanII	1,000 u	244
R6571	Csp45I	2,500 u	245
R6581	AccIII	200 u	243
R6591	SpeI	200 u	250
R6595	SpeI	1,000 u	250
R6601	SspI	500 u	250
R6621	BbuI	200 u	244
R6641	BstEII	2,000 u	245
R6651	BclI	1,000 u	244
R6661	HaeII	1,000 u	246
R6671	CspI	100 u	245
R6675	CspI	500 u	245
R6691	BalI	50 u	243
R6695	BalI	250 u	243
R6711	MboI	200 u	247
R6723	MboII	100 u	247
R6731	Eco47III	50 u	246
R6741	Bsp1286I	500 u	244
R6791	SnaBI	100 u	250
R6795	SnaBI	500 u	250
R6801	NdeI	500 u	248
R6811	XhoII	100 u	251
R6815	XhoII	500 u	251
R6821	Bsu36I	500 u	245
R6831	BssHII	100 u	245
R6835	BssHII	500 u	245
R6841	Tth111I	500 u	251
R6851	VspI	500 u	251
R6861	NarI	200 u	248
R6881	BstZI	500 u	245
R6891	BanI	200 u	244
R6921	Acc65I	1,500 u	243
R6931	BstOI	2,000 u	245
R6951	EcoICRI	1,000 u	246
R6991	BsaMI	500 u	244
R7011	Tru9I	200 u	251
R7021	MspA1I	1,000 u	248
R7031	I-PpoI	10,000 u	247
R7061	NciI	1,000 u	248
R7081	AccB7I	200 u	243
R7091	NruI	200 u	248
R7103	SgfI	250 u	250
R7131	NaeI	250 u	248
R7135	NaeI	1,000 u	248
R7141	Bst98I	500 u	245
R7151	Hsp92I	500 u	247
R7161	Hsp92II	1,000 u	247
R7241	BsrSI	500 u	244
R7251	AgeI	100 u	243
R7271	XmnI	500 u	251
R7273	XmnI	2,500 u	251
R7291	NdeII	200 u	248
R7295	NdeII	1,000 u	248
R9461	Bovine Serum Albumin, Acetylated	400 µl	24
R9921	4-CORE® Buffer Pack (Buffers A, B, C and D), 1ml each	4ml (4 × 1 ml)	252
R9991	MULTI-CORE™ Buffer Pack	3 × 1 ml	252
S1000	AttoPhos® AP Fluorescent Substrate System	3 × 36 mg	182
S1001	AttoPhos® AP Fluorescent Substrate System Trial Size	1 × 36 mg	182
S1011	AttoPhos® Substrate	36 mg	182
S1012	AttoPhos® Substrate	100 mg	182
S1013	AttoPhos® Substrate	1 g	182
S1021	AttoPhos® Buffer	60 ml	182
S1022	AttoPhos® Buffer	240 ml	182
S2001	Coelenterazine	250 µg	144
S2011	Coelenterazine-h	250 µg	144
S3721	Anti-Mouse IgG (H+L), AP Conjugate	100 µl	181

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S3731	Anti-Rabbit IgG (Fc), AP Conjugate	100 µl	181
S3761	*E. coli* Extract for Background Reduction	2 ml	182
S3771	BCIP/NBT Color Development Substrate	1.25/2.5 ml	24
S3821	Anti-Human IgG (H+L), AP Conjugate	100 µl	181
S3831	Anti-Rat IgG (H+L), AP Conjugate	100 µl	181
S3841	Western Blue® Stabilized Substrate for Alkaline Phosphatase	100 ml	32
SA1001	Maxwell® 16 Installation Qualification	1 each	88
SA1011	Maxwell® 16 Operational Qualification	1 each	88
SA1021	Maxwell® 16 Installation and Operational Qualification	1 each	88
SA2000	Maxwell® 16 Premier Warranty	1 each	88
SA2010	Maxwell® 16 Standard Service Agreement	1 each	88
SA2015	Maxwell® 16 Premier Service Agreement	1 each	88
SA2020	Maxwell® 16 Preventative Maintenance	1 each	88
SA3000	GloMax® 20/20 Base Instrument Service Agreement	1 each	158
