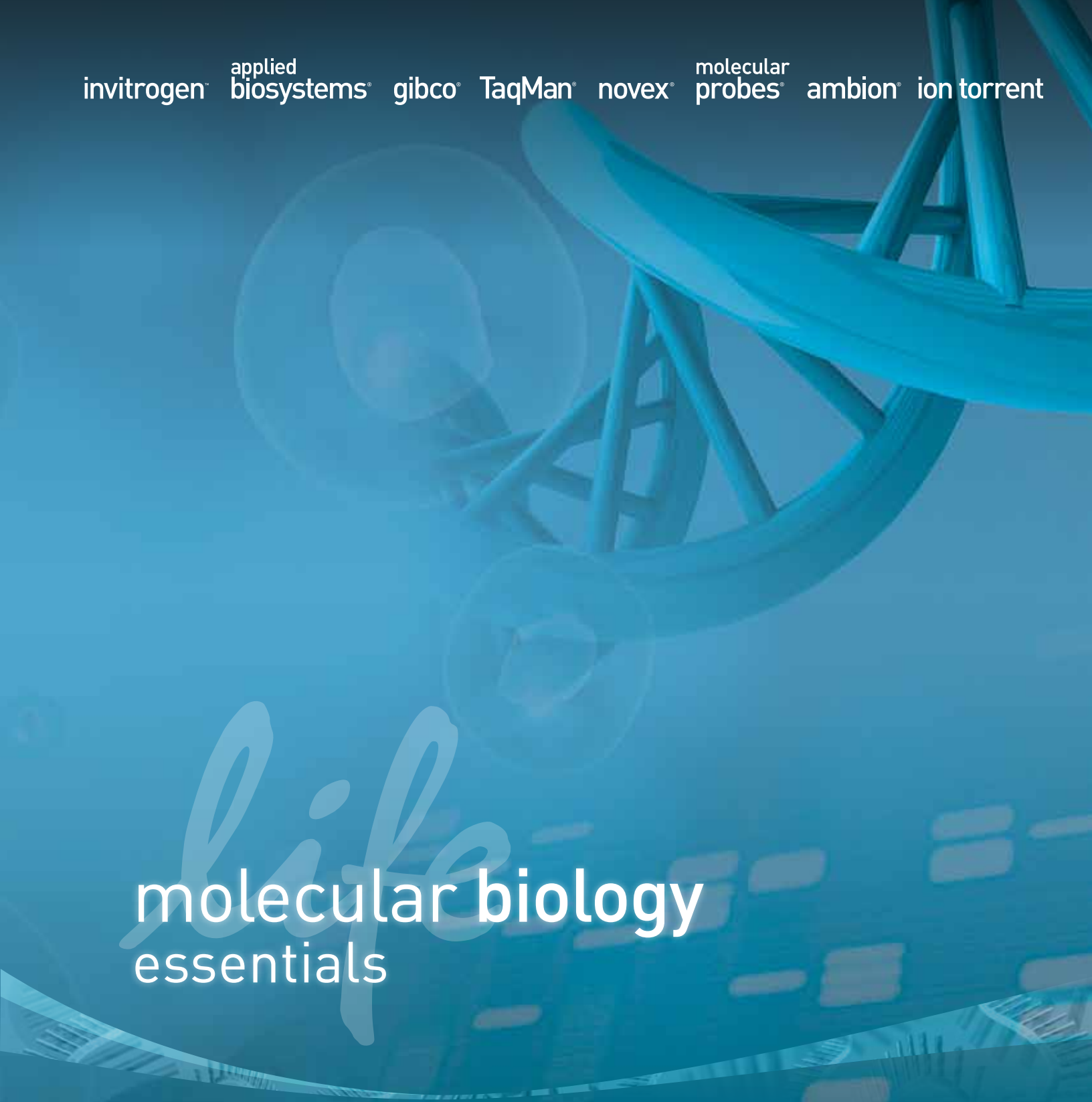
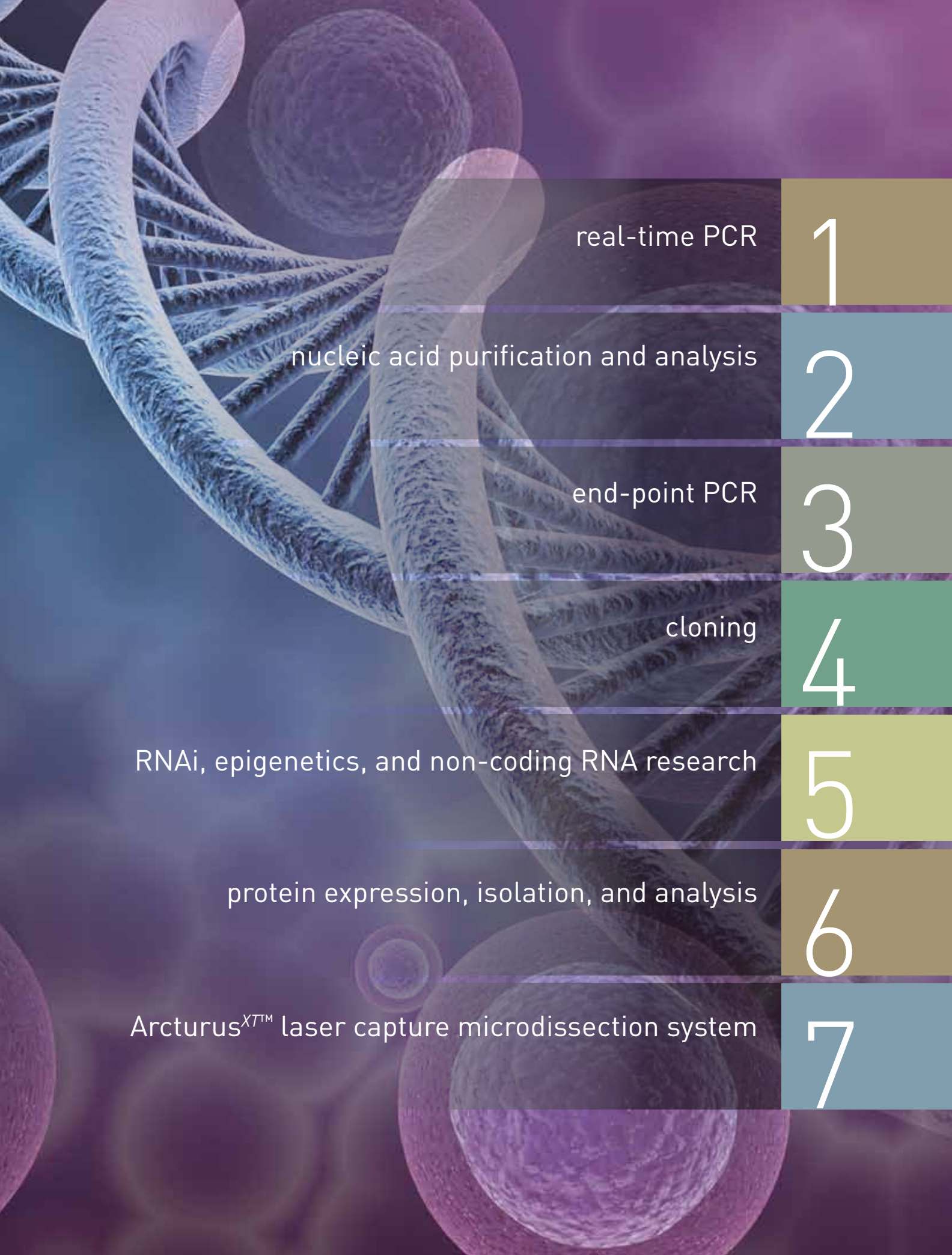
invitrogen™ applied biosystems® gibco® TaqMan® novex® molecular probes® ambion® ion torrent



molecular biology essentials

life technologies™



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2011/12 Life Technologies
Molecular Biology Essentials Catalogue

Comprising trusted and familiar Invitrogen™ and Applied Biosystems® products, the Life Technologies molecular biology portfolio offers life sciences researchers the most-published, high-performance, scalable solutions across the breadth of molecular biology applications.

The chart below shows an organized snapshot of these solutions, which have been cited in more than 200,000 publications within the past 10 years. To help you quickly find products that meet your needs, we've included icons to denote benefits like time-savings and high-throughput. Refer to the chapter noted for complete product information.

	Gene analysis and cloning	Protein expression
Sample preparation	• PureLink® Plasmid Prep **(Ch. 2)** • PureLink® RNA Mini Kit **(Ch. 2)** • BenchPro® 2100 Plasmid Purification System **(Ch. 2)** • MagMax™ sample preparation system **(Ch. 2)** • iPrep™ Purification Instrument **(Ch. 2)**	• Invitrogen™ Vector NTI® Cloning & Sequence Analysis Software **(Ch. 4)** • BenchPro® 2100 Plasmid Purification System
Key technologies	• GeneArt® Gene Synthesis and Gene Assembly **(Ch. 4)** Invitrogen™ cloning • SuperScript® Reverse Transcriptase, TOPO® cloning, Gateway,® TA Cloning® **(Ch. 4)** • Platinum® *Taq*, AmpliTaq Gold® 360 Master Mix **(Ch. 3)** • E-Gel® DNA & RNA separation gels **(Ch. 2)**	• Invitrogen™ protein expression vectors including Gateway® vectors **(Ch. 6)** • *E. coli*, baculoviral, lentiviral, adenoviral, yeast, & mammalian expression systems **(Ch. 6)** • Invitrogen™ Lipofectamine® transfection reagents **(Ch. 6)** • Neon® Transfection System **(Ch. 6)** • MembranePro™ Functional Protein Expression Kit **(Ch. 6)**
Detection instruments	• Invitrogen™ Qubit® DNA & RNA Quantitation System **(Ch. 2)** • Applied Biosystems® Veriti®, 9700, 2720 thermal cyclers **(Ch. 3)**	• Tali™ Image-Based Cytometer **(information at www.lifetechnologies.com)** • Countess® Automated Cell Counter **(information at www.lifetechnologies.com)**
Essentials	• TrackIt™ DNA Ladders **(Ch. 2)** • Oligos/primers **(Ch. 3)**, dNTPs **(Ch. 1)**, Invitrogen™ One Shot® competent cells **(Ch. 4)**, UltraPure™ agarose & reagents **(Ch. 2)** • Ambion® RNA Essentials **(Ch. 2)**, ligases & restriction enzymes **(Ch. 4)**	• Invitrogen™ One Shot® Competent Cells **(Ch. 4)** • imMedia™ growth medium & LB broth **(Ch. 4)** • Selection antibiotics **(Ch. 6)**

Time saver High throughput

You may notice that we've included only our most popular products and featured new technologies within these pages. Follow the web link within the chapters for a complete view of all our products. While you're on our website, be sure to check out our new products, protocols, technical guides, videos, and handbooks at www.lifetechnologies.com.

If you have any questions about which of our solutions is right for you, please contact Life Technologies technical support by phone, email, Facebook, or Twitter at www.facebook.com/LifeTechnologies and twitter.com/LifeTechSupport.

	Functional genetic analysis	Protein analysis
	• TRIzol® Reagent **(Ch. 2)** • Ambion® Cells-to-Ct™ **(Ch. 2)** • Ambion® *mir*Vana™ miRNA Isolation Kit **(Ch. 2)** • Ambion® *mir*Vana™ PARIS™ Kit **(Ch. 2)** • Arcturus^{XT}™ Laser Capture Dissection System **(Ch. 7)**	• Dynabeads® streptavidin magnetic beads **(Ch. 6)** • Invitrogen™ Qubit® Protein Quantitation System **(Ch. 6)** • IP antibodies **(Ch. 5)**
	• TaqMan® and SYBR® master mixes **(Ch. 1)** • TaqMan® Gene Expression, Genotyping (SNP), and CNV Assays **(Ch. 1)** • TaqMan® Non-coding RNA and Ambion® Pre-miR Assays **(Ch. 5)** • Epigenetics: methylation, histone/chromatin remodeling reagents **(Ch. 5)** • Ambion® *Silencer*® Select, Lipofectamine® RNAiMAX reagents, *mir*Vana™ miRNA Mimics and Inhibitors **(Ch. 5)**	• Novex® and NuPAGE® Gels **(Ch. 6)** • XCell SureLock® gel box **(Ch. 6)** • TaqMan® Protein Assays **(Ch. 1)**
	Applied Biosystems® Real-Time PCR Instruments • StepOnePlus™ System & 7500 Fast System **(Ch. 1)** • ViiA™ 7 Real-Time PCR System **(Ch. 1)** • OpenArray® System **(Ch. 1)**	• iBlot® Dry Blotting System **(Ch. 6)** • BenchPro® 4100 Western Processing Station **(Ch. 6)**
	• Applied Biosystems® primers **(Ch. 1)** • SYBR® Green assays **(Ch. 1)** • SuperScript® VILO kits **(Ch. 3)**	• Novex® Protein Stains **(Ch. 6)** • Novex® Buffers and protein standards **(Ch. 6)** • Novex® Gel cassettes and pour-your-own essentials (www.lifetechnologies.com/novexcassettes)

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Cell and Tissue Analysis solutions, go to www.lifetechnologies.com/cellularanalysis.
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real-time PCR

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Overview of real-time PCR

Advantages of real-time PCR

Quantitative real-time PCR, also called qPCR, combines PCR amplification and detection into a single step, thus eliminating the need to detect products using gel electrophoresis and enabling the PCR method to be truly quantitative. Real-time PCR systems utilize fluorescent dyes to detect the accumulation of PCR products during the exponential phase of the reaction, which allows for fast and precise product quantification and objective data analysis. In contrast, end-point PCR requires subjective analysis of product via gel electrophoresis, northern blots, or arrays. qPCR has several advantages over these semi-quantitative detection methods, including:

- Lower risk of contamination
- Easier workflow
- Faster time-to-results
- Broad dynamic range: up to 9 logs
- Higher sensitivity
- Ease of optimization
- Cost-effectiveness
- High throughput
- Easy-to-design controls
- Small input amounts of RNA or DNA required
- High reproducibility

TaqMan® and SYBR® Green detection methods

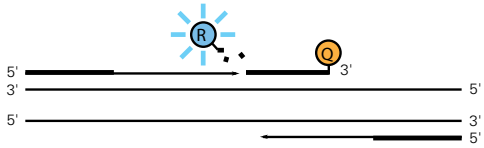
TaqMan® probe–based detection

TaqMan® chemistry entails the use of a fluorogenic probe to enable the detection of a specific PCR product as it accumulates during PCR. A single-stranded DNA probe is constructed containing a reporter fluorescent dye on the 5´ end and a quencher dye on the 3´ end. While the probe is intact, the proximity of the quencher dye greatly reduces the fluorescence emitted by the reporter dye by fluorescence resonance energy transfer (FRET) through space. If the target sequence is present, the probe anneals downstream from one of the primer sites and is cleaved by the 5´ nuclease activity of *Taq* DNA polymerase as this primer is extended.

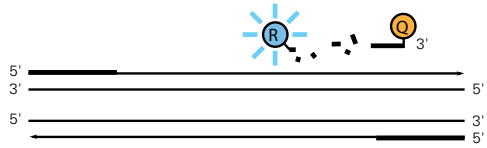
Forward and reverse primers are designed to hybridize to the target sequence flanking either side of the probe. At the start of the PCR, in conjunction with the reverse primer, the probe attaches to the complementary region of the target DNA. As the enzyme begins extending the reverse primer creating a new strand of DNA, it also degrades the attached probe that lies in its path. As the probe breaks apart, the reporter and quencher dye are separated. Their loss of proximity, and thus quenching, results in a fluorescent signal from the reporter dye. The reporter dye signal increases with each PCR cycle, and this signal is directly proportional to the amount of target DNA initially present in the reaction. A single cycle of the TaqMan® Assay is described in the figure below.

SYBR® Green–based detection

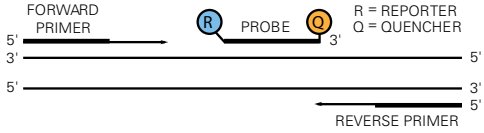
While SYBR® Green–based detection offers less specificity than TaqMan® probe–based approaches and does not enable assay multiplexing, it is often a less expensive method for qPCR analysis. However, more time is typically required to develop and optimize assays with



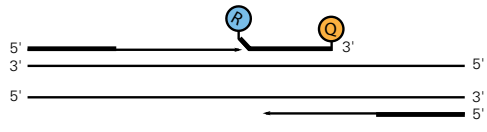
Step 1: Polymerization. A reporter (R) and a quencher (Q) are attached to the 5´ and 3´ ends of a TaqMan® probe.



Step 2: Strand displacement. When both labels are attached to the probe, reporter dye fluorescence is quenched.



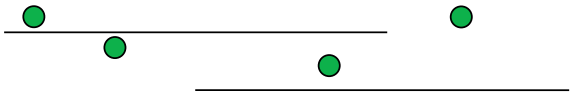
Step 3: Cleavage. During each extension cycle, *Taq* DNA polymerase cleaves the reporter dye from the probe.



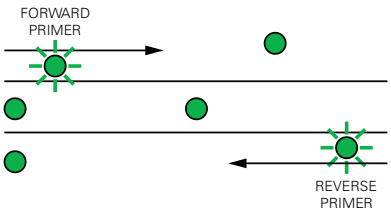
Step 4: Polymerization completed. Once separated from the quencher, the reporter dye emits its characteristic fluorescence.

TaqMan® Assay. TaqMan® chemistry uses a fluorogenic probe to enable the detection of a specific PCR product as it accumulates during PCR.

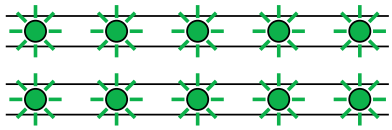
Step 1: Reaction setup. The SYBR® Green I dye fluoresces when bound to double-stranded DNA.



Step 2: Denaturation. When the DNA is denatured, the SYBR® Green I dye is released and fluorescence is drastically reduced.



Step 3: Polymerization. During extension, primers anneal and PCR product is generated.



Step 4: Polymerization completed. SYBR® Green I dye binds to the double-stranded product, resulting in a net increase in fluorescence detected by the instrument.

SYBR® Green Assay. SYBR® Green I dye is nonspecific and will bind to any double-stranded DNA molecule.

this approach. SYBR® Green I is a dye that binds to the minor groove of double-stranded DNA. When bound, the intensity of its fluorescent emission increases. Hence, as more double-stranded amplicons are produced during PCR, the SYBR® Green I dye signal increases proportionally (see figure above). The sensitivity of SYBR® Green I exceeds that of ethidium bromide and other conventional dyes by more than one order of magnitude. Researchers often use SYBR® Green chemistry for initial screening assays using qPCR. SYBR® Green technology is ideal for screening multiple targets or when a target-specific probe is not readily available. These assays can later be converted to TaqMan® probe–based detection systems, which have much greater specificity. SYBR® Green reagents include primers and master mixes that contain the SYBR® Green I dye.

Real-time PCR instruments

Real-time PCR instruments are used to perform the PCR as well as the real-time detection and monitoring of the resulting fluorescent signal. The Applied Biosystems® family of real-time PCR systems, which includes the ViiA™ 7, OpenArray®, 7900HT Fast, 7500 Fast, 7500, StepOnePlus™, and StepOne™ Real-Time PCR Systems, make qPCR more accessible than ever. These systems offer cutting-edge software designed to support your application and are flexible, accommodating the real-time chemistry of your choice.

Real-time PCR reagents and kits

Life Technologies' optimized master mixes and kits provide the ideal solution for sensitive, reliable, and reproducible results. The master mixes are available in multiple formats for different applications. qPCR reagents include the same components as traditional PCR, such as primers, enzymes, and dNTPs. However, TaqMan® chemistry is distinguished by the use of fluorescent dyes. TaqMan® technology, based on the 5´ nuclease assay, offers the highest specificity and sensitivity. SYBR® Green technology is ideal for screening multiple targets or when a target-specific probe is not readily available.

TaqMan® Assays and custom probes

Life Technologies offers predesigned, off-the-shelf, gene-specific probe and primer sets and Custom TaqMan® probes and primers manufactured to your desired target sequences. All products use TaqMan® probe–based chemistry and are designed for use on the suite of Applied Biosystems® real-time PCR systems. Together these components provide the gold standard in quantitative gene expression and genotyping applications, offering great sensitivity, specificity, reproducibility, and broad dynamic range.

Web resources

Real-Time PCR (Applications and Technologies Overview)
http://www.appliedbiosystems.com/absite/us/en/home/applications-technologies/real-time-pcr.html

Real-Time PCR Reagents Selection Guide
http://marketing.appliedbiosystems.com/mk/get/REALTIME_PCR_REAGENT_PRODUCT

Real-Time PCR Instruments Selection Guide
www.appliedbiosystems.com/absite/us/en/home/applications-technologies/real-time-pcr/real-time-pcr-instruments.html?ICID=EDI-Ord6

Real-Time PCR vs. Traditional PCR vs. Digital PCR
www.appliedbiosystems.com/absite/us/en/home/applications-technologies/real-time-pcr/real-time-pcr-vs-traditional-pcr.html?ICID=EDI-Lrn2

Real-Time PCR: Understanding C_t (Application Note)
www.appliedbiosystems.com/understandingct

Real-Time PCR Learning Area
http://www.appliedbiosystems.com/absite/us/en/home/applications-technologies/real-time-pcr/rtpcr-learn.html

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Real-time PCR instruments selection guide

Applied Biosystems® real-time PCR instruments offer the gold standard for high-performance, multicolor real-time PCR. With these real-time PCR platforms, you get the convenience of universal cycling conditions with TaqMan® Assays and the flexibility of unlimited real-time chemistry choices.

	Instruments				
					
Instrument/ capacity	StepOnePlus™ 96-well plates, 0.1-mL tubes StepOne™ 48-well plates, 0.1-mL tubes	7500 Fast 96-well optical Fast plates 7500 96-well optical plates or 0.2-mL tubes	7900HT 96-well, 384-well, and TaqMan® Array Microfluidic Cards	ViiA ™ 7 96-well, 384-well, and TaqMan® Array Microfluidic Cards	OpenArrat® 3,072 through-hole plates
Temperature uniformity	±0.5°C, within 30 sec of clock start	±0.5°C, within 30 sec of clock start	±0.5°C, within 30 sec of clock start	±0.5°C, within 30 sec of reaching temperature set point	±0.5°C
Fast block	Yes	Yes—7500 Fast No—7500	Yes—96-well Fast	Yes—96- and 384-well	No
Optics					
Excitation source	Blue LED	Tungsten-halogen	488 nm argon-ion laser	OptiFlex® System with halogen lamp	3 LEDs
Filters/colors	StepOnePlus™: 4 filters factory-calibrated for FAM™/ SYBR® Green, VIC®/JOE™, NED™/ TAMRA™, and ROX™ dyes StepOne™: 3 filters factory-calibrated for FAM™/SYBR® Green, VIC®/JOE™, and ROX™ dyes	5 filter sets calibrated for FAM™ /SYBR® Green, VIC®/JOE™, NED™/TAMRA™/Cy®3, ROX™/Texas Red®, Cy®5 dyes	None, CCD acts as spectrograph with continuous detection 500–650 nm	6 excitation filters (450–670 nm) 6 emission filters (500–720 nm) Filters are decoupled to allow for 21 filter combinations	3 filters factory-calibrated for FAM™/SYBR® Green, VIC®/JOE™, and ROX™ dyes
Detector	Photodiodes	CCD	CCD	CCD	CCD
Performance specifications					
Demonstrated sensitivity	10 copies	10 copies	10 copies	Down to 1 copy	100 copies
Dynamic range	Up to 9 logs of linear dynamic range	Up to 9 logs of linear dynamic range	Up to 9 logs of linear dynamic range	Up to 9 logs of linear dynamic range	Up to 6 logs of linear dynamic range
Instrument specifications					
Size	W 23.6 cm D 51.2 cm H 42.7 cm	W 34 cm D 45 cm H 49 cm	w/ drawer open: W 72 cm D 84 cm H 64 cm	W 53.3 cm D 63.5 cm H 64.5 cm	W 63 cm D 77 cm H 72.5 cm
Multiplex quantitation	Yes	Yes	Yes	Yes	No

OpenArray® Real-Time PCR Platform

Gene expression, genotyping, microRNA, and digital PCR in a mid-density, high-throughput format

OpenArray® Real-Time PCR Platform.

- Reliable, multiple sample loading—fill 3,072 through-holes (33 nL each) minimizing cross-contamination and with minimal hands-on time
- Economical, highly scalable approach—run up to three OpenArray® Plates simultaneously on the OpenArray® Real-Time PCR Instrument
- The OpenArray® Real-Time PCR Platform provides PCR-based genotyping, gene expression, microRNA, and digital PCR analysis of hundreds to thousands of samples and assays per day, enabling mid-density, high-throughput results with ease of use

Complete platform

The OpenArray® Real-Time PCR Platform includes:

- OpenArray® Real-Time PCR Instrument—images three OpenArray® Plates simultaneously
- OpenArray® AccuFill™ System—loads samples into OpenArray® Plates
- OpenArray® Case Sealing Station—seals OpenArray® Plates
- OpenArray® Instrument Accessories (includes PC system)
- OpenArray® Real-Time PCR Analysis Software and OpenArray® Genotyping Analysis Software

Hundreds of samples and assays

Researchers can analyze up to 144 samples on a single OpenArray® Plate. Three OpenArray® Plates can be cycled and imaged simultaneously on the OpenArray® Real-Time PCR Instrument, providing high-throughput results with ease. The system workflow thermally cycles and fluorescently images the OpenArray® Plates. Results are captured on the external PC, and the OpenArray® Analysis Software enables researchers to view specific assay data.

Well-equipped cycler

The OpenArray® Real-Time PCR Instrument uses three filtered high-brightness LEDs to excite, and a cooled CCD camera to detect fluorescence emission from dyes. The excitation and emission wavelengths support TaqMan® FAM™ dye detection and SYBR® Green I, ROX™, FAM™, and VIC® dye detection, making it an ideal platform for running all TaqMan® and SYBR® Green assays. The OpenArray® Real-Time PCR Instrument includes an integrated thermal block for gene expression, miRNA, and digital cycling. For genotyping applications, a Dual Flat Block GeneAmp® PCR System 9700 is required and can be purchased separately.

Analysis software

A number of software options make it easy to acquire, analyze, and manage OpenArray® data. OpenArray® Real-Time PCR Software exports gene expression data as C_t measurements. DataAssist™ Software povides additional multi-plate functionality for advanced gene expression and miRNA analysis. For digital PCR, OpenArray® Digital PCR Software fits real-time PCR data to a Poisson statistical model and displays digital results as copies per through-hole. OpenArray® SNP Genotyping Software automates genotype calling, with easy export for further analysis. TaqMan® Genotyper Software provides additional multi-plate functionality for powerful downstream analysis.

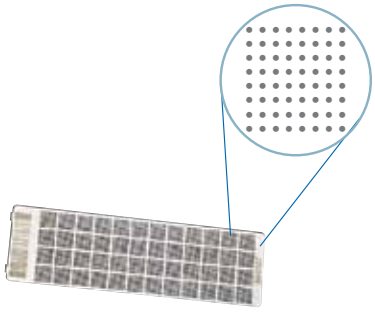
OpenArray® data can be easily manipulated and analyzed. Data from mutiple plates can be combined into the same project, and values such as C_t, T_m, ΔC_t, and $\Delta\Delta C_t$ can be quickly calculated. Data can be analyzed numerically or graphically, and can be exported in .cvs format for further analysis with third-party analysis packages.

Product	Quantity	Cat. No.
OpenArray® Real-Time PCR Instrument with AccuFill™ System	1 unit	4459283
OpenArray® AccuFill™ System	1 unit	4457243
OpenArray® AccuFill™ System Tips	1 box	4457246
OpenArray® AccuFill™ System Tips, 10 pack	10 boxes	4458107
OpenArray® Loader Tips	1 box	4404571
OpenArray® Loader Tips, 10 pack	10 boxes	4404604
OpenArray® Case Sealing Station	1 unit	4409361
Dual Flat Block GeneAmp® PCR System 9700 Sample Module Only	1 unit	4425757

OpenArray® Platform Technology

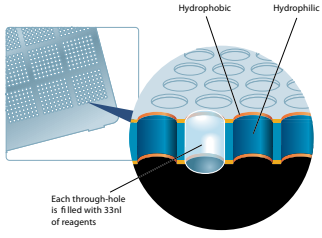
OpenArray® Platform technology is a broadly applicable nanoliter fluidics technology platform for low-volume, solution-phase reactions. Researchers using this technology benefit from the parallelism of microarrays and the data quality of solution-phase reactions such as PCR.

OpenArray® Platform technology utilizes a microscope slide–sized plate with 3,072 through-holes. Each through-hole is 300 µm in diameter with a depth of 300 µm. The plates are arranged in 48 subarrays of 64 through-holes. For gene expression and genotyping applications, TaqMan® or SYBR® Assays (for up to 48 samples) are preloaded and dried down in the plate, and are delivered ready for processing. For digital PCR applications, each through-hole is pre-treated to accept your assays and samples in your lab.



Each OpenArray® plate contains 48 subarrays with 64 through-holes each.

Proprietary processes are used to coat the surfaces of the plates so that they are hydrophobic, while rendering the interiors of the through-holes hydrophilic and biocompatible. When processed, each of the 3,072 through-holes contains 33 nL of fluid held in place by means of surface tension.



OpenArray® plate anatomy: cross-section of several through-holes in an OpenArray® plate. Each through-hole is coated with hydrophilic and hydrophobic coatings. Reagents are retained in through-holes via surface tension.

ViiA™ 7 Real-Time PCR System

High-performance features to maximize your productivity

- Proven Applied Biosystems reliability and accuracy
- Faster to set up, easier to run, and more convenient to automate
- Using TaqMan® Array Microfluidic Cards provides an integrated workflow and faster results
- Easy to use—intuitive software, responsive touch-screen, automation, effortless block exchange without the need for tools

Applied Biosystems® ViiA™ 7 Real-Time PCR System.

The ViiA™ 7 Real-Time PCR System combines all of the real-time PCR features you want in a single high-performance instrument, so that you can optimize your research productivity. With a streamlined workflow, intuitive software, touch-screen interface, and one-button protocols for error minimization, the ViiA™ 7 System offers exceptional reproducibility with minimal well-to-well and instrument-to-instrument variation.

Productivity

Ideal for performing medium- to high-throughput real-time PCR, the ViiA™ 7 System enhances your lab's productivity.

- Quick block changes—front access makes it easy to change thermal cycling block formats without having to move attached peripherals such as robots or computers
- Easy touch-screen interface—instrument touch screen provides one-touch protocols for fast and easy assay setup for a broad range of applications
- Automation compatibility—integrated with Applied Biosystems® Twister® II Robot, the ViiA™ 7 System allows you to maximize productivity for automated environments

Integration

Fully compatible with TaqMan® Array Microfluidic Cards and Applied Biosystems® TaqMan® Assays: the gold standard in real-time PCR analysis, providing high sensitivity, high specificity, and broad dynamic range.

- 384-well throughput without robotics—with TaqMan® Array Microfluidic Cards, there's no need for liquid-handling robotics or complex pipetting to load; simply add your sample and master mix and run on the ViiA™ 7 System
- Compatible with the full range of TaqMan® Assays—the ViiA™ 7 System is optimized to enable clear, clean data for all TaqMan® Assays, including MicroRNA, Protein, Non-coding RNA, and Pri-miRNA Assays

Performance

The ViiA™ 7 System has the following features:

- OptiFlex® System—enhanced fluorescence detection enabling accurate and sensitive data analysis
- Maximum multiplexing—six decoupled excitation and emission filter channels for the greatest number of dye combinations and maximum multiplexing capabilities
- Flexible data collection—multiple ramp method detection formats provide more flexibility for collecting data during a ramp stage
- Precise quantification—detect as small as 1.5-fold changes in target quantities in singleplex reactions

ViiA™ 7 Software offers:

- Intuitive interface and innovative design
- Convenient walk-away operation
- Improved data analysis
- Fully compatible with high-throughput environments

Expandable software architecture

Optional add-ons are available to upgrade features to existing software.

- 21 CFR Part 11 compliance—the optional SAE module assists with Security, Auditing and Electronic signature of records for full data traceability
- High Resolution Melt (HRM)—using the built-in MeltDoctor™ protocols and calibrations, you can spend less time optimizing the run and more time customizing your results. Define the number of variants you want to see by quickly changing analysis settings

Product	Quantity	Cat. No.
Applied Biosystems® ViiA™ 7 Real-Time PCR System with 384-Well Block	1 instrument	4453536
Applied Biosystems® ViiA™ 7 Real-Time PCR System with TaqMan® Array Block	1 instrument	4453537
Applied Biosystems® ViiA™ 7 Real-Time PCR System with 96-Well Fast Block	1 instrument	4453535
Applied Biosystems® ViiA™ 7 Real-Time PCR System with 96-Well Block	1 instrument	4453534
ViiA™ 7 System Automation Accessory Robot (100–240 V)	1 unit	4453551
384-Well Block Upgrade Kit	1 kit	4453545
TaqMan® Array Microfluidic Card Upgrade Kit	1 kit	4453546
96-Well Fast Block Upgrade Kit	1 kit	4453544
96-Well Block Upgrade Kit	1 kit	4453543

To download free DataAssist™ Analysis Software go to www.appliedbiosystems.com/dataassist

Applied Biosystems® 7500 Fast Real-Time PCR System

Maximum performance in minimum time, including high resolution melt analysis

- Powerful, five-color platform is calibrated for the broadest range of dyes available: FAM™/SYBR® Green I, VIC®/JOE™, NED™/ TAMRA™/ Cy®3, ROX™/Texas Red®, and Cy®5 dyes
- Specially designed Fast block helps ensure thermal uniformity at top speeds
- Fast ramp rates enable rapid results without compromising extension times or assay quality
- Fast optical plates provide excellent precision in 10–30 µL reaction volumes
- Ability to run multiple Fast assays on one plate
- Finish more runs per day with the excellent performance, equivalent to that offered by the 7500 System

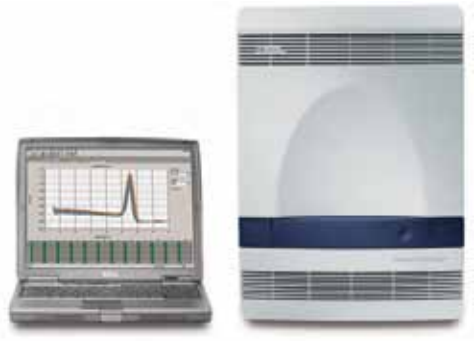
The Applied Biosystems® 7500 Fast Real-Time PCR System contains the same number of dyes and flexibility for multiplexing as the 7500 Real-Time PCR System, but also offers trusted performance in minimum time for labs running a variety of applications, including high resolution melt (HRM) analysis. Fully optimized for Fast cycling, the 7500 Fast delivers high-quality results in as little as 30 minutes.

7500 Software

The 7500 Software includes a variety of plate setup wizards, standard curve dilution and master mix recipe calculators, QC flags, data filters, and email notification when a run is finished. The 7500 Software v2.0 also includes a powerful Gene Expression Study package to accommodate large studies, and has a variety of melt curve protocol options, including multiple peak detection, step and hold temperature control, and customizable ramp rates.

One system—many applications

Applications include gene expression analysis, pathogen quantitation, SNP genotyping, copy number variation, isothermal, and +/– assays utilizing internal positive controls. The 7500 Fast can also be used for HRM analysis using Applied Biosystems® HRM Software. HRM Software is an easy-to-use melt analysis software enabling real-time PCR melt curve assays to be used more accurately for mutation scanning and genotyping. To facilitate many of these applications, Life Technologies provides preformulated, ready-to-use, quality-tested, TaqMan® Assays for use with the 7500 Fast system.



Applied Biosystems® 7500 Fast Real-Time PCR System.

21 CFR Part 11 module available

The SDS v1.4 21 CFR Part 11 Module is a powerful tool for assisting with 21 CFR Part 11 compliance, while still offering the flexibility of user-customizable configuration settings.

The 7500 Fast System includes:

- Choice of Dell® Business Line notebook or tower computer system with flat panel monitor
- Chemical Installation Kit containing dye calibration plates, RNase P Instrument Verification Plate, and application-specific starter kit
- Instrument operation and analysis software, plus Primer Express® primer and probe design software
- An extensive documentation set with Getting Started guides for all major real-time PCR applications
- Installation by a Life Technologies Service Engineer
- One-day on-site application training by a Life Technologies Field Applications Specialist

Product	Quantity	Cat. No.
Applied Biosystems® 7500 Fast Real-Time PCR System with Dell® Notebook	1 instrument	4351106
Applied Biosystems® 7500 Fast Real-Time PCR System with Dell® Tower	1 instrument	4351107
High Resolution Melt Software v3.0	1 license	4461357
	10 licenses	4461456
Applied Biosystems® 7500 Fast Real-Time PCR System Upgrade Kit	1 kit	4362143
7500 Fast System SDS v1.4 21 CFR Part 11 Module	1 unit	4377355

Applied Biosystems® 7500 Real-Time PCR System

Powerful platform for labs requiring superior performance and maximum dye versatility

- Powerful, five-color platform is calibrated for the broadest range of dyes available: FAM™/SYBR® Green I, VIC®/JOE™, NED™/ TAMRA™/ Cy®3, ROX™/Texas Red®, and Cy®5 dyes

The Applied Biosystems® 7500 Real-Time PCR System is a reliable platform for labs requiring maximum dye versatility. This platform features a sensitive optical system that lets you access a broad range of fluorophores. The variable excitation capability allows greater sensitivity for longer wavelength (red) dyes.

7500 Software

The 7500 Software incorporates easy-to-use features, such as plate setup wizards, standard curve dilution and master mix recipe calculators, QC flags, data filters, and email notification when a run is finished. The 7500 Software also includes a powerful Gene Expression Study package and has a variety of melt curve protocol options, including multiple-peak detection, step and hold temperature control, and customizable ramp rates.