SA3010	GloMax® 96 Base Instrument Service Agreement	1 each	157
SA3020	GloMax®-Multi Base Instrument Service Agreement	1 each	155
SA3030	GloMax®-Multi+ Base Instrument Service Agreement, 1 year	1 each	154
SA3040	GloMax® Injectors Service Agreement, 1 year	1 each	154
SA3040	GloMax® Injectors Service Agreement, 1 year	1 each	155
SA3040	GloMax® Injectors Service Agreement, 1 year	1 each	157
SA3040	GloMax® Injectors Service Agreement, 1 year	1 each	158
SA3060	QuantiFluor™ Service Agreement	1 each	158
SA3070	ReliaPrep™ LV 32 HSM Standard Service Agreement	1 each	101
SA3080	GloMax®-Multi Jr Service Agreement	1 each	156
U1100	Prime-a-Gene® Labeling System	30 reactions	267
U1151	Labeling 5X Buffer	300 µl	267
U1191	dUTP	40 µmol	17
U1201	dATP	40 µmol	17
U1202	dATP	200µmol (2 × 100 µmol)	17
U1205	dATP	25 µmol	17
U1211	dGTP	40 µmol	17
U1212	dGTP	200µmol (2 × 100 µmol)	17
U1215	dGTP	25 µmol	17
U1221	dCTP	40 µmol	17
U1222	dCTP	200µmol (2 × 100 µmol)	17
U1225	dCTP	25 µmol	17
U1231	dTTP	40 µmol	17
U1232	dTTP	200µmol (2 × 100 µmol)	17
U1235	dTTP	25 µmol	17
U1240	Set of dATP, dCTP, dGTP, dTTP	40µmol each	17
U1240	Set of dATP, dCTP, dGTP, dTTP	40µmol each	255
U1240	Set of dATP, dCTP, dGTP, dTTP	40µmol each	256
U1245	Set of dATP, dCTP, dGTP, dUTP	40µmol each	17
U1300	DNA Polymerase I Large (Klenow) Fragment Mini Kit	150 u	253
U1330	Set of dATP, dCTP, dGTP, dTTP	10µmol each	17
U1330	Set of dATP, dCTP, dGTP, dTTP	10µmol each	253
U1335	Set of dATP, dCTP, dGTP, dUTP	10µmol each	17
U1410	Set of dATP, dCTP, dGTP, dTTP	200µmol (2 × 100µmol each)	17
U1420	Set of dATP, dCTP, dGTP, dTTP	25 µmol each	17
U1511	dNTP Mix	200 µl	16
U1515	dNTP Mix	1,000 µl	16
U2010	DNA 5´ End-Labeling System	10 reactions	267
V1061	Chymotrypsin, Sequencing Grade	25 µg	231
V1062	Chymotrypsin, Sequencing Grade	100µg (4 × 25 µg)	231
V1071	Endoproteinase Lys-C, Sequencing Grade	5 µg	233
V1111	Anti-ACTIVE® CaM KII pAb, Rabbit, (pT^{286})	40 µl	170
V1121	MEK Inhibitor U0126	5mg (5 × 1 mg)	48
V1141	Anti-ERK 1/2 pAb, Rabbit	40 µl	173
V1151	Donkey Anti-Goat IgG, AP	60 µl	181
V1161	SB 203580	1 mg	50
V1171	PMA	5 mg	51
V1181	4α-PMA	1 mg	51
V1191	PD 98059	5 mg	50
V1201	LY 294002	5 mg	50
V1211	Anti-ACTIVE® p38 pAb, Rabbit, (pTGpY)	100 µl	172
V1221	DNA IQ™ Spin Baskets	1,000 /bag	119

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V1231	Microtubes, 1.5ml	1,000 /bag	119
V1240	ProFluor® PKA Assay	4 plate	44
V1241	ProFluor® PKA Assay	8 plate	44
V1260	ProFluor® Ser/Thr PPase Assay	4 plate	53
V1261	ProFluor® Ser/Thr PPase Assay	8 plate	53
V1270	ProFluor® Src-Family Kinase Assay	4 plate	44
V1271	ProFluor® Src-Family Kinase Assay	8 plate	44
V1280	ProFluor® Tyrosine Phosphatase Assay	4 plate	54