21 CFR Part 11 Module available

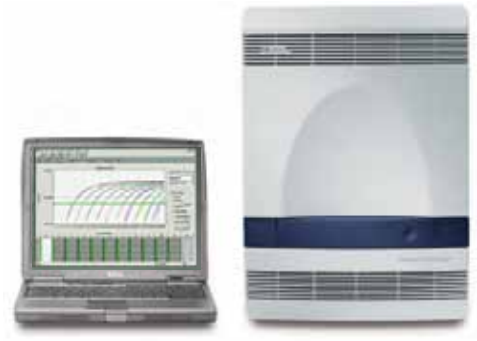
The SDS v1.4 21 CFR Part 11 Module is a powerful tool for assisting with 21 CFR Part 11 compliance, while still offering the flexibility of user-customizable configuration settings.

Supports many applications

Applications include gene expression analysis, pathogen quantitation, SNP genotyping, copy number variation, isothermal, and +/- assays utilizing internal positive controls. To facilitate many of these applications, Life Technologies provides preformulated, ready-to-use, quality-tested TaqMan® Assays for use with the 7500 System. Now you can reduce your assay optimization efforts.

Upgrade to high-speed thermal cycling

An optional upgrade to the 7500 Fast System is available. This 7500 Fast System uses our master mix formulations and enables you to shorten your real-time PCR runs to as little as 30 minutes.



Applied Biosystems® 7500 Real-Time PCR System.

The 7500 System includes:

- Choice of a Dell® Business Line notebook or tower computer system with flat-panel monitor
- Chemical Installation Kit containing dye calibration plates, RNase P Instrument Verification Plate, and application-specific starter kit
- Instrument operation and analysis software, plus Primer Express® primer and probe design software
- An extensive documentation set with Getting Started guides for all major real-time PCR applications
- Installation by a Life Technologies Service Engineer
- One-day on-site application training by a Life Technologies Field Applications Specialist

Product	Quantity	Cat. No.
Applied Biosystems® 7500 Real-Time PCR System with Dell® Notebook	1 instrument	4351104
Applied Biosystems® 7500 Real-Time PCR System with Dell® Tower	1 instrument	4351105
Applied Biosystems® 7500 Fast System Upgrade Kit	1 kit	4362143
7500 System SDS v1.4 21 CFR Part 11 Module	1 unit	4377354

StepOnePlus™ Real-Time PCR System

Simple and powerful platform for new and advanced real-time PCR researchers

- Advanced software and instrumentation for performing a wide array of genomic assays
- Long-life LED-based, 4-color optical system that can record fluorescence from FAM™/SYBR® Green, VIC®/JOE™, NED™/TAMRA™, and ROX™ dyes
- Intuitive and robust software guides users through their real-time PCR experiments in easy-to follow steps—perfect for first-time and advanced users
- Simple and flexible instrument set-up and usage
- Ultra-compact footprint fits any laboratory setting
- LCD touchscreen and USB drive provide configuration flexibility and enable PC-free operation

The StepOnePlus™ Real-Time PCR System is a 96-well real-time PCR instrument perfect for both first-time and experienced users. The StepOnePlus™ Real-Time PCR System can be set up in a variety of configurations and comes ready to use, out of the box, with intuitive data analysis and instrument control software. Utilizing robust LED-based, 4-color optical recording, the StepOnePlus™ Real-Time PCR System is designed to deliver precise, quantitative real-time PCR results for a variety of genomic research applications.

StepOne™ Software

The StepOne™ Software included with the StepOnePlus™ Real-Time PCR System runs on Windows® XP, Windows Vista®, and Windows® 7 operating systems and provides instrument control, data collection, and data analysis capabilities. This latest version includes:

- Capabilities to collect melt curve data for high resolution melt (HRM) experiments
- The option to export in Real-Time PCR Data Markup Language (RDML) for compatibility with MIQE guidelines
- Experimental design wizards to help you design and set up experiments
- Remote monitoring to view amplification in real-time from a remote monitor and email notification when a run is finished

In addition, the advanced software provided with the StepOnePlus™ Real-Time PCR System now includes the powerful Gene Expression Study Package. This software package allows for greater flexibility and accuracy in your gene expression assays.

StepOnePlus™ Real-Time PCR System.

Supports many applications

Applications include gene expression analysis, SNP genotyping, copy number variation, microRNA expression, protein expression, translocation analysis, gene detection, and viral load analysis. To facilitate many of these applications, Life Technologies provides preformulated, ready-to-use, quality-tested, TaqMan® Assays for use with the StepOnePlus™ System so you can reduce your assay optimization efforts.

The StepOnePlus™ System includes:

- Precalibration of the instrument
- RNase P Instrument Verification Plate
- Instrument operation and analysis software, plus Primer Express® primer and probe design software
- An extensive documentation set with Getting Started guides for all major real-time PCR applications
- Web-based training package provided by experienced applications personnel (Option to purchase on-site application training by a Life Technologies Field Applications Specialist)

Product	Quantity	Cat. No.
StepOnePlus™ Real-Time PCR System	1 instrument	4376600
StepOnePlus™ Real-Time PCR System with Laptop Computer	1 instrument	4376598
StepOnePlus™ Real-Time PC System with Tower Computer	1 instrument	4376599
High Resolution Melt Software v3.0	1 license	4461357
	10 licenses	4461456

StepOne™ Real-Time PCR System

Plug and play convenience with easy-to-use software for new and experienced real-time PCR instrument users

- Advanced software and instrumentation for performing a wide array of genomic assays
- Sensitive, long-life, 3-color optical LED recording system precalibrated for FAM™/SYBR® Green, VIC®/JOE™, and ROX™ dyes
- Intuitive and robust software perfect for first-time and advanced users
- Simple and flexible instrument set-up and usage
- Ultra-compact footprint fits any laboratory setting
- LCD touchscreen and USB drive provide configuration flexibility and enable PC-free operation

StepOne™ Real-Time PCR System.

The StepOne™ Real-Time PCR System is a 48-well, low-throughput real-time PCR instrument perfect for both first-time and experienced users. The StepOne™ Real-Time PCR System can be set up in a variety of configurations and comes ready to use, out of the box, with intuitive data analysis and instrument control software. Utilizing robust LED-based, 3-color optical recording, the StepOne™ Real-Time PCR System is designed to deliver precise, quantitative real-time PCR results for a variety of genomic research applications.

StepOne™ Software

The StepOne™ Software included with the StepOne™ Real-Time PCR System runs on Windows® XP, Windows Vista®, and Windows® 7 operating systems and provides instrument control, data collection, and data analysis capabilities. This latest version includes:

- Capabilities to collect melt curve data for High Resolution Melt (HRM) experiments
- The option to export in Real-Time PCR Data Markup Language (RDML) for compatibility with MIQE guidelines
- Experimental design wizards to help you design and set up experiments
- Remote monitoring to view amplification in real-time from a remote monitor and email notification when a run is finished

In addition, the advanced software provided with the StepOne™ Real-Time PCR System now includes the powerful Gene Expression Study Package. This software package allows for greater flexibility and accuracy in your gene expression assays.

Supports many applications

Applications include gene expression analysis, SNP genotyping. microRNA expression, translocation analysis, gene detection, and viral load analysis. To facilitate many of these applications, Life Technologies provides preformulated, ready-to-use, quality-tested, TaqMan® Assays for use with the StepOne™ System so you can reduce your assay optimization efforts.

Upgrade your StepOne™ System to a StepOnePlus System™

The StepOnePlus™ Real-Time PCR System Upgrade is an easy upgrade solution for your StepOne™ System. You can go from 3 colors and 48 wells to 4 colors and 96 wells in no time at all. During the upgrade process, we provide a StepOnePlus™ instrument on loan so you can continue your research until your own instrument arrives.

The StepOne™ System includes:

- Precalibration of the instrument
- RNase P Instrument Verification Plate
- Instrument operation and analysis software, plus Primer Express® primer and probe design software
- An extensive documentation set with Getting Started guides for all major real-time PCR applications
- Web-based training package provided by experienced applications personnel (option to purchase on-site application training by a Life Technologies Field Applications Specialist)

Product	Quantity	Cat. No.
StepOne™ Real-Time PCR System	1 instrument	4376357
StepOne™ Real-Time PCR System with Laptop Computer	1 instrument	4376373
StepOne™ Real-Time PCR System with Tower Computer	1 instrument	4376374
StepOnePlus™ Real-Time PCR System Upgrade Kit	1 kit	4379216
High Resolution Melt Software v3.0	1 license	4461357
	10 licenses	4461456

Applied Biosystems® 7900HT Fast Real-Time PCR System

Throughput and flexibility in real-time PCR instrumentation

- Fast PCR option reduces run time to about 35 minutes in a standard 96-well format, or about 55 minutes in a 384-well plate
- Continuous wavelength detection from 500–660 nm allows the use of multiple fluorophores in a single reaction
- 96- or 384-well plate compatibility (including the TaqMan® Array Microfluidic Card)
- Optional Enterprise Edition Software provides data analysis tools that help support 21 CFR Part 11 guidelines, providing data integrity and security
- Hands-free plate-loading and unloading provides true walk-away automation, allowing you to increase your lab's productivity
- Proven assay development guidelines help save time and money

Applied Biosystems® 7900HT Fast Real-Time PCR System.

The Applied Biosystems® 7900HT Fast Real-Time PCR System combines 96- and 384-well plate compatibility with fully automated robotic loading—and also offers optional Fast real-time PCR capability. The 7900HT Fast system combined with TaqMan® Assays enables you to achieve both throughput and flexibility. With researcher-friendly software, a 384-well TaqMan® Array Microfluidic Card format, and convenient TaqMan® Assay products, it's easy for labs of all sizes to realize the full potential of this powerful research tool.

High throughput

An Automation Accessory combined with 384-well plate capability make the 7900HT Fast system well suited to meet the high-throughput requirements of today's drug discovery process.

Flexibility

The 7900HT Fast system is a versatile research tool that can accommodate a variety of real-time PCR needs. User-interchangeable thermal cycling block formats let you select the format that's right for your project, using industry-standard 96- and 384-well formats, as well as a novel 384-well TaqMan® Array Microfluidic Card and a Fast 96-well block that reduces run times from 2 hours to about 30 minutes. An easy automation upgrade path lets you add features to meet throughput demands.

Powerful software

Using two automation software tools, Plate Utility and Automation Controller, applications can be fully automated for high-throughput applications and require minimal user intervention. You can process 5,000 x 384-well plates (384 markers), which equates to 1.92 million genotypes. Using Relative Quantification (RQ) Manager, you can also process 200 x 384-well plates (96 detectors with quadruplicate data points/multiplexed endogenous control), which equates to 153,600 data points. Fully integrated into one complete Enterprise system, SNP Manager and RQ Manager eliminate data analysis bottlenecks in high-throughput SNP and gene expression research by allowing significantly more plates to be analyzed simultaneously.

Product	Quantity	Cat. No.
Applied Biosystems® 7900HT Fast Real-Time PCR System with 384-Well Block Module	1 instrument	4329001
Applied Biosystems® 7900HT Fast Real-Time PCR System with 384-Well Block Module and Automation Accessory	1 instrument	4329002
Applied Biosystems® 7900HT Fast Real-Time PCR System with Fast 96-Well Block Module	1 instrument	4351405
Applied Biosystems® 7900HT Fast Real-Time PCR System with Standard 96-Well Block Module	1 instrument	4329003
High Resolution Melt (HRM) Software v2.0	1 unit	4397808

To download free DataAssist™ Analysis Software, go to www.appliedbiosystems.com/dataassist.

High Resolution Melt (HRM) Software

- Minimal user input to achieve results
- Easy recall analysis settings for future experiments
- Greater accuracy and increased sensitivity
- Ability to achieve more in one run
- Shortcuts in experiment setup save time

High resolution melt (HRM) analysis is an alternative to dHPLC sequencing screening of new gene variants. The Applied Biosystems HRM application does not require temperature shifting, which results in a greater likelihood of identifying new homozygous mutations than methods that require temperature shifting.

High Resolution Melt Software v3.0 is a new and improved tool for HRM analysis. HRM analysis is a post-PCR analysis method used to identify variation in nucleic acid sequences. The method is based on detecting small differences in PCR melt (dissociation) curves. It is enabled by high-brightness dsDNA-binding dyes used in conjunction with real-time PCR instrumentation that has precise temperature ramp control, advanced data capture capabilities, and access to software designed specifically for HRM analysis. High Resolution Melt Software v3.0 brings HRM to the StepOne™ and StepOnePlus™ Real-Time PCR Systems, as well as the 7500 Fast Real-Time PCR System, and contains features that make fast and accurate HRM results more accessible than ever.

Product	Quantity	Cat. No.
High Resolution Melt (HRM) Software v3.0 for StepOne™, StepOnePlus™, and 7500 Systems	1 license	4461357
	10 licenses	4461456
High Resolution Melt (HRM) Software v3.0 for ViiA™ 7 System	1 license	4453724
High Resolution Melt (HRM) Software v3.0 for 7900HT System	1 license	4397808

RealTime StatMiner® Software

Advanced data mining software for real-time PCR data analysis

RealTime StatMiner® Software provides a unique, straightforward solution to data analysis for real-time PCR for both life scientists and bioinformaticians. The intuitive user interface offers step-by-step guidance to simplify data processing and interpretation, helping researchers to extract biological relevance from real-time PCR data quickly and reliably. RealTime StatMiner® Software is widely used by pharmaceutical corporations, biotechnology companies, research centers, and core facilities around the world for high-throughput analysis of gene expression data. It combines commonly used interactive visualizations with advanced statistics to offer rapid, reliable analysis, allowing researchers to organize their samples based on technical and biological replicates, as well as rapidly create publication-ready reports.

RealTime StatMiner® Software automatically detects outliers and filters low-expressed or undetermined detectors, alerting the user to problems within their data and logging all activities during analysis. It also offers the unique ability to compute ΔC_t-based multiple endogenous controls, automatically determining the most stable controls using algorithms such as geNorm, NormFinder, and Minimum Variance Median. RealTime StatMiner® Software is available as a standalone as well as a TIBCO® Spotfire® software compatible plug-in that can be integrated with Integromics Biomarker Discovery® and SeqSolve™ software to provide a complete solution for gene expression analysis.

Compatibility

- Multi-platform compatibility—co-developed with Applied Biosystems for use with OpenArray®, ViiA™ 7, 7900HT, 7500, and 7500 Fast, StepOne™, StepOnePlus™, and other systems
- Multiple experimental assays—including TaqMan® and SYBR® Green
- Universal Data Loader—simplifying use of nonstandard data formats
- High throughput—allows use of multiple plates and samples, with more than 10 plates per run

Product	Quantity	Cat. No.
RealTime StatMiner® Software (stand-alone & TIBCO® Spotfire® software) for Academic Use	1-yr license	4398545
	3-yr license	4398087
RealTime StatMiner® Software (stand-alone & TIBCO®Spotfire® software) for Commercial Use	1-yr license	4398532
	3-yr license	4398086

Master mix selection table

Application	Detection chemistry	Starting material	Run speed	Product	Benefits
Gene expression or general purpose	TaqMan® probe	cDNA (2-step)	Fast*	TaqMan® Fast Advanced Master Mix	Highest-performing master mix for gene expression applications
		cDNA (2-step)	Standard	TaqMan® Universal Master Mix II	Ideal when working with multiple applications including miRNA, SNP genotyping, and copy number variation
		RNA (1-step)	Fast*	EXPRESS One-Step SuperScript® qRT-PCR Reagent Kit	Formulated for high yield and less PCR inhibition; includes premixed UDG
		RNA (1-step)	Standard	TaqMan® RNA-to-C$_T$™ 1-Step Kit	Delivers consistent RNA target quantification on a wide variety of targets
	SYBR® Green I	cDNA (2-step)	Fast	Fast SYBR® Green Master Mix	Designed to minimize primer-dimers and nonspecific amplification
		cDNA (2-step)	Standard	*Power* SYBR® Green Master Mix	Sensitivity and reproducibility with two copies of a target gene over a broad range of target template
		RNA (1-step)	Standard	*Power* SYBR® Green RNA-to-C$_T$™ *1-Step* Kit	Reduces false positive results due to primer-dimers with reliable detection of low abundance targets
Genotyping	TaqMan® probe	DNA	Fast	TaqMan® GTXpress™ Master Mix	Delivers accurate genotyping results with robust performance in less than 50 minutes; available with the TaqMan® Sample-to-SNP™ Kit for rapid SNP genotyping from samples
		DNA	Standard	TaqMan® Genotyping Master Mix	Specifically formulated for detection of SNPs and insertions/deletions
		DNA	Fast*	TaqMan® Fast Advanced Master Mix	Fast master mix works well for genotyping and other applications
miRNA	TaqMan® probe	cDNA	Fast*	TaqMan® Fast Advanced Master Mix	Detection of miRNA and other targets with fast cycling conditions
		cDNA	Standard	TaqMan® Universal Master Mix II	Ideal when working with multiple applications including miRNA, SNP genotyping, and copy number variation
Virus detection	TaqMan® probe	RNA (1-step) or DNA	Fast*	TaqMan® Fast Virus 1-Step Master Mix	Features designed for sensitive detection of RNA or DNA viruses
		DNA	Fast*	TaqMan® Fast Advanced Master Mix	High-performance master mix for multiple applications
Preamplification	Not applicable	cDNA	Standard	TaqMan® PreAmp Master Mix	Enables amplification of cDNA/DNA from limited samples for various applications, including single-cell studies
High resolution melt (HRM)	MeltDoctor™ HRM Dye	DNA	Standard	MeltDoctor™ HRM Master Mix	Formulated for HRM performance across a wide range of genomic targets without optimization
Protein expression	TaqMan® probe	DNA	Fast	TaqMan® Protein Assays Fast Master Mix	Used with TaqMan® Protein Expression Assays to quantify protein using real-time PCR

*Works in Fast and Standard mode.

TaqMan® Fast Advanced Master Mix

Performance superior to standard master mixes in less than half the time

- Best-in-class performance
- Engineered for enhanced benchtop stability
- Optimized for multiplexing
- Reduced run times on fast and standard instrumentation
- Validated for use with multiple real-time PCR applications, including microRNA assays

TaqMan® Fast Advanced Master Mix has been designed to match or exceed the performance of standard master mixes, delivering shorter run times (<40 minutes) with results equal to or better than what is achieved today.

Best-in-class performance

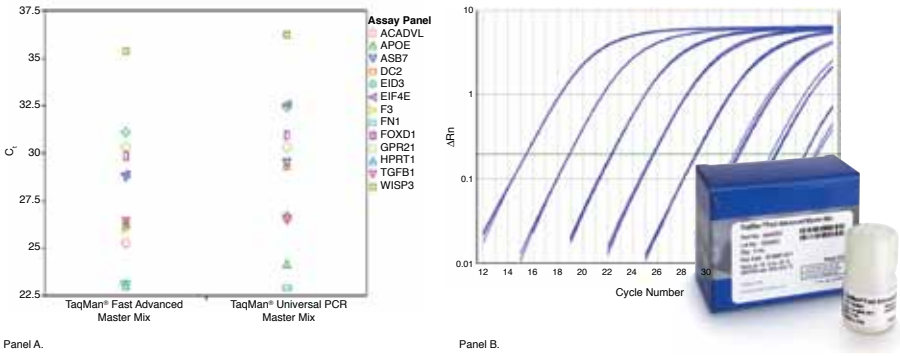
TaqMan® Fast Advanced Master Mix has been designed to provide performance equal to or better than the results you currently expect from your standard master mix. The master mix has been benchmarked against the leading suppliers' standard and fast master mixes to help ensure that it succeeds in providing best-in-class sensitivity, accuracy, dynamic range, and specificity.