V1281	ProFluor® Tyrosine Phosphatase Assay	8 plate	54
V1320	HisLink™ Spin Protein Purification System	25 reactions	228
V1361	PDE-Glo™ Phosphodiesterase Assay	1,000 assays	39
V1362	PDE-Glo™ Phosphodiesterase Assay	10,000 assays	39
V1391	Slicprep™ 96 Device	10 pack	119
V1401	MAO-Glo™ Assay	200 assays	82
V1402	MAO-Glo™ Assay	1,000 assays	82
V1452	MAO-A	500 µl	82
V1501	cAMP-Glo™ Assay	300 assays (384-well plate)	38
V1502	cAMP-Glo™ Assay	3,000 assays (384-well plate)	38
V1503	cAMP-Glo™ Assay	30,000 assays (384-well plate)	38
V1531	PhosphoCatch™ Phosphopeptide Enrichment System	10 pack	230
V1532	PhosphoCatch™ Phosphopeptide Enrichment System	20 pack	230
V1560	MAO-Glo™ Assay with MAO-A	1,000 assays	82
V1591	Manual Differex™ Magnet	1 each	120
V2011	SoftLink™ Soft Release Avidin Resin	1 ml	228
V2012	SoftLink™ Soft Release Avidin Resin	5 ml	228
V2020	PinPoint™ Xa Protein Purification System	1 system	229
V2031	PinPoint™ Xa-1 Vector	10 µg	229
V2071	ProteaseMAX™ Surfactant, Trypsin Enhancer	1 mg	231
V2072	ProteaseMAX™ Surfactant, Trypsin Enhancer	5 × 1 mg	231
V2081	UGT-Glo™ Assay	200 assays	82
V2082	UGT-Glo™ Assay	1,000 assays	82
V2111	Agarose, Low Melting Point, Analytical Grade	25 g	23
V2120	UGT-Glo™ UGT1A1 Screening System	200 assays	82
V2121	UGT-Glo™ UGT1A1 Screening System	1,000 assays	82
V2130	UGT-Glo™ UGT2B7 Screening System	200 assays	82
V2131	UGT-Glo™ UGT2B7 Screening System	1,000 assays	82
V2211	cdc2 Protein Kinase Peptide Substrate	1 mg	51
V2372	Olomoucine (cdc2 Protein Kinase Inhibitor)	0.5 mg	50
V2373	Olomoucine (cdc2 Protein Kinase Inhibitor)	10 mg	50
V2460	Serine/Threonine Phosphatase Assay System	96 reactions	52
V2471	Tyrosine Phosphatase Assay System	96 reactions	52
V2591	TetraLink™ Tetrameric Avidin Resin	1 ml	228
V2592	TetraLink™ Tetrameric Avidin Resin	5 ml	228
V2791	Guanidine Thiocyanate, Molecular Grade	100 g	26
V2831	Agarose, LMP, Preparative Grade for Large Fragments (>1,000bp)	25 g	22
V2861	SAM2® Biotin Capture Membrane	96 samples	45
V3011	PEG 8000 Powder, Molecular Biology Grade	500 g	28
V3021	Proteinase K	100 mg	102
V3021	Proteinase K	100 mg	102
V3021	Proteinase K	100 mg	103
V3021	Proteinase K	100 mg	119
V3021	Proteinase K	100 mg	120
V3021	Proteinase K	100 mg	233
V3031	Deep Well MagnaBot® 96 Magnetic Separation Device	1 each	108
V3111	Acrylamide, Molecular Grade	100 g	22
V3115	Acrylamide, Molecular Grade	500 g	22
V3121	Agarose, LE, Analytical Grade	100 g	22
V3125	Agarose, LE, Analytical Grade	500 g	22
V3131	Ammonium Persulfate, Molecular Grade	25 g	23
V3141	Bisacrylamide, Molecular Grade	25 g	24
V3143	Bisacrylamide, Molecular Grade	125 g	24
V3151	DTT, Molecular Grade (Dry Powder)	5 g	25
V3151	DTT, Molecular Grade (Dry Powder)	5 g	119

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V3151	DTT, Molecular Grade (Dry Powder)	5 g	120
V3155	DTT, Molecular Grade (Dry Powder)	25 g	25
V3171	Urea	1 kg	31
V3175	Urea	5 kg	31
V3181	Sephacryl® S-400	10 ml	16
V3181	Sephacryl® S-400	10 ml	28
V3281	Anti-ACTIVE® MAPK Family Sampler	1 each	171
V3471	MagnaBot® Large Volume Magnetic Separation Device	1 each	108
V3591	Pgp-Glo™ Assay System	10 ml	81
V3601	Pgp-Glo™ Assay System with P-glycoprotein	10 ml	81
V3680	HisLink™ 96 Protein Purification System	1 × 96	227
V3681	HisLink™ 96 Protein Purification System	5 × 96	227
V3691	Shaker Integration Plate	1 each	120
V3771	Kinase-Glo® Plus Luminescent Kinase Assay	10 ml	43
V3772	Kinase-Glo® Plus Luminescent Kinase Assay	10 × 10 ml	43
V3773	Kinase-Glo® Plus Luminescent Kinase Assay	100 ml	43
V3774	Kinase-Glo® Plus Luminescent Kinase Assay	10 × 100 ml	43
V3841	Agarose, LMP, Preparative Grade for Small Fragments (10 to 1,000bp)	25 g	22
V3941	X-Gal	100mg/2 ml	32
V3951	IPTG, Dioxane-Free	5 g	27
V3953	IPTG, Dioxane-Free	50 g	27
V3955	IPTG, Dioxane-Free	1 g	27
V4211	PinPoint™ Vector Sequencing Primer	2 µg	229
V4211	PinPoint™ Vector Sequencing Primer	2 µg	229
V4221	5M Sodium Chloride, Molecular Biology Grade	1 L	28
V4231	EDTA, 0.5M (pH 8.0), Molecular Biology Grade	100 ml	25
V4231	EDTA, 0.5M (pH 8.0), Molecular Biology Grade	100 ml	102
V4231	EDTA, 0.5M (pH 8.0), Molecular Biology Grade	100 ml	103
V4233	EDTA, 0.5M (pH 8.0), Molecular Biology Grade	400 ml	25
V4251	TBE Buffer, 10X, Molecular Biology Grade	1,000 ml	30
V4261	SSC Buffer, 20X, Molecular Grade	1,000 ml	29
V4271	TAE Buffer, 10X, Molecular Biology Grade	1,000 ml	29
V4281	TAE Buffer, 40X, Molecular Biology Grade	1,000 ml	29
V5111	Sequencing Grade Modified Trypsin	100 µg	232
V5113	Sequencing Grade Modified Trypsin, Frozen	100 µg	233
V5161	cAMP-Dependent Protein Kinase, Catalytic Subunit	2,500 u	47
V5171	cGMP-Dependent Protein Kinase (α-Isozyme)	6,000 u	47
V5261	Protein Kinase C	1µg (2 × 0.5 µg)	48
V5280	Trypsin Gold, Mass Spectrometry Grade	100 µg	232
V5291	384-Well Plate, Flat	10 /pk	108
V5311	384-Well Plate, Conical	10 /pk	108
V5330	PepTag® Non-Radioactive PKC Assay	120 reactions	47
V5340	PepTag® Non-Radioactive cAMP-Dependent Protein Kinase Assay	120 reactions	47
V5551	EGF Receptor	10 u	48
V5581	Factor Xa Protease	50 µg	233
V5591	Streptavidin Alkaline Phosphatase	0.5 ml	29
V5601	Kemptide (PKA) Peptide Substrate	1 mg	50
V5611	Neurogranin$_{(28–43)}$ (PKC) Peptide Substrate	1 mg	50
V5621	Casein Kinase II	100 u	48
V5631	Casein Kinase I	100 u	48
V5661	Casein Kinase II Peptide Substrate	1 mg	51