Benchtop stability for high-throughput handling and convenience

TaqMan® Fast Advanced Master Mix has been engineered to retain a high level of performance in preassembled reactions for up to 72 hours. The stability of this mix provides users of high-throughput, liquid-handling systems the assurance that the results on the first plate will mimic those of the last plate. For those with less extreme throughput needs, the enhanced stability of this master mix provides an overall added convenience to your workflow, as you are no longer constrained to immediately running your plates upon assembly.

Reduced run times on standard instrumentation

TaqMan® Fast Advanced Master Mix has been optimized for use with both fast and standard instrumentation, enabling researchers who currently own standard instruments to also reap the performance benefits and time savings this mix provides.



Performance of TaqMan® Fast Advanced Master Mix vs. TaqMan® Universal PCR Master Mix.

Product	Quantity	Cat. No.
TaqMan® Fast Advanced Master Mix (1 x 1 mL)	100 reactions	4444556
TaqMan® Fast Advanced Master Mix (1 x 5 mL)	500 reactions	4444557
TaqMan® Fast Advanced Master Mix (2 x 5 mL)	1,000 reactions	444 4963
TaqMan® Fast Advanced Master Mix (5 x 5 mL)	2,500 reactions	4444964
TaqMan® Fast Advanced Master Mix (10 x 5 mL)	5,000 reactions	4444965
TaqMan® Fast Advanced Master Mix (1 x 50 mL)	5,000 reactions	4444558

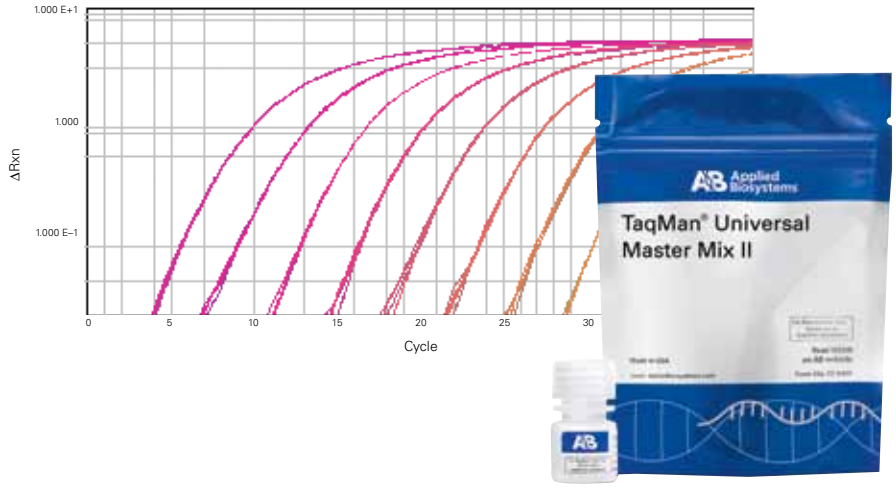
Quantity shown is for number of 20-µL reactions.

TaqMan® Universal Master Mix II

The real-time PCR master mix for multiple TaqMan® applications

- Stable at room temperature for 24 hours in preassembled PCR reactions
- Validated with TaqMan® Assays for gene expression, SNP genotyping, copy number, and microRNA
- Uses universal thermal cycling conditions for TaqMan® Assays
- Can be directly substituted into your existing protocols

TaqMan® Universal Master Mix II brings sensitivity and precision across a broad range of input target quantities, reliable detection of low copy number targets, and accurate quantification to discriminate subtle differences in target abundance. Like all TaqMan®Assay–based technologies, this real-time PCR master mix offers single-base discrimination between homologous sequences, and reactions can be run using universal thermal cycling conditions.



TaqMan® Universal Master Mix II.

Product	Quantity	Cat. No.
TaqMan® Universal Master Mix II, no UNG, Mini-Pack (1 mL tube)	100 reactions	4440043
TaqMan® Universal Master Mix II, no UNG, 1-Pack (5 mL bottle)	500 reactions	4440040
TaqMan® Universal Master Mix II, no UNG, 2-Pack (2 x 5 mL bottle)	1,000 reactions	4440047
TaqMan® Universal Master Mix II, no UNG, 5-Pack (5 x 5 mL bottle)	2,500 reactions	4440048
TaqMan® Universal Master Mix II, no UNG, 10-Pack (10 x 5 mL bottle)	5,000 reactions	4440049
TaqMan® Universal Master Mix II, no UNG, Bulk Pack (50 mL bottle)	5,000 reactions	4440041
TaqMan® Universal Master Mix II, with UNG, Mini-Pack (1 mL tube)	100 reactions	4440042
TaqMan® Universal Master Mix II, with UNG, 1-Pack (5 mL bottle)	500 reactions	4440038
TaqMan® Universal Master Mix II, with UNG, 2-Pack (2 x 5 mL bottle)	1,000 reactions	4440044
TaqMan® Universal Master Mix II, with UNG, 5-Pack (5 x 5 mL bottle)	2,500 reactions	4440045
TaqMan® Universal Master Mix II, with UNG, 10-Pack (10 x 5 mL bottle)	5,000 reactions	4440046
TaqMan® Universal Master Mix II, with UNG, Bulk Pack (50 mL bottle)	5,000 reactions	4440039

Quantity shown is for number of 20-µL reactions.

TaqMan® PreAmp Master Mix

Preamplify small amounts of cDNA without introducing bias

- Amplify cDNA targets equally without introducing bias
- Analyze mRNA from any precious sample such as laser capture microdissections (LCM), needle biopsies, and formalin-fixed paraffin-embedded tissues (FFPE)
- Stretch as little as 1 ng of cDNA into 200 real-time PCR reactions for gene expression analysis using TaqMan® Gene Expression Assays
- Analyze up to 100 gene expression targets with minimal hands-on time

TaqMan® PreAmp Master Mix preamplifies small amounts of cDNA without introducing amplification bias to the sample. Preamplification enables you to stretch your precious sample into many more real-time PCR reactions.

Preserves equilibrium of targets without preamplification bias

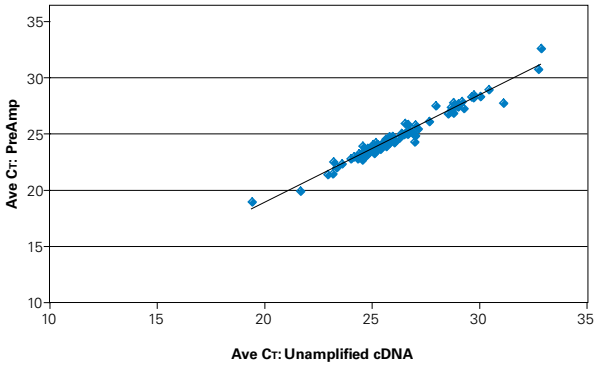
In the past, preamplification kits and techniques often resulted in uneven amplification of targets and incorrect or biased data (as shown by low correlation coefficients between preamplified and unamplified samples). TaqMan® PreAmp Master Mix amplifies with no bias and provides extremely high correlation between amplified and unamplified cDNA.

Easy workflow

With TaqMan® PreAmp Master Mix, preamplification setup takes only a few minutes. After a one-time pooling of the TaqMan® Assays, preamplification setup (mixing the pooled assays with the cDNA sample and the TaqMan® PreAmp Master Mix) typically takes only an additional 5 minutes. Cycling takes 1.5 hours using a 9700 thermal cycler.

Product	Quantity	Cat. No.
TaqMan® PreAmp Master Mix	40 reactions	4391128

Quantity shown is for number of 20-µL reactions.



High correlation between preamplified and unamplified cDNA. cDNA (1 ng) was preamplified using a pool of 96 TaqMan® Gene Expression Assays and subsequently amplified with a specific TaqMan® Assay from that set. The average C_t values for preamplified reactions are the y-axis values, and the average C_t values for the control (unamplified) cDNA sample (3 ng/reaction) are the x-axis values. $R^2 = 0.977$.

TaqMan® Sample-to-SNP™ Kit

One hour. One kit. Any sample.

- Fast—raw biological samples to SNP genotyping results typically in less than one hour
- Simple—a brief protocol with few pipetting steps
- Robust—highly accurate results for virtually any sample, without DNA quantitation
- Flexible—validated with many types of TaqMan® SNP Genotyping Assays and easily scalable to meet variable throughput needs

The TaqMan® Sample-to-SNP™ Kit provides a streamlined protocol for performing TaqMan® chemistry–based genotyping analysis from any sample with a single kit. The kit is comprised of two parts: the DNA Extract All Lysis Reagents and the TaqMan® GTXpress™ Master Mix. The DNA Extract All Lysis Reagents reduce prolonged procedures for the release of real-time PCR-ready DNA to a 5-minute protocol. They can process a wide variety of samples ranging from blood to buccal swabs to plant tissues (see Figure 2). TaqMan® GTXpress™ Master Mix enables robust PCR amplification typically in less than 50 minutes.

Robust reagent system

Many master mix products available today can perform reasonably well with highly purified DNA. However, even purified DNA from inhibitory samples such as blood or FFPE tissues can still pose challenges for these mixes. TaqMan® GTXpress™ Master Mix has been formulated to handle a broad spectrum of inhibitors contained in samples as varied as blood and cotton.

Concordance with purified DNA

The TaqMan® Sample-to-SNP™ Kit streamlines the SNP genotyping workflow without purifying genomic DNA. The kit was tested extensively for call concordance between the DNA lysate and purified DNA. The DNA lysate provides excellent cluster separation when compared to purified DNA. While the actual clustering is assay-dependent, a broad study comparing the genotyping call concordance between purified DNA and the DNA lysate across 46 EDTA-treated blood samples and two "no-template" controls showed excellent agreement. These 46 samples were genotyped with a 400-assay panel including 200 TaqMan® Predesigned SNP Genotyping Assays, 100 TaqMan® Drug Metabolism Genotyping Assays, and 100 Custom TaqMan® SNP Genotyping Assays. *Continued on next page.*

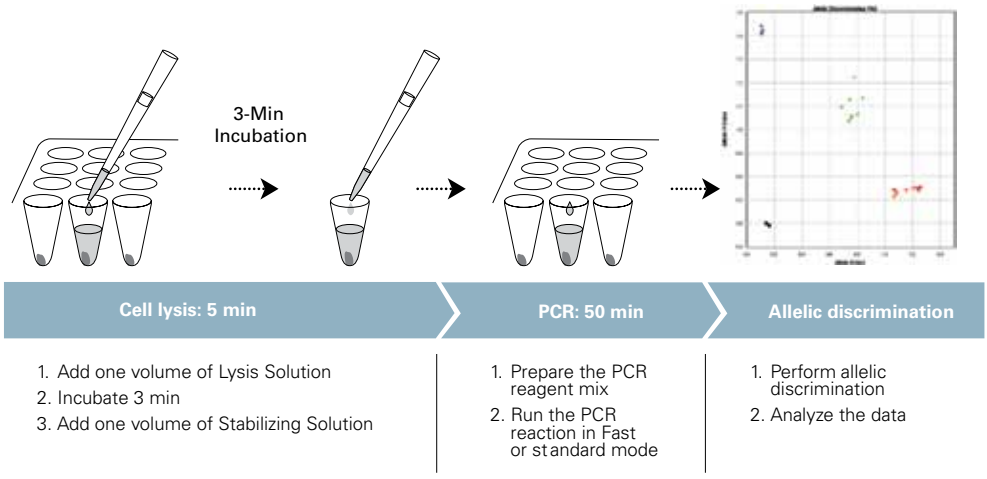


Figure 1. TaqMan® Sample-to-SNP™ Kit workflow.

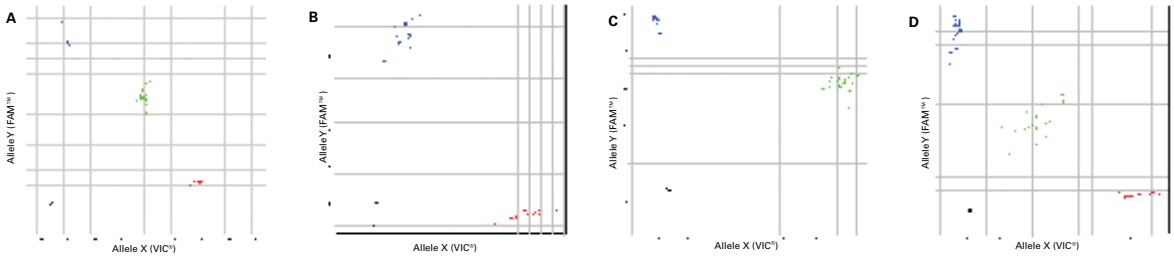


Figure 2. Allelic discrimination plots from various samples. Samples of buccal swab (A), mouse tail (B), corn leaf (C), and FFPE sample (D) were processed with the TaqMan® Sample-to-SNP™ Kit and the genotype results were obtained following the recommended user protocol. All amplifications and allelic discriminations were performed on an Applied Biosystems® 7900HT Fast Real-Time PCR System with the 384-well format.

Product	Quantity	Cat. No.
TaqMan® Sample-to-SNP™ Kit, Mini-Pack (5 mL sample prep, 1 mL PCR)	1 kit	4403313
TaqMan® Sample-to-SNP™ Kit (20 mL sample prep, 10 mL PCR)	1 kit	44033083
TaqMan® Sample-to-SNP™ Kit (20 mL sample prep, 50 mL PCR)	1 kit	4403087
TaqMan® Sample-to-SNP™ Kit (200 mL sample prep, 10 mL PCR)	1 kit	4403081
TaqMan® GTXpress™ Master Mix, Mini-Pack (1 mL)	100 reactions	4403311
TaqMan® GTXpress™ Master Mix, Mini-Pack (10 mL)	1,000 reactions	4401892
TaqMan® GTXpress™ Master Mix (1 x 50 mL)	5,000 reactions	4401890
TaqMan® GTXpress™ Master Mix (2 x 50 mL)	10,000 reactions	4401857

Quantity shown is for number of 20-µL reactions.

TaqMan® Genotyping Master Mix

Optimized for end-point fluorescence detection in SNP genotyping applications

- Distinct clusters and high call rates enable unambiguous allelic discrimination
- Validated with TaqMan® SNP Genotyping and Copy Number Assays
- Excellent pre- and post-PCR stability for high-throughput setup and analysis
- Exceptional performance from an optimized mix (2X)

Accurate genotype assignments result from preferential binding of the allele-specific probe to the matching target. TaqMan® Genotyping Master Mix enables specific binding of the probe to achieve exceptional cluster resolution. Key features include:

- AmpliTaq Gold® DNA Polymerase, UP (Ultra Pure)—automatic hot-start enzyme designed to be active during thermal cycling and inactive at room temperature for easy reaction setup
- Optimized mix components provide excellent specificity for discrimination between alleles
- Passive internal reference based on proprietary ROX™ dye for increased precision on Applied Biosystems® real-time PCR instruments
- Single thermal cycling condition enables consistent results with TaqMan® Assays

Validated with TaqMan® Genotyping Assays and instruments

TaqMan® Genotyping Master Mix is recommended for use with TaqMan® Copy Number Assays and has been tested across all types of TaqMan® SNP Genotyping Assays: TaqMan® Drug Metabolism Genotyping Assays, TaqMan® SNP Genotyping Assays, and Custom TaqMan® SNP Genotyping Assays. In addition, the master mix is validated with the Applied Biosystems® thermal cyclers and real-time PCR systems.

High-throughput setup and analysis

The combination of several low-cost thermal cyclers for PCR and a single real-time PCR instrument for allelic discrimination make high-throughput SNP genotyping manageable. TaqMan® Genotyping Master Mix further improves high-throughput setup and analysis with excellent room temperature stability both before and after PCR. This provides the flexibility required to run long experiments unattended overnight or over a weekend.

Reliable discrimination with challenging targets

Amplicon design constraints make SNP detection challenging, particularly when targets contain an abundance of GC- or AT-rich regions. The optimized components of TaqMan® Genotyping Master Mix result in tight, well-separated clusters and more accurate allele calls compared to other mixes, even for challenging targets.

Product	Quantity	Cat. No.
TaqMan® Genotyping Master Mix, Mini-Pack (1 mL)	100 reactions	4371353
TaqMan® Genotyping Master Mix, 1-Pack (1 x 10 mL)	1,000 reactions	4371355
TaqMan® Genotyping Master Mix, 2-Pack (2 x 10 mL)	2,000 reactions	4381656
TaqMan® Genotyping Master Mix, 1 Bulk Pack (1 x 50 mL)	5,000 reactions	4371357
TaqMan® Genotyping Master Mix, Multi-Bulk Pack (2 x 50 mL)	10,000 reactoins	4381657

Quantity shown is for number of 20-µL reactions.

TaqMan® Genotyping Master Mix.

MeltDoctor™ HRM Master Mix

Superior HRM performance across a wide range of genomic targets

- Low background fluorescence
- High brightness in the presence of double-stranded DNA
- Minimal temperature shift of DNA melting due to dye binding
- Thermal stability to tolerate PCR cycling conditions
- No inhibition of polymerase activity, resulting in high PCR efficiency
- A dNTP blend including dUTP, which minimizes carryover contamination by allowing amplicon degradation by uracil-DNA glycosylase (UDG) in subsequent PCR

High resolution melt (HRM) analysis is a post-PCR analysis method used to identify genetic variation in nucleic acid sequences. The MeltDoctor™ HRM Master Mix contains all components (excluding template and primers) formulated for superior HRM performance across a wide range of genomic targets. Unlike some other mixes, the MeltDoctor™ HRM Master Mix does not require additional mixing prior to use and was developed and optimized solely for HRM applications.

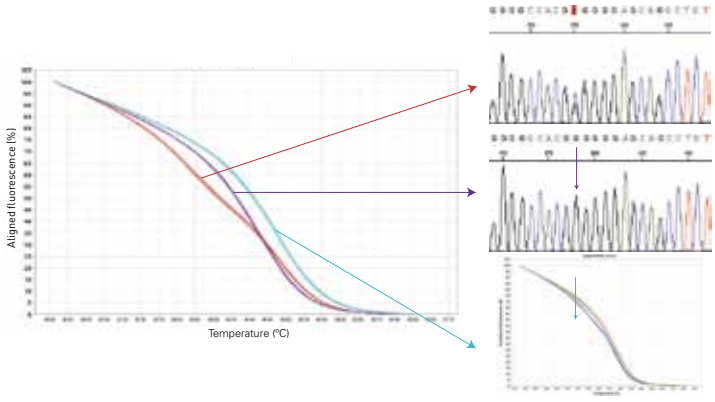
Perform mutation scanning

HRM analysis can be used to scan for mutations in target genes for the identification of variant samples prior to sequencing analysis (see figure). As a mutation-scanning technique, HRM offers significant advantages over conventional methods such as denaturing high-performance liquid chromatography (DHPLC) and denaturing gradient gel electrophoresis (DGGE). Specifically, the advantages of HRM for mutation scanning include:

- Low reagent consumption, with little waste: HRM requires only a 20-µL PCR for analysis of each sample, eliminating the need for HPLC solvents or DGGE gels
- Simple, fast workflow: no additional instrumentation is required after PCR amplification. An HRM step can be simply added to the end of the PCR profile for immediate analysis
- Fast optimization: unlike DHPLC, thermal optimization is not required
- Low sample consumption: following HRM analysis, the PCR product can be used directly in a Sanger sequencing reaction

Quality components

- Magnesium salts and other buffer components, precisely formulated to obtain optimal HRM results
- AmpliTaq Gold® 360 DNA Polymerase, a highly purified DNA polymerase that provides hot-start performance, minimizing nonspecific product formation and enabling reactions to be set up at room temperature
- MeltDoctor™ HRM Dye, a stabilized form of the fluorescent Molecular Probes® SYTO® 9 double-stranded nucleic acid stain



Mutation scanning using the Applied Biosystems® HRM workflow. Genomic DNA samples from three cell lines (HeLa, Raji, and Jurkat) were analyzed by HRM using MeltDoctor™ HRM Master Mix on an Applied Biosystems® 7500 Fast Real-Time PCR System and analyzed using Applied Biosystems® HRM Software v2.0, followed by DNA sequencing. Primers were designed to amplify 152 bp of exon 4 of the *p53* tumor suppressor gene. Three genotypes are clearly distinguishable in the aligned HRM profile (left), and they were called accurately by the analysis software. Following HRM, the genotype of each sample (GC, GG, CC) was identified by sequencing (right).