V5671	DNA-Dependent Protein Kinase Peptide Substrate	1 mg	50
V5681	cAMP-Dependent Protein Kinase Peptide Inhibitor	1 mg	49
V5691	Myristoylated Protein Kinase C Peptide Inhibitor	1 mg	49
V5811	DNA-Dependent Protein Kinase	2,500 u	47
V6041	MagnaBot® Flat Top Magnetic Separation Device	1 each	119
V6041	MagnaBot® Flat Top Magnetic Separation Device	1 each	120
V6051	ProTEV Protease	1,000 u	225
V6052	ProTEV Protease	10,000 u	225
V6071	Kinase-Glo® Max Luminescent Kinase Assay	10 ml	43
V6072	Kinase-Glo® Max Luminescent Kinase Assay	10 × 10 ml	43

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V6073	Kinase-Glo® Max Luminescent Kinase Assay	100 ml	43
V6074	Kinase-Glo® Max Luminescent Kinase Assay	10 × 100 ml	43
V6231	TE Buffer, 1X, Molecular Biology Grade	100 ml	30
V6232	TE Buffer, 1X, Molecular Biology Grade	500 ml	30
V6311	PPase-2A	25 u	52
V6361	PPase-2B	10 u	52
V6411	cGMP, 1mM	500 µl	52
V6421	cAMP, 1mM	500 µl	52
V6430	SignaTECT® cdc2 Protein Kinase Assay System	96 reactions	46
V6480	SignaTECT® Protein Tyrosine Kinase (PTK) Assay System	96 reactions	46
V6551	SDS Solution, Molecular Biology Grade (10% w/v)	100 ml	28
V6553	SDS Solution, Molecular Biology Grade (10% w/v)	500 ml	28
V6711	Kinase-Glo® Luminescent Kinase Assay	10 ml	43
V6712	Kinase-Glo® Luminescent Kinase Assay	10 × 10 ml	43
V6713	Kinase-Glo® Luminescent Kinase Assay	100 ml	43
V6714	Kinase-Glo® Luminescent Kinase Assay	10 × 100 ml	43
V6741	Deep Well Heat Transfer Block	1 each	120
V6751	VARIOMAG® Teleshake (110V, for North America use only)	1 each	120
V6761	V&P Scientific Heating Block (110V, North America use only)	1 each	120
V6771	1.2ml, Round-Bottom Deep Well Plate	50 /case	120
V6781	2.2ml, Square-Well Deep Well Plate	50 /case	120
V6791	Pyramid-Bottom Reservoir, 12 Column	25 /case	120
V6801	Pyramid-Bottom Reservoir	25 /case	120
V6811	U-Bottom Microplate	50 /case	120
V6821	1.1ml, Square-Well, V-Bottom Deep Well Plate	25 /case	120
V6831	10ml, 24-Well Deep Well Plate	25 /case	120
V6911	GSH-Glo™ Glutathione Assay	10 ml	68
V6912	GSH-Glo™ Glutathione Assay	50 ml	68
V7441	Casein Kinase I Peptide Substrate	1 mg	51
V7451	cGMP-Dependent Protein Kinase Peptide Substrate	1 mg	51
V7470	SignaTECT® Protein Kinase C (PKC) Assay System	96 reactions	46
V7480	SignaTECT® cAMP-Dependent Protein Kinase (PKA) Assay System	96 reactions	46
V7541	SAM²® 96 Biotin Capture Plate	96 -well plate	45
V7542	SAM²® 96 Biotin Capture Plate	5 × 96 -well plate	45
V7861	SAM²® Biotin Capture Membrane	7.6 × 10.9 cm	45
V7870	SignaTECT® DNA-Dependent Protein Kinase Assay System	96 reactions	46
V7931	Anti-ACTIVE® JNK pAb, Rabbit, (pTPpY)	40 µl	171
V7932	Anti-ACTIVE® JNK pAb, Rabbit, (pTPpY)	120 µl	171
V7951	Donkey Anti-Rabbit IgG (H+L), HRP	60 µl	180
V7971	Donkey Anti-Rabbit IgG (H+L), AP	60 µl	180
V7981	Antibiotic G-418 Sulfate	100 mg	23
V7982	Antibiotic G-418 Sulfate	1 g	23
V7983	Antibiotic G-418 Sulfate	5 g	23
V8031	Anti-ACTIVE® MAPK pAb, Rabbit, (pTEpY)	40 µl	171
V8051	Donkey Anti-Goat IgG, HRP	60 µl	181
V8081	Anti-pT^{183} MAPK pAb, Rabbit	50 µl	173
V8091	Antibiotic G-418 Sulfate Solution	20 ml	23
V8151	MagnaBot® 96 Magnetic Separation Device	1 each	108
V8151	MagnaBot® 96 Magnetic Separation Device	1 each	115
V8151	MagnaBot® 96 Magnetic Separation Device	1 each	119
V8161	SignaTECT® Calcium/ Calmodulin-Dependent Protein Kinase (CaM KII) Assay System	96 reactions	46
V8211	InCELLect™ AKAP St-Ht31 Inhibitor Peptide	150 µl	49
V8221	InCELLect™ St-Ht31P Control Peptide	150 µl	49
V8241	MagnaBot® 384 Magnetic Separation Device	1 each	108
V8251	Plate Clamp 96	1 each	108
V8261	Plate Stand	1 each	108
V8351	MagnaBot® II Magnetic Separation Device	1 each	108
V8381	MagnaBot® Spacer	1 each	108
V8481	MagnaBot® Adapter T1	1 each	108
V8491	Broad Range Protein Molecular Weight Markers	100 lanes	210
V8500	MagneHis™ Protein Purification System	65 reactions	226

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V8560	MagneHis™ Ni-Particles	2 ml	226
V8565	MagneHis™ Ni-Particles	10 ml	226
V8571	FastBreak™ Cell Lysis Reagent, 10X	15 ml	227
V8572	FastBreak™ Cell Lysis Reagent, 10X	60 ml	227
V8573	FastBreak™ Cell Lysis Reagent, 10X	100 ml	227
V8581	MagnaBot® Spacer 1/8 inch	1 each	108
V8600	MagneGST™ Protein Purification System	40 reactions	226
V8603	MagneGST™ Protein Purification System	200 reactions	226
V8611	MagneGST™ Glutathione Particles	4 ml	226
V8612	MagneGST™ Glutathione Particles	20 ml	226
V8681	MagnaBot® Spacer 1/16 inch	1 each	108
V8751	P450-Glo™ CYP1A1 Assay	10 ml	80
V8752	P450-Glo™ CYP1A1 Assay	50 ml	80
V8761	P450-Glo™ CYP1B1 Assay	10 ml	80
V8762	P450-Glo™ CYP1B1 Assay	50 ml	80
V8771	P450-Glo™ CYP1A2 Assay	10 ml	80
V8772	P450-Glo™ CYP1A2 Assay	50 ml	80
V8781	P450-Glo™ CYP2C8 Assay	10 ml	80
V8782	P450-Glo™ CYP2C8 Assay	50 ml	80
V8791	P450-Glo™ CYP2C9 Assay	10 ml	80
V8792	P450-Glo™ CYP2C9 Assay	50 ml	80
V8801	P450-Glo™ CYP3A4 Assay	10 ml	80
V8802	P450-Glo™ CYP3A4 Assay	50 ml	80
V8811	P450-Glo™ CYP3A7 Assay	10 ml	80
V8812	P450-Glo™ CYP3A7 Assay	50 ml	80
V8821	HisLink™ Protein Purification Resin	50 ml	227
V8823	HisLink™ Protein Purification Resin	5 ml	227
V8830	MagZ™ Protein Purification System	30 reactions	227
V8870	MagneGST™ Pull-Down System	80 reactions	240
V8881	P450-Glo™ CYP2C19 Assay	10 ml	80
V8882	P450-Glo™ CYP2C19 Assay	50 ml	80
V8891	P450-Glo™ CYP2D6 Assay	10 ml	80
V8892	P450-Glo™ CYP2D6 Assay	50 ml	80