Product	Quantity	Cat. No.
MeltDoctor™ HRM Master Mix (5 mL)	500 reactions	4415440
MeltDoctor™ HRM Master Mix (5 x 5 mL)	2,500 reactions	4415452
MeltDoctor™ HRM Master Mix (10 x 5 mL)	5,000 reactions	4415450
MeltDoctor™ HRM Master Mix (50 mL)	5,000 reactions	4409535
MeltDoctor™ HRM Reagent Kit	1 kit	4425557

Quantity shown is for number of 20-µL reactions.

Fast SYBR® Green Master Mix

- Fast—real-time PCR results in as fast as 35 minutes
- Sensitive—detect very low numbers of copies of target
- Specific—minimize primer-dimer and nonspecific amplification
- Reproducible—consistent amplification across a wide dynamic range

Fast SYBR® Green Master Mix delivers a fast and reliable solution for your real-time PCR applications without compromising sensitivity, specificity, dynamic range, or PCR efficiency. Fast SYBR® Green Master Mix contains all of the components, excluding the template and primers, in a convenient 2X mix. It includes the following components in an optimized buffer:

- AmpliTaq® Fast DNA Polymerase, UP, a highly purified DNA polymerase designed to allow instant hot start, minimizing nonspecific product formation and enabling reactions to be set up at room temperature
- SYBR® Green I dye enabling detection of double-stranded DNA
- Deoxynucleotides (dNTPs) to help maintain optimal PCR results
- Uracil-DNA glycosylase (UDG) designed to reduce carryover contamination
- Passive internal reference based on proprietary ROX™ dye to enable increased precision

Exceptional speed without compromise

Fast SYBR® Green Master Mix is designed to deliver PCR results on Fast instruments in less than half the time of standard real-time PCR reagents, while maintaining the equivalent PCR performance on Fast PCR. The dynamic range, sensitivity, and resulting PCR efficiency of Fast SYBR® Green Master Mix is comparable to *Power* SYBR® Green PCR Master Mix, while PCR run time is decreased by at least 60%.

Minimizing nonspecific amplification without compromising sensitivity

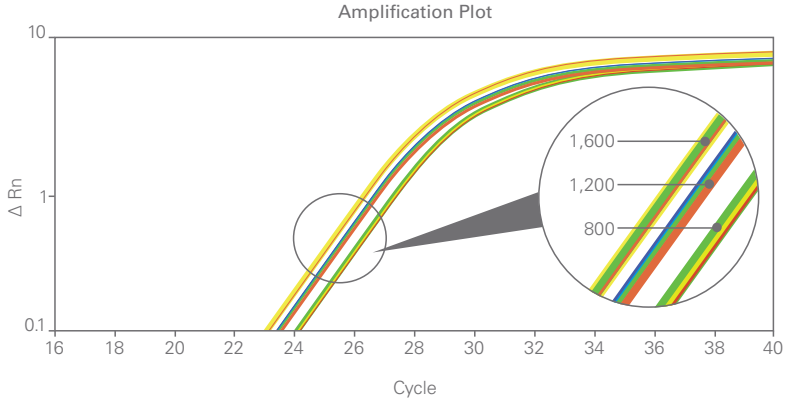
The presence of secondary, nonspecific PCR products and primer-dimers can help reduce amplification efficiency, and ultimately the accuracy of the data. Primer-dimers can also limit the dynamic range of the desired standard curve due to competition for reaction components during amplification. Fast SYBR® Green Master Mix was formulated to minimize primer dimerization and nonspecific amplification to optimize the efficiency and accuracy of the reaction without compromising sensitivity.

Reliable quantitation of abundant and limited targets

Fast SYBR® Green Master Mix is designed to provide dependable target quantitation over a wide dynamic range and is not limited only to abundant transcripts such as plasmids. Sensitivity of the master mix is validated by amplifying a single-copy gene and obtaining the expected target quantity and corresponding mean C_t values at a single-copy level.

Discriminating less than 2-fold difference

The high sensitivity provided by Fast SYBR® Green Master Mix facilitates quantifying small differences in target amount between samples. For example, the amplification plot shown in the figure below shows a clear, statistically significant discrimination down to 1.33-fold differences between samples of RNase P gene with 99.7% confidence.



High precision in target quantification with similar abundance levels using the Fast SYBR® Green Master Mix. Discrimination between 800, 1,200, and 1,600 copies of the RNase P gene, amplified from human gDNA (32 replicate reactions) on the StepOnePlus™ Real-Time PCR System with up to 99.97% confidence.

Product	Quantity	Cat. No.
Fast SYBR® Green Master Mix, Mini-Pack (1 x 1 mL)	100 reactions	4385610
Fast SYBR® Green Master Mix, 1-Pack (1 x 5 mL)	500 reactions	4385612
Fast SYBR® Green Master Mix, 2-Pack (2 x 5 mL)	1,000 reactions	4385616
Fast SYBR® Green Master Mix, 5-Pack (5 x 5 mL)	2,500 reactions	4385617
Fast SYBR® Green Master Mix, 10-Pack (10 x 5 mL)	5,000 reactions	4385618
Fast SYBR® Green Master Mix, Bulk Pack (1 x 50 mL)	5,000 reactions	4385614

Quantity shown is for number of 20-µL reactions.

Power SYBR® Green PCR Master Mix

Superior sensitivity and reproducibility for your real-time PCR experiments

- Highly sensitive DNA quantitation enables low copy-number detection
- Optimized formulation offers improved consistency
- Compatible with existing Applied Biosystems® SYBR® Green reagent protocols

The new *Power* SYBR® Green PCR Master Mix delivers superior sensitivity and reproducibility without compromising other performance parameters, such as specificity, wide dynamic range, and uniformity in your real-time PCR experiments.

Detect as few as 2 copies of a target gene

Power SYBR® Green PCR Master Mix delivers highly sensitive nucleic acid quantitation, detecting as few as two copies of a target gene over a broad range of template concentrations. *Power* SYBR® Green PCR Master Mix also provides high fluorescent signal strength for consistent detection of low copy numbers. The newly optimized formulation contains highly purified AmpliTaq Gold® DNA Polymerase, LD, to offer greater sensitivity. Additionally, our new formulation minimizes variation to help ensure consistency between experiments. Most importantly, the new *Power* SYBR® Green reagent–based PCR chemistry easily replaces the existing SYBR® Green PCR Master Mix in Applied Biosystems protocols using the same setup and thermal cycling conditions.

Optimized formulation for maximum performance

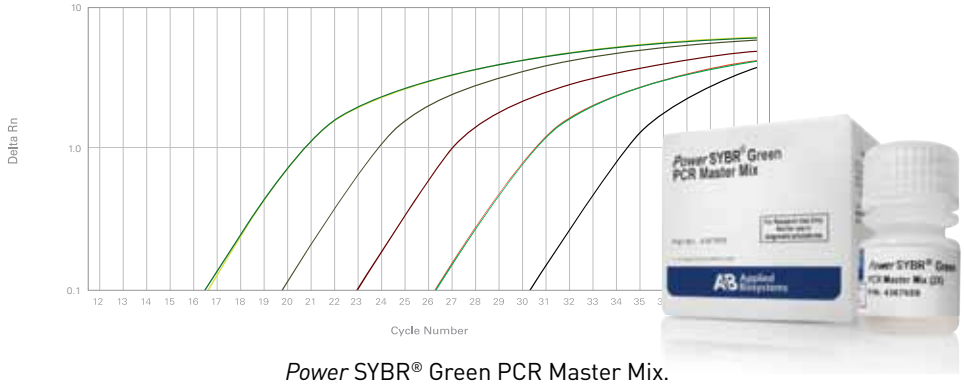
The new formulation contains a blend of dTTP/dUTP and employs a highly purified enzyme, AmpliTaq Gold® DNA Polymerase, LD, to provide greater specificity and reliability. *Power* SYBR® Green PCR Master Mix is compatible with AmpErase® UNG, which minimizes carryover contamination.

Ready to use

Power SYBR® Green PCR Master Mix provides greater flexibility, reduces experimental design time, and requires fewer reagents. *Power* SYBR® Green PCR Master Mix contains all of the components, excluding the template and primers, for superior SYBR® Green reagent–based real-time PCR. The AmpliTaq Gold® DNA Polymerase, LD is a hot-start PCR enzyme provided in an inactive state allowing for room temperature setup and minimizing nonspecific amplification.

High precision

Power SYBR® Green PCR Master Mix contains a proprietary version of ROX™ dye, an internal passive reference, to normalize non–PCR-related fluorescence fluctuations. Normalizing with an internal passive reference minimizes well-to-well variability that results from a variety of causes, such as pipetting error and sample evaporation.



Power SYBR® Green PCR Master Mix.

Product	Quantity	Cat. No.
Power SYBR® Green PCR Master Mix, Mini Pack (1 x 1 mL)	100 reactions	4368577
Power SYBR® Green PCR Master Mix, 1-Pack (1 x 5 mL)	500 reactions	4367659
Power SYBR® Green PCR Master Mix, 2-Pack (2 x 5 mL)	1,000 reactions	4368706
Power SYBR® Green PCR Master Mix, 5-Pack (5 x 5 mL)	2,500 reactions	4368702
Power SYBR® Green PCR Master Mix, 10-Pack (10 x 5 mL)	5,000 reactions	4368708
Power SYBR® Green PCR Master Mix, Bulk Pack (1 x 50 mL)	5,000 reactions	4367660

Quantity shown is for number of 20-µL reactions.

Express One-Step SuperScript® qRT-PCR Universal

Get the turnaround time you want with the data quality you need

- Highly sensitive and reproducible—uses fast-activating, antibody-mediated Platinum® *Taq* DNA Polymerase, which provides complete, rapid activation
- Better yield and less PCR inhibition—one-step SuperScript® III Reverse Transcriptase module has a unique formulation specifically for one-step qRT-PCR
- Flexible formats—supplied with ROX™ dye premixed or in a separate tube, designed to work with default instrument protocols on a wide range of high-throughput fast instruments (e.g., Applied Biosystems® 7500 Fast, 7900 HT Fast, StepOnePlus™ and StepOne™; Roche LightCycler® 480, and Qiagen Rotor-Gene® instruments)
- Significantly reduced carryover contamination risk—the only commercially available one-step qRT-PCR kit containing heat-labile uracil-DNA glycosylase (UDG)

EXPRESS One-Step SuperScript® qRT-PCR Universal is designed for sensitive, convenient, and contamination-free qPCR assays. It incorporates Platinum® *Taq* DNA Polymerase, which activates more quickly than chemically modified enzymes, to help deliver superior results on high-throughput, fast-cycling instruments for your probe-based qPCR experiments.

Product	Quantity	Cat. No.
Express One-Step SuperScript® qRT-PCR Universal	500 reactions	11781200
	2,500 reactions	117810K
Express One-Step SuperScript® qRT-PCR Universal with Premixed ROX™ Dye	500 reactions	11791200
	2,500 reactions	1179101K

Quantity shown is for number of 20-µL reactions.

TaqMan® Fast Virus 1-Step Master Mix

High-sensitivity detection of viral targets

- High sensitivity—one-tube, one-step 4X master mix to amplify both RNA and DNA with high sensitivity
- Consistent results—formulated to handle common RT-PCR inhibitors found in blood, stool, and other difficult samples
- Simplified application—single run profile with RNA and DNA, and on any supported instrument, which allows for easy mix-and-match of targets on a plate
- Target flexibility—works in singleplex, multiplex, and with exogenous or endogenous internal controls
- Fast results—increased real-time RT-PCR speed on fast and on standard instruments

TaqMan® Fast Virus 1-Step Master Mix is designed for reliable, high-sensitivity real-time RT-PCR even in the presence of common reaction inhibitors. The features of the kit have been selected to enhance virus detection on commonly used sample types. A single-tube format allows for uniform handling and processing for both RNA and DNA viruses.

With the TaqMan® Fast Virus 1-Step Master Mix you can perform reverse transcription and PCR all in one reaction well. It includes:

- AmpliTaq® Fast DNA Polymerase UP, for rapid hot-start PCR
- A rapid thermostable MMLV reverse transcriptase for high sensitivity on viral nucleic acid targets
- Additives to greatly improve success using samples that contain RT-PCR inhibitors, such as blood, anticoagulants, dirt, and stool
- A buffer solution that does not freeze at the –20°C storage temperature

High sensitivity

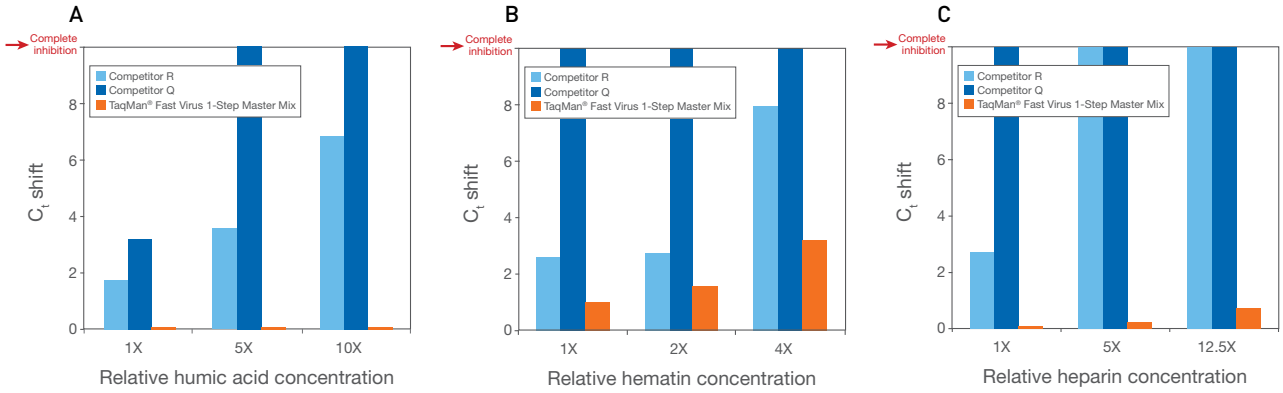
TaqMan® Fast Virus 1-Step Master Mix is optimized for high-sensitivity detection of viral targets. The higher-concentration master mix allows you to set up smaller reactions, which means that you can perform fast cycling protocols and obtain at least the same sensitivity as is expected from standard-cycling, real-time PCR. Alternatively, larger sample input amounts can be added to standard reaction volumes for more accurate quantification of low-titer samples.

Consistent results in the presence of inhibitors

Research samples commonly assayed for viruses include blood, dirt, and tissues. Buffer components and proprietary additives in the TaqMan® Fast Virus 1-Step Master Mix have been optimized to handle RT-PCR inhibitors (see figure) to help ensure consistent performance even with these difficult samples, so that you can be more confident in your results.

Product	Quantity	Cat. No.
TaqMan® Fast Virus 1-Step Master Mix (1 x 1 mL)	200 reactions	4444432
TaqMan® Fast Virus 1-Step Master Mix (5 x 1 mL)	1,000 reactions	4444434
TaqMan® Fast Virus 1-Step Master Mix (1 x 10 mL)	2,000 reactions	4444436

Quantity shown is for number of 20-µL reactions.



Comparison of inhibitor tolerance of TaqMan® Fast Virus 1-Step Master Mix and one-step kits made by other vendors. Three inhibitors of RT-PCR (**A**, humic acid; **B**, hematin; C, heparin) were titrated at three concentrations and added to real-time RT-PCRs with a viral target to assess the magnitude of the shift in C_t values caused by these inhibitors. Graphs show the change in C_t values from a baseline value with no inhibitor present. TaqMan® Fast Virus 1-Step Master Mix is clearly more robust than the other kits tested to humic acid, heparin, and hematin, and real-time RT-PCR results are often achievable with minimal loss of sensitivity, even at concentrations that will completely inhibit other products.

Power SYBR® Green RNA-to-C$_T$™ *1-Step* Kit

High sensitivity and specificity in an easy-to-use, one-step qRT-PCR kit

- Minimized hands-on time with a one-step protocol
- Reduced error and contamination caused by multiple pipetting steps
- Accuracy across a wide dynamic range for dependable performance
- Highly specific and consistent results, even at low quantities of target RNA

The *Power* SYBR® Green RNA-to-C$_T$™ *1-Step* Kit provides high sensitivity and specificity in an easy-to-use, one-step qRT-PCR kit. A novel formulation helps reduce false positive results caused by primer-dimers that are common in SYBR® Green–based assays. This significantly improves data accuracy and enables the reliable detection of low-expressing targets or targets present at low amounts.

Formulation

With the *Power* SYBR® Green RNA-to-C$_T$™ *1-Step* Kit you can perform reverse transcription and PCR all in one reaction tube. It includes:

- AmpliTaq Gold® DNA Polymerase UP (UltraPure) for hot-start PCR to prevent amplification during reaction setup
- ArrayScript™ UP Reverse Transcriptase for highly efficient reverse transcription across a wide range of targets
- A dNTP blend including dUTP which allows resulting amplicons to be degraded by uracil-DNA glycosylase (UDG) to minimize carryover PCR contamination in subsequent reactions (UDG not included)
- RNase inhibitor to decrease degradation of RNA template
- An additive that reduces primer-dimer formation, enhancing specificity
- Passive internal reference based on the proprietary ROX™ dye for increased data precision

Minimal nonspecific amplification

The nonspecific binding of SYBR® Green I dye to primer-dimers can give significant background and show confusing results. The *Power* SYBR® Green RNA-to-C$_T$™ *1-Step* Kit is designed to reduce primer-dimer formation and nonspecific amplification.

High sensitivity and confidence

The *Power* SYBR® Green RNA-to-C$_T$™ *1-Step* Kit formulation helps to enhance sensitivity while reducing nonspecific signals. The kit can detect as little as 0.1 pg of RNA when run on a target gene in total human RNA in a serial dilution. Coupled with the ability to minimize nonspecific amplification, the *Power* SYBR® Green RNA-to-C$_T$™ *1-Step* Kit gives you confidence in data quality.