V8901	P450-Glo™ CYP3A4 Assay (Luciferin-PFBE)Cell-Based/ Biochemical Assay	10 ml	80
V8902	P450-Glo™ CYP3A4 Assay (Luciferin-PFBE)Cell-Based/ Biochemical Assay	50 ml	80
V8911	P450-Glo™ CYP3A4 Assay (Luciferin-PPXE)DMSO-Tolerant Assay	10 ml	80
V8912	P450-Glo™ CYP3A4 Assay (Luciferin-PPXE)DMSO-Tolerant Assay	50 ml	80
V8920	Luciferin Detection Reagent	10 ml	80
V8921	Luciferin Detection Reagent	50 ml	80
V8930	Luciferin Detection Reagent with esterase	10 ml	80
V8931	Luciferin Detection Reagent with esterase	50 ml	80
V9001	P450-Glo™ CYP3A4 Assay with Luciferin-IPA	10 ml	80
V9002	P450-Glo™ CYP3A4 Assay with Luciferin-IPA	50 ml	80
V9012	Immobilized Trypsin	2 ml	231
V9013	Immobilized Trypsin	4ml (2 × 2 ml)	231
V9101	ADP-Glo™ Kinase Assay	1,000 assays	40
V9102	ADP-Glo™ Kinase Assay	10,000 assays	40
V9103	ADP-Glo™ Kinase Assay	100,000 assays	40
V9510	NADPH Regeneration System	1,000 assays	80
V9770	P450-Glo™ CYP1A2 Screening System	1,000 assays	80
V9790	P450-Glo™ CYP2C9 Screening System	1,000 assays	80
V9800	P450-Glo™ CYP3A4 Screening System	1,000 assays	80
V9880	P450-Glo™ CYP2C19 Screening System	1,000 assays	80
V9890	P450-Glo™ CYP2D6 Screening System	1,000 assays	80
V9910	P450-Glo™ CYP3A4 Screening System (Luciferin-PPXE)DMSO-Tolerant Assay	1,000 assays	80
V9920	P450-Glo™ CYP3A4 Screening System with Luciferin-IPA	1,000 assays	80
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W1015	ECL Western Blotting Substrate	500 ml	181
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W3841	Blot-Qualified BSA	10 g	182
W3940	ProtoBlot® II AP System with Stabilized Substrate, Human	1 each	182
W3950	ProtoBlot® II AP System with Stabilized Substrate, Mouse	1 each	182
W3960	ProtoBlot® II AP System with Stabilized Substrate, Rabbit	1 each	182
W4011	Anti-Rabbit IgG (H+L), HRP Conjugate	300 µl	181
W4021	Anti-Mouse IgG (H+L), HRP Conjugate	300 µl	181
W4031	Anti-Human IgG (H+L), HRP Conjugate	300 µl	181

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Y5101	MOPS/EDTA Buffer	3 × 10 ml	9
Y9341	Promega Flipper® Rack, Blue	8 × 8 tubes	252
Y9422	Promega Flipper® Rack, Purple	8 × 12 tubes	252
Z3051	RNA Lysis Buffer (RLA)	50 ml	110
Z3051	RNA Lysis Buffer (RLA)	50 ml	111
Z3051	RNA Lysis Buffer (RLA)	50 ml	112
Z3052	Wizard® SV Lysis Buffer	50 ml	102
Z3052	Wizard® SV Lysis Buffer	50 ml	103
Z3091	RNA Wash Solution (RWA)	58.8 ml	111
Z3091	RNA Wash Solution (RWA)	58.8 ml	112
Z3100	SV Total RNA Isolation System	50 preps	110
Z3101	SV Total RNA Isolation System	10 preps	110
Z3105	SV Total RNA Isolation System	250 preps	110
Z3141	Red Blood Cell Lysis Solution (CLB)	200 ml	110
Z3141	Red Blood Cell Lysis Solution (CLB)	200 ml	111
Z3191	Lysis Buffer B, Food	100 ml	106