Product	Quantity	Cat. No.
Power SYBR® Green RNA-to-C$_T$™ 1-Step Kit, Mini-Pack	100 reactions	4391178
Power SYBR® Green RNA-to-C$_T$™ 1-Step Kit	500 reactions	4389986

Quantity shown is for number of 20-µL reactions.

TaqMan® Exogenous Internal Positive Control Reagents

- Predesigned primers and TaqMan® probe eliminate assay design
- Rapid assay development guidelines can minimize optimization time
- Separate components provide flexibility in assay setup

This kit contains a preoptimized internal positive control (IPC) with predesigned primers and TaqMan® probe. The IPC can be spiked into samples to distinguish true target negatives from PCR inhibition. This TaqMan® Exogenous Internal Positive Control Reagents kit allows you to amplify a low-copy target DNA in the same tube with the IPC. Although the target and IPC DNAs may differ in initial copy number, the concentration of the IPC primers in the PCR reaction is limiting so that the amplification efficiency of the target reaction is not compromised.

Compatibility note

Note that this kit has a VIC® Probe with TAMRA™ quencher. Therefore, it is not compatible with the StepOne™ System. It is compatible with the StepOnePlus™ System and all other Applied Biosystems® real-time PCR instruments.

Product	Quantity	Cat. No.
TaqMan® Exogenous Internal Positive Control Reagents (VIC® Dye)	200 reactions	4308323
TaqMan® Exogenous Internal Positive Control Reagents (VIC® Dye), 5-Pack	1,000 reactions	4308321

TaqMan® Exogenous Internal Positive Control Reagents include10X exogenous IPC primer and probe (VIC® dye) mix, 10X exogenous IPC blocking reagent, and 50X exogenous IPC DNA target.

Other control reagents

TaqMan® Ribosomal RNA Control Reagents

TaqMan® Ribosomal RNA Control Reagents are designed to detect the 18S ribosomal RNA (rRNA) gene. Probe and primer sequences are conserved among a diverse group of eukaryotes including human, rat, mouse, *Xenopus*, *Saccharomyces*, *Giardia*, and *Arabidopsis*.

Product	Quantity	Cat. No.
TaqMan® Ribosomal RNA Control Reagents (VIC® Dye)	1,000 reactions	4308329
Includes human control RNA, 50 ng/µL (100 µL), rRNA probe (VIC® dye), rRNA forward primer, and rRNA reverse primer		

TaqMan® ß-Actin Detection Reagents

TaqMan® ß-Actin Detection Reagents provide the necessary components for using ß-actin as a normalization control in applications with human cells.

Product	Quantity	Cat. No.
TaqMan® ß-Actin Detection Reagents	100 reactions	401846
Includes human genomic DNA, 10 ng/µL (100 µL), ß-actin probe (FAM™ dye), ß-actin forward primer, and ß-actin reverse primer		

TaqMan® GAPDH Control Reagents (Human)

The TaqMan® GAPDH Control Reagents (Human) provide the necessary components for using human GAPDH as a normalization control. Either DNA or RNA can be used as a target template.

Product	Quantity	Cat. No.
TaqMan® GAPDH Control Reagents (Human)	100 reactions	402869
Includes human control RNA, 50 ng/µL (100 µL), GAPDH probe (JOE™ dye), GAPDH forward primer, and GAPDH reverse primer		

TaqMan® Rodent GAPDH Control Reagents

The TaqMan® Rodent GAPDH Control Reagents provide the necessary components for using rodent GAPDH as a normalization control in applications with rat, mouse, and Chinese hamster cells.

Product	Quantity	Cat. No.
TaqMan® Rodent GAPDH Control Reagents (VIC® Dye)	1,000 reactions	4308313
Includes rodent control RNA, 50 ng/µL (100 µL), rodent GAPDH probe (VIC® dye), Rodent GAPDH forward primer, and rodent GAPDH reverse primer		

Other enzymes, primers, and reagents

Product	Quantity	Cat. No.
AmpErase® Uracil N-glycosylase (UNG) Includes AmpErase® Uracil N-glycosylase, 100 µL, 1 unit/µL	100 units	N8080096
GeneAmp® dNTP Blend (10 mM) Includes deoxyribonucleotide triphosphates, 10 mM (2.5 mM each dATP, dCTP, dGTP, and dTTP) dissolved in DI water, titrated to pH 7.0 with NaOH	1 mL	N8080260
TaqMan® DNA Template Reagents Includes human DNA standard curve dilution series from 0.6 to 12.0 ng/µL, unknown DNA control, β-actin probe (FAM™ dye), β-actin forward primer, and β-actin reverse primer	100 reactions	401970
TaqMan® Control Total RNA (Human) Includes human RNA, 50 ng/µL	100 µL	4307281
Oligo $(dT)_{16}$ (50 µM) Includes poly(dT)-tailed primer, 50 µM, for reverse transcription of RNA, in 10 mM Tris-HCl, pH 8.3	100 µL	N8080128
Random Hexamers (50 µM) Includes random primers, 50 µM, for reverse transcription of RNA, in 10 mM Tris-HCl, pH 8.3	100 µL	N8080127

Spectral calibration kits

Product	Quantity	Cat. No.
For Applied Biosystems® ViiA™ 7 Real-Time PCR System		
Array card block		
ViiA™ 7 Array Card Spectral Calibration Kit	1 kit	4432314
384-well block		
384-Well Spectral Calibration Plate with FAM™ Dye	1 plate	4432271
384-Well Spectral Calibration Plate with VIC® Dye	1 plate	4432278
384-Well Spectral Calibration Plate with ROX™ Dye	1 plate	4432284
384-Well Spectral Calibration Plate with SYBR® Green Dye	1 plate	4432290
384-Well Spectral Calibration Plate with TAMRA™ Dye	1 plate	4432296
384-Well Spectral Calibration Plate with NED™ Dye	1 plate	4432302
384-Well Region of Interest (ROI) and Background Plates	2 plates	4432320
384-Well Normalization Plates with FAM™/ROX™ and VIC®/ROX™ Dyes	2 plates	4432308
MeltDoctor™ HRM Calibration Plate, 384-Well	1 plate	4425559
96-well block		
96-Well Spectral Calibration Plate with FAM™ Dye	1 plate	4432327
96-Well Spectral Calibration Plate with VIC® Dye	1 plate	4432334
96-Well Spectral Calibration Plate with ROX™ Dye	1 plate	4432340
96-Well Spectral Calibration Plate with SYBR® Green Dye	1 plate	4432346
96-Well Spectral Calibration Plate with TAMRA™ Dye	1 plate	4432352
96-Well Spectral Calibration Plate with NED™ Dye	1 plate	4432358
96-Well Region of Interest (ROI) and Background Plates	2 plates	4432364
96-Well Normalization Plates with FAM™/ROX™ and VIC®/ROX™ Dyes	2 plates	4432370
Fast 96-well block		
Fast 96-Well Spectral Calibration Plate with FAM™ Dye	1 plate	4432389
Fast 96-Well Spectral Calibration Plate with VIC® Dye	1 plate	4432396
Fast 96-Well Spectral Calibration Plate with ROX™ Dye	1 plate	4432402
Fast 96-Well Spectral Calibration Plate with SYBR® Green Dye	1 plate	4432408
Fast 96-Well Spectral Calibration Plate with TAMRA™ Dye	1 plate	4432414
Fast 96-Well Spectral Calibration Plate with NED™ Dye	1 plate	4432420
Fast 96-Well Region of Interest (ROI) and Background Plates	2 plates	4432426
Fast 96-Well Normalization Plates with FAM™/ROX™ and VIC®/ROX™ Dyes	2 plates	4432432
MeltDoctor™ HRM Calibration Plate, Fast 96-Well	1 plate	4425618
For Applied Biosystems® 7900HT Real-Time PCR System		
Applied Biosystems® 7900HT Fast 96-Well Spectral Calibration Kit	3 plates	4351653
7900HT Sequence Detection Systems 96-Well Spectral Calibration Kit	3 plates	4328639
7900HT Sequence Detection Systems 384-Well Spectral Calibration Kit	2 plates	4323977
TaqMan® Array Card Spectral Calibration Kit (for 7900HT)	1 kit	4362745
MeltDoctor™ HRM Calibration Plate, 384-Well	1 plate	4425559
MeltDoctor™ HRM Calibration Plate, Fast 96-Well	1 plate	4425618

Spectral calibration kits, cont.

Product	Quantity	Cat. No.
For Applied Biosystems® 7500 Fast Real-Time PCR System		
Applied Biosystems® 7500 Fast Real-Time PCR System Spectral Calibration Kit I	9 plates	4360788
Applied Biosystems® 7500 Fast Real-Time PCR System Spectral Calibration Kit II (red dyes)	3 plates	4362201
MeltDoctor™ HRM Calibration Plate, Fast 96-Well	1 plate	4425618
For Applied Biosystems® 7500 Real-Time PCR System		
Applied Biosystems® 7500 Real-Time PCR System Spectral Calibration Kit I	9 plates	4349180
Applied Biosystems® 7500 Real-Time PCR System Spectral Calibration Kit II (red dyes)	3 plates	4351151
For StepOnePlus™ Real-Time PCR System		
StepOnePlus™ Real-Time PCR System Spectral Calibration Kit	3 plates	4371435
MeltDoctor™ HRM Calibration Plate, Fast 96-Well	1 plate	4425618
For StepOne™ Real-Time PCR System		
StepOne™ Real-Time PCR System Spectral Calibration Kit	3 plates	4371433

TaqMan® RNase P Instrument Verification Plates

Product	Quantity	Cat. No.
For Applied Biosystems® ViiA™ 7 Real-Time PCR System		
ViiA™ 7 Array Card RNase P Verification Kit	1 kit	4432265
TaqMan® RNase P 384-Well Instrument Verification Plate	1 plate	4455280
TaqMan® RNase P 96-Well Instrument Verification Plate	1 plate	4432382
TaqMan® RNase P Fast 96-Well Instrument Verification Plate	1 plate	4351979
For Applied Biosystems® 7900HT Real-Time PCR System		
7900HT TaqMan® Low Density Array Instrument Verification RNase P Kit	1 kit	4351468
TaqMan® RNase P 384-Well Instrument Verification Plate	1 plate	4323306
TaqMan® RNase P 96-Well Instrument Verification Plate	1 plate	4310982
TaqMan® RNase P Fast 96-Well Instrument Verification Plate	1 plate	4351979
For Applied Biosystems® 7300 & 7500 Real-Time PCR System		
TaqMan® RNase P 96-Well Instrument Verification Plate	1 plate	4350584
For StepOnePlus™ Real-Time PCR System		
TaqMan® RNase P Fast 96-Well Instrument Verification Plate	1 plate	4351979
For StepOne™ Real-Time PCR System		
TaqMan® RNase P Fast 48-Well Instrument Verification Plate	1 plate	4371439
For Applied Biosystems® Fast 7500 Real-Time PCR System		
TaqMan® RNase P Fast 96-Well Instrument Verification Plate	1 plate	4351979

For a complete listing of our calibration kits and verification plates, please go to www.appliedbiosystems.com and look under Products > Real-Time PCR > Instrument Calibration Kits.

TaqMan® Gene Expression Assay selection guide

TaqMan® Gene Expression Assays consist of a pair of unlabeled PCR primers and a TaqMan® probe with a FAM™ or VIC® dye label and minor groove binder (MGB) moiety on the 5´ end, and nonfluorescent quencher (NFQ) dye on the 3´ end. RNA from samples of interest is reverse transcribed into cDNA, and the synthesized cDNA serves as the template for quantitative PCR.

Which TaqMan® Assay or Array Product Is Right for You?

I want to use TaqMan® probe and primer sets (assays) designed by Life Technologies.

Choose from over 1.2 million TaqMan® Assays designed by Life Technologies scientists.

What assay format do you want?	Do you want to choose the assays and configure how assays are arranged on your strip-tube, plate or card?	Use this TaqMan® Assay/Array product
Single tube	NA	TaqMan® Gene Expression Assays
Strip-tube with assay and master mix precombined	NA	Lyophilized TaqMan® Gene Expression Assays with Master Mix
96-well plate	Yes	TaqMan® Array Plates
	No, I want a preconfigured plate for studying a particular disease or pathway	TaqMan® Array Gene Signature Plates
96-well plate with assays and master mix precombined	No, I want a preconfigured plate for studying a particular disease or pathway	Lyophilized TaqMan® Array Plates with Master Mix
384-well microfluidic card (for use only on Applied Biosystems® ViiA™ 7 and 7900HT Real-Time PCR Systems)	Yes	Custom TaqMan® Array Cards
	No, I want a preconfigured card for studying a particular disease or pathway	TaqMan® Array Gene Signature Cards
OpenArray® Plate (for use only on the OpenArray® Real-Time PCR Platform)	Yes	TaqMan® OpenArray® Real-Time PCR Plates

I want to design my own probes and primers.

Submit your own sequences and have Life Technologies make TaqMan® Assays to your specifications.

What assay format do you want?	Do you know the exact sequence you need?	Use this TaqMan® Assay/Array product
Single tube	No	TaqMan® Custom Gene Expression Assays
	Yes	Custom TaqMan® Probes and Primers
96-well plate	Yes/No	TaqMan® Custom Plating Service
384-well plate	Yes/No	

TaqMan® Gene Expression Assays

The industry's largest collection of probe and primer sets

- Gene-specific TaqMan® probe and primer sets for quantitative gene expression studies in 19 different species and select pathogens
- High specificity, high sensitivity, and large dynamic range
- Ready-to-go format—TaqMan® Assays require no optimization, and eliminate the labor, expense, and bioinformatics expertise required to design quantitative real-time PCR assays
- Universal thermal cycling conditions
- Choice of FAM™ or VIC® dye labels

TaqMan® Gene Expression Assays include a compact disc with an electronic assay information file.

TaqMan® Gene Expression Assays are preoptimized PCR primer and probe sets for real-time PCR formulated at 20X or 60X concentration. Life Technologies offers over 1.2 million TaqMan® Gene Expression Assays, the most comprehensive set of predesigned real-time PCR assays available. All TaqMan® Gene Expression Assays have been designed using our validated bioinformatics pipeline and run with the same PCR protocol, eliminating the need for primer design or PCR optimization. The assays are now available with a choice of FAM™ or VIC® reporter dye labels and in four different sizes.

Size	Number of 20-µL reactions, conc.	Reporter dye	Approximate delivery time (business days)	Cat. No. (PL = primer-limited)
Extra Small (Made-to-Order. Minimum order quantity 2—any 2 assays)	75, 20X	FAM™	5–12	4448892
Extra Small (Inventoried)	75, 20X	FAM™	1–6	4453320
Small (Inventoried)	250, 20X	FAM™	1–6	4331182
Small (Made-to-Order)	360, 20X	FAM™ or VIC®	5–12	4351372 4448489 (VIC®) 4448484 (VIC® - PL)
Medium (Made-to-Order)	750, 20X	FAM™ or VIC®	5–12	4351370 4448490 (VIC®) 4448485 (VIC® - PL)
Large (Made-to-Order)	2,900, 60X	FAM™ or VIC®	5–12	4351368 4448491 (VIC®) 4448486 (VIC® - PL)

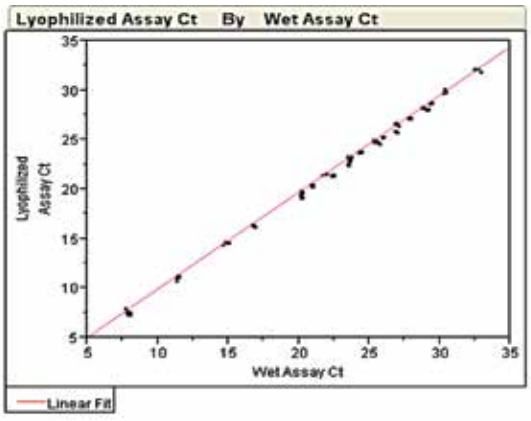
To access our extensive online catalog of TaqMan® Gene Expression Assays go to http://www.appliedbiosystems.com. Assays can be searched using a number of public ID numbers (including RefSeq ID, Entrez Gene ID, or Unigene ID), common gene names, symbols, or aliases, and functional categories and groups (such as kinase, cytokine, and transcription factors).

Lyophilized TaqMan® Gene Expression Assays with Master Mix

Just add sample and go

- Gold standard performance—TaqMan® Gene Expression Assays with Master Mix premixed in optimal quantities for optimal performance
- Limited variability—less pipetting and mixing errors
- Greater convenience—fewer workflow steps: just add sample and go. Helps reduce time spent purchasing, storing, and disposing

Lyophilized TaqMan® Gene Expression Assays with Master Mix are mixtures of TaqMan® probe, primers, and master mix enzyme in strip-tubes or 96-well plates. These Lyophilized TaqMan® Assays simplify the experimental workflow and help limit variability in the assay results because the number of steps is reduced. Lyophilization, also known as freeze drying, is a widely-used technique in food and pharmaceutical industries for room temperature storage and transportation of compounds. We have applied this technique to qPCR so that probe and primers for any gene can be combined with enzyme into a mixture that is stable at room temperature. With this new lyophilized format, you just need to add sample solution into the reaction tube (strip-tubes or 96-well plates) containing the lyophilized assay and enzyme mix, and run on a real-time PCR instrument.



Lyophilized TaqMan® Gene Expression Assays performance. Lyophilized assays and wet assays were run in parallel on a plate with replicates (n = 4). The C_t values between lyophilized and wet assays were similar (R^2 = 0.998; slope 1.02). The standard deviation of the C_t replicates was <0.24.

For ordering information, go to www.appliedbiosystems.com/lyophilizedtaqman.

Custom TaqMan® Expression Assays

Custom quantitative expression assays for any transcript, any species

- Gold standard qPCR assays for quantitative RNA detection
- Powerful online tools for flexible assay design and ordering
- Ready-to-use, preformulated assays designed to run under universal cycling conditions

Custom TaqMan® Expression Assays are the custom version of our original, gold standard 5´ nuclease TaqMan® Assays for relative quantification of RNA expression. Each assay is a mix of forward primer, reverse primer, and FAM® dye-labeled TaqMan® MGB probe. Designed to run under universal oligo concentration and thermal cycling conditions for two-step RT-PCR, these custom assays are simple to use. Just add TaqMan® Universal Master Mix II and your cDNA sample to generate sensitive, reproducible, and truly quantitative RNA expression data.

Two available options

Life Technologies offers two custom expression assay options to help meet your RNA research needs: Custom TaqMan® Gene Expression Assays and Custom Plus TaqMan® RNA Assays. Both options allow you to submit a target sequence to our TaqMan® Assay design pipeline, which will use your input sequence to design custom primer and probe sequences. However, there are several key differences between the two options (see table).

Custom Plus TaqMan® RNA Assays are designed using the full bioinformatic power of the TaqMan® Assay design pipeline, which uses proprietary algorithms that have generated millions of assay designs and is regarded as the benchmark in the industry for qPCR design. If you do not need bioinformatics or QC checks performed on your sequences, then our standard Custom TaqMan® Gene Expression Assays will meet your needs.

Easy ordering

Target sequence submission is easy using the online Custom TaqMan® Assay Design Tool (CADT). Simply select your bioinformatic analysis and assay specificity preferences and enter your target sequence. Sequence submissions are completely confidential; Life Technologies will not share your target sequences or assay sequences with any third parties.

If you do not have a predefined input sequence, the CADT can help you to search for sequences by keyword (gene symbol, accession number, etc.) or by genomic location in a broad range of the most popular and well-studied model species ranging from human and mouse to pig, dog, cow, *Arabidopsis*, and more.

Comparison of Custom TaqMan® Expression Assay products.

Feature	Custom Plus TaqMan® RNA Assays	Custom TaqMan® Gene Expression Assays
Bioinformatic analysis performed on input target sequences	●	
In silico QC performed on assay sequences	●	
Specificity option for gene-level or transcript-level detection	●	
Contains TaqMan® FAM™-MGB probe and 2 unlabeled primers	●	●
Runs under universal thermal cycling conditions	●	●
Assay sequences provided		●
Available for any species	*Available for any species with genome information in Applied Biosystems database	●
Automatic check for predesigned TaqMan® Assays which may detect input sequence	●	●

Product	Conc.	# of 20-µL reactions	Cat. No.
Custom TaqMan® Gene Expression Assays Size			
Small	20X	360	4331348
Medium	20X	750	4332078
Large	60X	2,900	4332079
Custom Plus TaqMan® RNA Assays Size			
Small	20X	360	4441114
Medium	20X	750	4441117
Large	60X	2,900	4441118

Order custom assays at www.appliedbiosystems.com/cadt.

TaqMan® RNase P Detection Reagents (FAM™ dye)

- Predesigned primers and TaqMan® probe eliminate assay design
- Easy-to-use primer and TaqMan® probe mix minimizes assay set-up time
- Rapid Assay Development Guidelines can minimize optimization time
- Includes a passive internal reference for signal normalization

The TaqMan® RNase P Detection Reagents Kit provides the components needed to detect and quantitate genomic copies of the human RNase P gene using the 5´ nuclease assay. The human RNase P gene is a single-copy gene encoding the RNA moiety for the RNase P enzyme. This 5´ nuclease assay employs TaqMan® Universal PCR Master Mix or TaqMan® PCR Core Reagents for PCR using the provided human genomic DNA as template.

Product	Quantity	Cat. No.
TaqMan® RNase P Detection Reagents Kit	100 reactions	4316831

TaqMan® Endogenous Controls

Convenient controls for quantitative gene expression

- Optimized, preformulated, ready-to-use control assays
- Cost-effective gene expression quantitative controls for human, mouse, rat, and other eukaryotic organisms
- Choice of FAM™ or VIC® dye labels

TaqMan® Endogenous Controls are a collection of predesigned probe and primer sets that enable you to normalize the amount of sample RNA or DNA in a reaction. Now you can avoid all the trial and error of selecting controls for most common human, mouse, rat, and eukaryotic genes.

Simple to use

All components of the TaqMan® Endogenous Controls are QC tested, formulated into a single 20X mix, and functionally tested. The controls are not only simple to use, but they are also fully compatible with universal conditions for two-step RT-PCR. Just add TaqMan® Universal Master Mix II and your cDNA sample to generate sensitive, reproducible, and truly quantitative gene expression data on Applied Biosystems instruments including the Applied Biosystems® ViiA™ 7, 7900HT, 7300, 7500, StepOne™,* and StepOnePlus™ Real-Time PCR Systems.

Flexible offering

Each endogenous control is built using our proven 5´ nuclease chemistry. For maximum flexibility, you can choose between two different reporter dyes and two quenchers:

- A FAM™ dye-labeled TaqMan® MGB probe (250 nM, final concentration) and two unlabeled PCR primers (900 nM each)
- A VIC® dye-labeled TaqMan® MGB probe (250 nM, final concentration) and two unlabeled PCR primers (150 nM each: primer-limited)
- A VIC® dye-labeled TAMRA™ probe (250 nM, final concentration) and two unlabeled PCR primers (150 nM each: primer-limited)

Choosing the right endogenous control

Endogenous controls can normalize the expression levels of target genes by correcting differences in the amount of cDNA that is loaded into PCR reaction wells. For best results, verify that the endogenous control is consistently expressed in the sample set to be tested. Endogenous control expression must be uniform across all samples in the study. For multiplexing, ensure that the gene expression level of the endogenous control is greater than that of the target.

Multiplex vs. singleplex PCR

All TaqMan® Endogenous Controls that contain probes labeled with the VIC® reporter dye are primer-limited. This allows multiplexing of TaqMan® Endogenous Controls with target gene expression assays, provided that the control gene is more abundantly expressed than the target gene. All TaqMan® Endogenous Controls that contain probes labeled with the FAM™ reporter dye are not primer-limited and are not intended for multiplexing.

To order from our line of TaqMan® Endogenous Controls, go to www.appliedbiosystems.com.

Product	Dye/quencher	Primer-limited	Conc.	Reactions	Cat. No.
Eukaryotic 18S rRNA	VIC®/MGB	Y	20X	2,500	4319413E
	VIC®/TAMRA™	Y	20X	2,500	4310893E
	FAM™/MGB	N	20X	125	4333760T
	FAM™/MGB	N	20X	500	4333760F
Human ACTB (ß-actin)	VIC®/MGB	Y	20X	1,000	4326315E
	VIC®/TAMRA™	Y	20X	1,000	4310881E
	FAM™/MGB	N	20X	125	4333762T
	FAM™/MGB	N	20X	500	4333762F
Human B2M (ß-2-microglobulin)	VIC®/MGB	Y	20X	2,500	4326319E
	VIC®/TAMRA™	Y	20X	2,500	4310886E
	FAM™/MGB	N	20X	125	4333766T
	FAM™/MGB	N	20X	500	4333766F
Human GAPD (GAPDH)	VIC®/MGB	Y	20X	1,000	4326317E
	VIC®/TAMRA™	Y	20X	1,000	4310884E
Human GAPD (GAPDH)	FAM™/MGB	N	20X	125	4333764T
	FAM™/MGB	N	20X	500	4333764F
Human GUSB (ß-glucuronidase)	VIC®/MGB	Y	20X	2,500	4326320E
	VIC®/TAMRA™	Y	20X	2,500	4310888E
	FAM™/MGB	N	20X	125	4333767T
	FAM™/MGB	N	20X	500	4333767F
Human HPRT1	VIC®/MGB	Y	20X	2,500	4326321E
	VIC®/TAMRA™	Y	20X	2,500	4310890E
	FAM™/MGB	N	20X	125	4333768T
	FAM™/MGB	N	20X	500	4333768F
Human PGK1 (phosphoglycerate kinase 1)	VIC®/MGB	Y	20X	2,500	4326318E
	VIC®/TAMRA™	Y	20X	2,500	4310885E
	FAM™/MGB	N	20X	125	4333765T
	FAM™/MGB	N	20X	500	4333765F
Human PPIA (cyclophilin A)	VIC®/MGB	Y	20X	2,500	4326316E
	VIC®/TAMRA™	Y	20X	2,500	4310883E
	FAM™/MGB	N	20X	125	4333763T
	FAM™/MGB	N	20X	500	4333763F
Human RPLO (large ribosomal protein)	VIC®/MGB	Y	20X	2,500	4326314E
	VIC®/TAMRA™	Y	20X	2,500	4310879E
	FAM™/MGB	N	20X	125	4333761T
	FAM™/MGB	N	20X	500	4333761F
Human TBP (TATA box–binding protein)	VIC®/MGB	Y	20X	2,500	4326322E
	VIC®/TAMRA™	Y	20X	2,500	4310891E
	FAM™/MGB	N	20X	125	4333769T
	FAM™/MGB	N	20X	500	4333769F
Human TFRC (CD71) (transferrin receptor)	VIC®/MGB	Y	20X	2,500	4326323E
	VIC®/TAMRA™	Y	20X	2,500	4310892E
	FAM™/MGB	N	20X	125	4333770T
	FAM™/MGB	N	20X	500	4333770F
Mouse GAPD (GAPDH)	VIC®/MGB	Y	20X	2,500	4352339E
Mouse ACTB (ß-actin)	VIC®/MGB	Y	20X	2,500	4352341E
Rat GAPD (GAPDH)	VIC®/MGB	Y	20X	2,500	4352338E
Rat ACTB (ß-actin)	VIC®/MGB	Y	20X	2,500	4352340E

*The StepOne™ Real-Time PCR System is only compatible with MGB quencher probes.

TaqMan® Array Gene Signature Plates

Select TaqMan® Assays preloaded in 96-well plates

- Affordable—minimum order of one plate; ideal for small, medium, or large projects
- Quick delivery—on your bench in days
- Convenient—dried PCR primer and TaqMan® probe sets supplied in a 96-well plate; just add master mix and your sample
- Available in Fast and standard formats

These 96-well plates come in Fast and standard formats, and are preconfigured with the most appropriate TaqMan® Gene Expression Assays for a specific biological process, pathway, or disease state. Each plate contains predefined assays and endogenous controls dried-down in the wells, ready for accurate assessment of an entire gene signature in one simple experiment. Extensive tests have found no difference in performance between TaqMan® Assays supplied in the traditional liquid format and assays dried into a TaqMan® Array Plate (Figure 1).

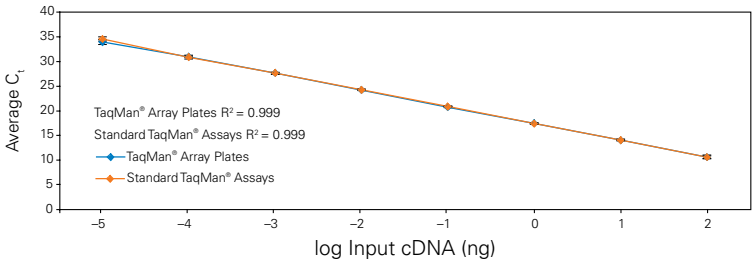


Figure 1. Linear range of TaqMan® Array Plates. TaqMan® Array Plates have the same performance as standard TaqMan® Assays (wet) on a 96-well plate. The linear dynamic range is maintained even down to low cDNA inputs, showing the assays are unaffected during the dry down process of the TaqMan® Array Plate. Each plate contained one assay (18s Hs99999901_s1) and eight replicates for each cDNA input.

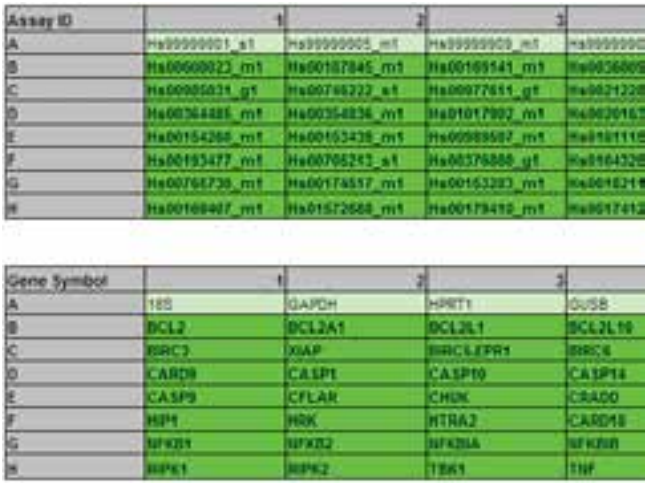


Figure 2. Example of TaqMan® Array Gene Signature Plate layout.

Simple to use

Ideal for projects such as validation of microarray and RNAi data as well as pathway studies, the plates include just the right amount of TaqMan® Assay in each well (i.e., enough for one 20-µL standard reaction or one 10-µL Fast reaction). The plates can be stored at room temperature or at 4°C. When you are ready to run your assays, simply add PCR master mix and your cDNA sample, seal the plate, and begin cycling on a real-time PCR instrument.

Plate map enables proper data labeling and easy re-ordering

Each pack of TaqMan® Gene Signature Array Plates is shipped with a CD containing the plate configuration map (Figure 2) along with details of each TaqMan® Assay. This information is imported into the real-time PCR instrument to enable proper labeling of experimental data and easy access to Assay ID numbers for simple reordering.

Compatibility

TaqMan® Array Plates must be run on a real-time PCR instrument with 96-well plate capability. TaqMan® Array Standard Plates have been optimized for use with TaqMan® Gene Expression Master Mix but can be used with TaqMan® Universal PCR Master Mix. Standard plates have been validated for use on Applied Biosystems® 7000, 7300, 7500, 7900HT Fast, and ViiA™ 7 Real-Time PCR Systems using standard 96-well blocks, standard PCR cycling conditions, and 20-µL reaction volumes.

TaqMan® Array Fast Plates have been optimized for use with TaqMan® Fast Universal PCR Master Mix but can be used with TaqMan® Gene Expression Master Mix or TaqMan® Universal PCR Master Mix. Fast plates are validated for use on Applied Biosystems® StepOnePlus™, 7500 Fast, 7900HT Fast, and ViiA™ 7 Real-Time PCR Systems using Fast 96-well blocks, Fast PCR cycling conditions, and 10-µL reaction volumes.

To choose from over 130 different gene signature plates, go to www.appliedbiosystems.com/signatureplates.

Custom TaqMan® Array Plates

Your choice of TaqMan® Assays preloaded in 96-well plates

- Easy to customize—use our online ordering tool to place assays on your Fast or standard plate, choose the number of replicates and controls, and more
- Affordable—order as few as six plates
- Efficient—plates arrive typically in less than two weeks, require as little as ten minutes or less of hands-on prep time
- Convenient—just add master mix and your sample directly into the 96-well plates and run
- Gold standard TaqMan® Assay quality

TaqMan® Array Plates are a cost-effective and convenient 96-well format complementing our line of individual TaqMan® Gene Expression Assays and prespotted 384-well TaqMan® Arrays. Customize any of the five available Fast or standard plate formats with dried-down TaqMan® Assays by positioning each in specific well locations. This flexibility enables an efficient means to screen biological samples for the presence and amount of specified gene targets expressed in human, mouse, rat, canine, and *Rhesus* species. When you are ready to run your assays, simply add PCR master mix and your cDNA sample, seal the plate, and begin cycling on a real-time PCR instrument with standard 96-well plate capability.

Product	Standard Plate Cat. No.	Fast Plate Cat. No.
TaqMan® Array Plate, Custom Format 96	4391524	4413255
TaqMan® Array Plate, Custom Format 96 Plus Candidate Endogenous Control Genes	4391525	4413256
TaqMan® Array Plate, Custom Format 48	4391526	4413257
TaqMan® Array Plate, Custom Format 48 Plus Candidate Endogenous Control Genes	4391527	4413258
TaqMan® Array Plate, Custom Format 32 TaqMan® Array Plate, Custom Format 32 Plus	4391528	4413259
Candidate Endogenous Control Genes	4391529	4413260
TaqMan® Array Plate Format 16	4413264	4413261
TaqMan® Array Plate Format 16 Plus Candidate Endogenous Control Genes	4413265	4413262
TaqMan® Array Plate Format 8	4413266	4413263

TaqMan® Custom Plating Service

Configure 96- and 384-well plates with any TaqMan® Assays

- Set up custom configurations for a variety of applications:
  - TaqMan® Gene Expression Assays (Inventoried, Made-to-Order, and Custom)
  - TaqMan® SNP Genotyping Assays (Predesigned, Custom, and DME)
  - TaqMan® Copy Number Assays (Predesigned, Custom Plus, or Custom) either alone or in duplex with TaqMan® Copy Number Reference Assays
  - Custom TaqMan® Probes and Primers
  - TaqMan® Non-coding RNA Assays (Inventoried, Made-to-Order)
  - TaqMan® MicroRNA Assays (Inventoried)
- Select from a variety of reaction volumes
- Receive in dried or liquid formulation
- Use with a wide range of instruments including most Applied Biosystems® real-time PCR systems

The TaqMan® Custom Plating Service offers the convenience of preplated TaqMan® Gene Expression Assays, SNP Assays, Copy Number Assay, Custom Assays, and Custom Probe/Primer Sets in 96- or 384-well plates.

Target applications

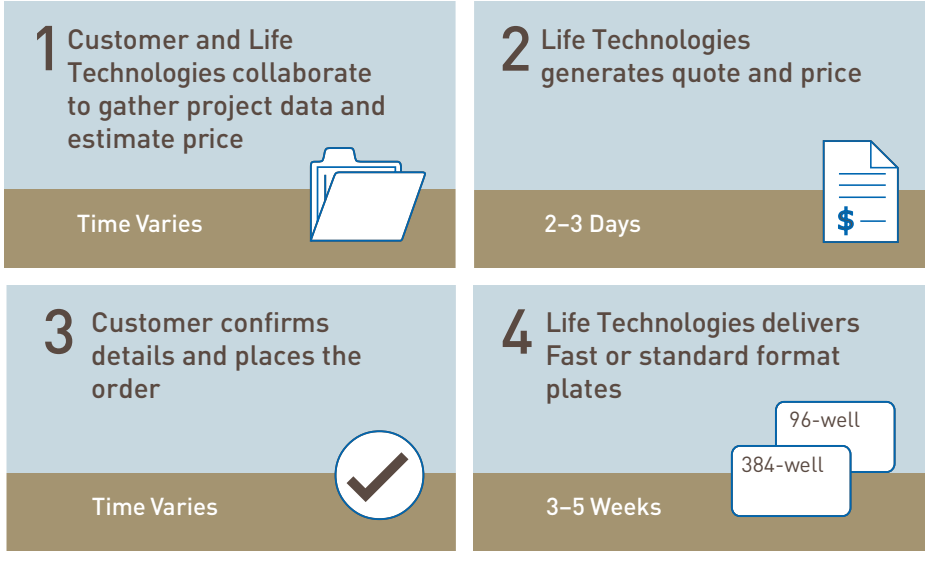
This cost-effective custom plating service gives you the flexibility and reliability you need for your screening applications. Whether you are looking for biomarkers, testing for toxicity, searching for microbes, genotyping, analyzing copy number variations, or performing other high-throughput screens (including validation of RNA interference experiments), TaqMan® Assay products provide the ideal solution. The custom plating service enables you to focus on your experimental design and results, while saving labor and time and reducing errors from pipetting or cross-contamination. Your local sales representative can assist you in selecting the assays and format that best suits your research.

Trusted technology

TaqMan® Assay chemistry offers high sensitivity, specificity, and reproducibility over a broad dynamic range for quantitative gene expression studies. Now you can also order any combination of TaqMan® Assay products preloaded in 96- or 384-well plates.

How TaqMan® custom plating works

To initiate the custom plating process, contact your local Life Technologies Sales Representative, or email us at customplating@appliedbiosystems.com.



How to order using the TaqMan® Custom Plating Service.

For more information, go to www.appliedbiosystems.com.

Custom TaqMan® Array Microfluidic Cards

Your choice of TaqMan® Gene Expression Assays preloaded in 384-well microfluidic cards

- Get the sensitivity and specificity of TaqMan® Assays in an easy-to-use microfluidic card format
- Create the perfect card by designing the Custom TaqMan® Array Microfluidic Card that meets your specific need
- Load 384 wells typically in less than five minutes without using liquid-handling robots or multi-channel pipettors
- Validate all of the "interesting" hits off a microarray quickly and economically
- Standardize the screening of gene panels across many samples and laboratories

Custom TaqMan® Array Microfluidic Cards enable you to perform 384 simultaneous real-time PCR reactions without the need to use liquid-handling robots or multichannel pipettors to fill the card. This low- to medium-throughput card allows for 1–8 samples to be run in parallel against twelve 384 TaqMan® Gene Expression Assay targets that are preloaded into each of the wells on the card. Custom TaqMan® Array Cards are completely customizable. Choose from over 50,000 TaqMan® Gene Expression Assays designed for human, mouse, and rat genes. A minimum order of 10 cards is required for all formats.