Z3201	Precipitation Solution, Food	150 ml	106
Z3271	Heat Transfer Block	1 each	108
Z3291	Luminometer Plates	50 plates	75
Z3301	1/4 inch Foam Spacer	1 each	108
Z3351	MagneSil® Total RNA mini-Isolation System	4 plate	113
Z3500	SV 96 Total RNA Isolation System	1 × 96 each	112
Z3505	SV 96 Total RNA Isolation System	5 × 96 each	112
Z3631	Tube Holder, 50ml Tubes	1 each	108
Z3651	Heat Block Insert	1 each	108
Z3661	Heat Block Adapter, 50ml Tubes	1 each	108
Z3671	Shaker Top Adapter	1 each	108
Z3740	PureYield™ RNA Midiprep System	10 preps	111
Z3741	PureYield™ RNA Midiprep System	50 preps	111
Z3781	Anti-β-Galactosidase, Purified Monoclonal Antibody	100 µg	175
Z3783	Anti-β-Galactosidase, Purified Monoclonal Antibody	2 mg	175
Z4000	ChipShot™ Indirect Labeling and Clean-Up System	25 reactions	192
Z4100	ChipShot™ Direct Labeling and Clean-Up System	25 reactions	192
Z5200	PolyATtract® mRNA Isolation System II with Magnetic Stand	3 isolations	114
Z5210	PolyATtract® mRNA Isolation System I (Refill for Z5200)	3 isolations	114
Z5261	Biotinylated Oligo(dT) Probe (50pmol/µl)	35 µl	114
Z5300	PolyATtract® mRNA Isolation System III with Magnetic Stand	15 isolations	114
Z5310	PolyATtract® mRNA Isolation System IV (Refill for Z5300)	15 isolations	114
Z5331	MagneSphere® Technology Magnetic Separation Stand (two-position)	0.5 ml	115
Z5332	MagneSphere® Technology Magnetic Separation Stand (two-position)	1.5 ml	114
Z5332	MagneSphere® Technology Magnetic Separation Stand (two-position)	1.5 ml	115
Z5332	MagneSphere® Technology Magnetic Separation Stand (two-position)	1.5 ml	119
Z5333	MagneSphere® Technology Magnetic Separation Stand (two-position)	12 × 75 mm	114
Z5333	MagneSphere® Technology Magnetic Separation Stand (two-position)	12 × 75 mm	115
Z5341	MagneSphere® Technology Magnetic Separation Stand (twelve-position)	0.5 ml	115
Z5342	MagneSphere® Technology Magnetic Separation Stand (twelve-position)	1.5 ml	115
Z5342	MagneSphere® Technology Magnetic Separation Stand (twelve-position)	1.5 ml	119
Z5343	MagneSphere® Technology Magnetic Separation Stand (twelve-position)	12 × 75 mm	115
Z5400	PolyATtract® System 1000 without Magnetic Stand	Scal able	114
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Z5410	PolyATtract® System 1000 Magnetic Separation Stand	1 each	115
Z5420	PolyATtract® System 1000 with Magnetic Stand	Scal able	114
Z5481	Streptavidin MagneSphere® Paramagnetic Particles	9ml (15 × 0.6 ml)	116
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Z5531	PolyATtract® GTC Extraction Buffer	120 ml	114
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Z7041	Streptavidin	1 mg	116

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