Simple and straightforward loading

The microfluidic card has eight sample-loading ports each connected to 48 reaction wells. Simply pipette your sample (premixed with TaqMan® Universal PCR Master Mix) into each sample-loading port and briefly centrifuge. Within minutes, your 384-well TaqMan® Array Card is ready to run on the Applied Biosystems® ViiA™ 7 or 7900HT Fast Real-Time PCR System.

Ultimate microarray validation tool

TaqMan® Array Cards are an ideal tool for validating the tens or hundreds of "interesting" hits from microarray experiments, because TaqMan® Array Cards can be customized to include up to 384 of those hits in one easy-to-use card. Custom TaqMan® Array Cards enable you to accomplish the validation necessary to arrive at the right answer easily and affordably.

Ideal screening technology

TaqMan® Array Cards are ideal for screening biomarkers and toxicology panels and for analyzing pathways, target classes, and complete disease sets. Because TaqMan® Array Cards are loaded without the need for liquid-handling robotics, you get standardized results with low variability across many users and laboratories. Plus, TaqMan® Gene Expression Assays, with excellent specificity and sensitivity in real-time PCR, are preloaded into the card, enabling reliable performance you can trust.

TaqMan® Array Card	Number of targets and controls	Number of assay replicates	Number of samples per card	Minimum order size	Cat. No.
Format 12	11 + 1	4	8	10	4342247
Format 16	15 + 1	3	8	10	4346798
Format 24	23 + 1	2 (4)	8 (4)	10	4342249
Format 32	31 + 1	3	4	10	4346799
Format 48	47 + 1	1 (2)	8 (4)	10	4342253
Format 64	63 + 1	3	2	10	4346800
Format 96a	95 + 1	1 (2)	4 (2)	10	4342259
Format 96b	95 + 1	4	1	10	4342261
Format 192	191 + 1	2	1	10	4346802
Format 384	380 + 1	1	1	10	4342265

TaqMan® Array Gene Signature Microfluidic Cards

Select TaqMan® Gene Expression Assays preloaded in 384-well microfluidic cards

- Choose from over 20 off-the-shelf, focused gene panels for quick delivery
- Available in smaller and more convenient package sizes
- Get quantitative gene expression data from low-expressing genes
- For use only with Applied Biosystems® ViiA™ or 7900HT Real-Time PCR Systems

TaqMan® Array Gene Signature Microfluidic Cards are predesigned TaqMan® Array Cards containing select TaqMan® Gene Expression Assays matching genes specific to disease target classes or pathways to facilitate drug discovery, disease research, and pathway analysis. The genes are chosen from pathway analysis tools, published articles, and collaborator and customer input. Reliable, sensitive TaqMan® Array Gene Signature Cards are ideal for gene expression analysis of comprehensive sets of gene targets related to a disease or pathway because they provide quantitative, reproducible results. They are cost-effective, affordable, and suitable for most throughput needs.

Product	Quantity	Cat. No.
Human ABC Transporter Card	4 cards	4378700
Human Alzheimer's Card	4 cards	4378713
Mouse Alzheimer's Card	4 cards	4378714
Human Angiogenesis Card	4 cards	4378710
Human Apoptosis Card	4 cards	4378701
Human Endogenous Control Card	2 cards	4367563
Mouse Endogenous Control Card	2 cards	4378702
Rat Endogenous Control Card	2 cards	4378704
Human GPCR Card	4 cards	4367785
Mouse GPCR Card	4 cards	4378703
Rat GPCR Card	4 cards	4378709
Human Nuclear Receptor Card	4 cards	4379961
Human Protein Kinase Card	4 cards	4367784
Human Immune Card	4 cards	4370573
Mouse Immune Card	4 cards	4367786
Human Inflammation Card	4 cards	4378707
Rat Inflammation Card	4 cards	4378708
Human Phosphodiesterase Card	4 cards	4378705
Rat Phosphodiesterase Card	4 cards	4378706
Human Stem Cell Pluripotency Card	4 cards	4385344
Mouse Stem Cell Pluripotency Card	4 cards	4385363
Rat Hepatocarcinogenicity Card	4 cards	4465484
Human Viral Pathogen Screening Card	Inquire	4465485

TaqMan® OpenArray® Real-Time PCR Plates

TaqMan® Gene Expression Assays in a high-throughput format

- More replicates—each plate generates over 2,500 data points, enabling technical replicates for better statistical power
- Simple workflows—each OpenArray® Plate comes preloaded with the assays of your choice—just add sample and master mix, then cycle and image
- Experimental flexibility—specify your desired inventoried TaqMan® Assays, and select the sample/assay format that best meets your project needs
- Reliable and economical—uses gold standard TaqMan® MGB probe chemistry in a nanofluidic design that reduces reagent usage

TaqMan® OpenArray® Real-Time PCR Plates consist of two world-class technologies: TaqMan® Gene Expression Assays and OpenArray® technology. TaqMan® Gene Expression Assays use one specific MGB probe and two PCR primers to provide highly robust and accurate gene expression analyses. OpenArray® technology uses nanoliter fluidics for higher sample throughput and lower costs per data point.

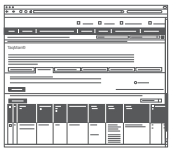
Simple workflow

TaqMan® OpenArray® Real-Time PCR Plates are delivered containing your defined selection of inventoried TaqMan® Gene Expression Assays preloaded into the OpenArray® Plate. Simply mix your sample and master mix, load onto the OpenArray® Plate, seal, cycle, and image. *Continued on next page.*

Performance specifications for the TaqMan® OpenArray® Real-Time PCR Plates.

Feature	Performance
Precision of replicates at 100 copies	C_t standard deviation <0.25
Specificity based on no-template control	No demonstrable amplification
No amplification in empty wells	>99% holes
Loading time of three OpenArray® Plates (8,064 reactions)	<30 min
Time from DNA or cDNA to real-time data	~3 hr
Throughput of one technician in one day	>30,000 reactions (576 samples)

1. Select Assays or Panels & Order Products

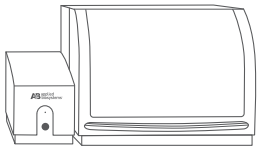


Visit **www.appliedbiosystems.com** to select assays and OpenArray® Plate formats for custom plates, or to select from pre-designed OpenArray® Pathway Panels or TaqMan® OpenArray® Digital PCR Plates. Digital PCR plates are pre-treated to accept your primers and samples in your lab. All other OpenArray® Plates are delivered with assays dried down in the plate through-holes.

2. Add Sample & Master Mix

For gene expression and genotyping applications, mix cDNA or DNA samples with Master Mix in 384-Well Sample Plates. For digital PCR applications, mix primers, samples and Master Mix in 384-Well Sample Plates.

3. Load Samples

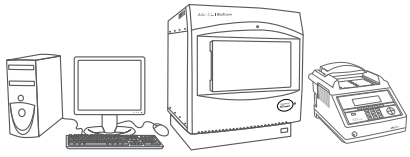


Load sample mixes onto an OpenArray® Plate with the AccuFill™ System.

4. Insert Into Case & Seal

Insert the OpenArray® Plate into the OpenArray® case filled with immersion fluid, and seal with glue.

5. Cycle, Image & Analyze Data



For genotyping applications, cycle in the Dual Flat Block GeneAmp® PCR System 9700 and transfer to the OpenArray® Real-Time PCR Instrument for imaging. For real-time and digital PCR applications, cycle and image on the OpenArray® Real-Time PCR Instrument and analyze data.

High throughout and high performance

The OpenArray® Real-Time PCR System provides high sample throughput for mid-density gene expression. In a single day, 12 TaqMan® OpenArray® Real-Time PCR Plates, providing 32,256 real-time PCR reactions can be run, screening up to 576 samples without the use of robotics. The OpenArray® System is ideal for academic research; AgBio validating complex disease associations; marker-assisted breeding; pharmaceutical target profiling and screening; or other studies requiring large sample sizes, high performance, and reproducibility.

Flexible formats

Each TaqMan® OpenArray® Real-Time PCR Plate contains 3,072 through-holes arranged in 48 subarrays of 64 through-holes each (Figures 1 and 2). For OpenArray® Real-Time PCR Plates, 8 through-holes per subarray are used as controls, yielding a total of 56 available through-holes per subarrray. The OpenArray® AccuFill™ System can precisely load one sample onto each subarray. Five plate format options are available (see table).

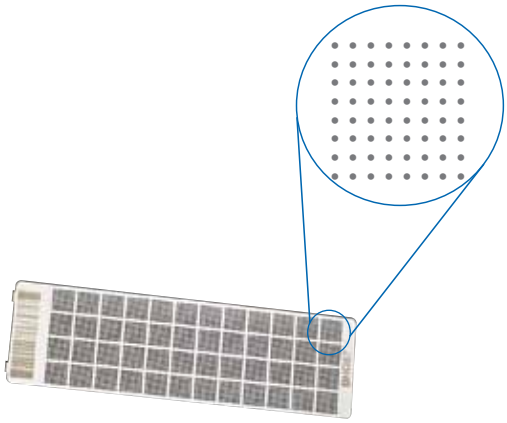


Figure 1. TaqMan® OpenArray® Real-Time PCR Plate.

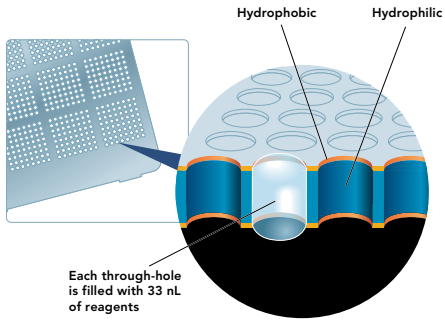


Figure 2. Plate anatomy.

TaqMan® OpenArray® Real-Time PCR Plate format options.

TaqMan® OpenArray® Real-Time PCR Plate formats	Number of samples/plate	Minimum order	Total number of samples/ minimum order
18 assays	48 samples	10	480
56 assays	48 samples	10	480
112 assays	24 samples	10	240
168 assays	16 samples	10	160
224 assays	12 samples	10	120

Product	Quantity	Cat. No.
OpenArray® Real-Time PCR Plates	18 assays in triplicate, up to 48 samples per plate	4456272
	56 assays, up to 48 samples per plate	4456276
	112 assays, up to 24 samples per plate	4456268
	168 assays, up to 16 samples per plate	4456270
	224 assays, up to 12 samples per plate	4456274

Custom TaqMan® probes and primers

Design your own TaqMan® probe and primer sets

- Choice of dye labels, quenchers, and synthesis scale
- Available for any species or organism
- For use in quantitative gene expression, SNP genotyping, other allelic discrimination applications, and pathogen detection

When you know the exact sequences you need for your TaqMan® probes and primers, Life Technologies can synthesize them for you. As a market leader in real-time PCR, our high-quality custom products can be used in all your real-time and end-point applications. These products offer you maximum flexibility if you prefer to optimize your own reaction formulation or if you simply prefer to buy in bulk.

TaqMan® probe usage chart for gene expression.

TaqMan® probe	Reaction volume = 20 µL (384-well plates)
Amount (pmol)	Number of reactions
6,000	1,200
20,000	4,000
50,000	10,000

For use on Applied Biosystems® real-time PCR systems.
For gene expression, the minimum number of reactions obtained from our TaqMan® probe products was calculated based on Universal Assay conditions, primer concentration of 900 nM, and probe concentration of 250 nM. The numbers are shown for 20-µL reaction volumes.

Product	Approximate delivery time (business days)	Quantity	Cat. No.
TaqMan® TAMRA™ Probes	4 to 5 days	6,000 pmol*	450025
	4 to 5 days	20,000 pmol*	450024
	4 to 5 days	50,000 pmol*	450003

TaqMan® probes are available with a choice of 5´ fluorescent label (6-FAM™, VIC® or TET™ dyes*) and 3´ quencher (TAMRA™). All probes are HPLC-purified and sequence-verified by mass spectrometry.

TaqMan® MGB Probes	6 to 7 days	6,000 pmol*	4316034
	6 to 7 days	20,000 pmol*	4316033
	6 to 7 days	50,000 pmol*	4316032

TaqMan® MGB probes are available with a choice of 5´ fluorescent label (6-FAM™, VIC®, TET™*, or NED™ dyes†) and a 3´ minor groove binder/nonfluorescent quencher. All probes are HPLC-purified and sequence-verified by mass spectrometry.

Sequence Detection Primers	3 days	10,000 pmol*	4304970
	4 days	80,000 pmol*	4304971
	4 days	130,000 pmol*	4304972

Sequence Detection Primers are provided purified by desalting for use in sequence detection with all Applied Biosystems® real-time PCR systems. Primers can be provided at a constant concentration in water or 1X TE buffer, or shipped dry.

Quantities are based on an average oligonucleotide length of 23 base pairs.

* Please note that filter-based instruments such as Applied Biosystems® 7300/7500/7500 Fast Real-Time PCR Systems are not supplied with calibration plates for TET™ dye. These instruments may be custom-calibrated to use TET™ dye in a singleplex reaction, but TET™ dye should not be used in a multiplex reaction with either FAM™ or VIC® dyes, as TET™ will not be distinguished by these instruments.

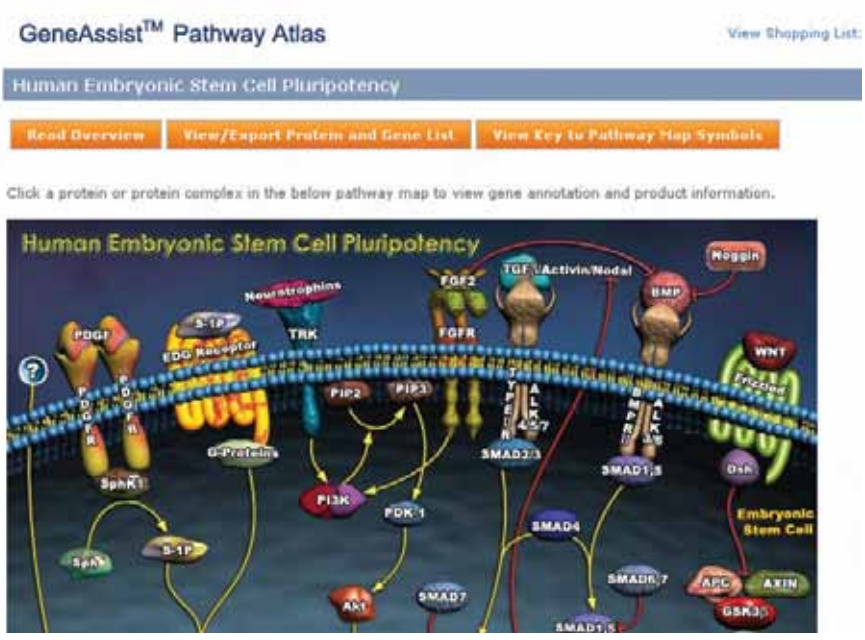
† Please note that the Applied Biosystems® 7500 Real-Time PCR System is optimized for use with NED™-labeled probes. Probes labeled with NED™ will give lower signal intensity on other real-time instrument systems than probes labeled with 6-FAM™, VIC®, or TET™ 3´ label.

GeneAssist™ Pathway Atlas

Quick generation of pathway-associated TaqMan® Gene Expression Assays

- Search or browse for TaqMan® Gene Expression Assays or Ambion® *Silencer*® siRNAs by biological pathway
- Easily export genes in a specific pathway to order Custom TaqMan® Array Cards and TaqMan® Array Plates
- Order TaqMan® Gene Expression Assays and *Silencer*® siRNAs in a single transaction online, by fax, or email

The GeneAssist™ Pathway Atlas is a free online tool that provides a set of more than 350 interactive, easy-to-understand signal transduction, metabolic, and disease state pathway maps, as well as their descriptions and relevant references. When a protein is selected from one of the maps, a detailed view appears showing additional gene information along with the recommended TaqMan® Gene Expression Assays and corresponding *Silencer*® siRNAs for studying that particular gene.



The GeneAssist™ Pathway Atlas online tool.

To access the GeneAssist™ Pathway Atlas, go to www.appliedbiosystems.com/pathway.

To order Custom TaqMan® Array Cards using your exported gene lists, go to www.appliedbiosystems.com/arrayplates.

Custom TaqMan® Assay Design Tool

When you want to design a TaqMan® Assay

The Custom TaqMan® Assay Design Tool allows you to order Custom TaqMan® SNP Genotyping Assays, Custom Plus TaqMan® RNA Assays, and Custom TaqMan® Gene Expression Assays by entering sequences and then submitting for assay design. Upon notification of successful assay design, simply add the desired custom assays to your shopping basket.

To use the Custom TaqMan® Assay Design Tool, go to www.appliedbiosystems.com/cadt

TaqMan® MicroRNA and Non-coding RNA Assay selection guide

The TaqMan® Non-coding RNA Assays family quantitates small and non-coding RNA with the specificity and sensitivity of TaqMan® Assay chemistry. A simple, two-step protocol requires only reverse transcription with a miRNA-specific primer, followed by quantitative real-time PCR (qPCR) with TaqMan® probes.

Which TaqMan® MicroRNA or Non-coding RNA Assay or Array product is right for you?

I want to use TaqMan® probe and primer sets (assays) designed by Life Technologies.

Choose from a variety of TaqMan® Assays designed by Life Technologies scientists based on format and research area.

What assay format do you want?	What type of research are you doing?	Use this TaqMan® Assay/Array product
Single tube	microRNA	TaqMan® MicroRNA Assays
	Pri-miRNA (human/rodent)	TaqMan® Pri-miRNA Assays
	Long non-coding RNA (human/rodent)	TaqMan® Non-coding RNA Assays
384-well microfluidic card (for use only on Applied Biosystems® ViiA™ 7 and 7900HT Real-Time PCR Systems)	Profiling using limited samples or material (human/rodent)	TaqMan® Array MicroRNA Cards
OpenArray® Plate (for use only on the OpenArray® Real-Time PCR System)	Large-scale profiling (human/rodent)	TaqMan® OpenArray® MicroRNA Panels

I want to design my own primers and probes.

Submit your own sequences and have Life Technologies make TaqMan® Assays to your specifications.

What assay format do you want?	Use this TaqMan® Assay/Array product
Single tube	Custom TaqMan® Small RNA Assays

TaqMan® MicroRNA Assays

Quantitative microRNA analysis with state-of-the-art TaqMan® Assay performance

- Highly specific—quantitate only mature microRNAs, not precursors
- Sensitive—conserve limited samples; requires only 1-10 ng of total RNA or equivalent
- Fast, simple, and scalable—two-step quantitative RT-PCR assay provides high-quality results in less than three hours

TaqMan® MicroRNA Assays quantitate microRNAs (miRNAs) with the specificity and sensitivity of TaqMan® Assay chemistry. A simple, two-step protocol requires only reverse transcription with a miRNA-specific primer, followed by real-time PCR with TaqMan® probes. TaqMan® MicroRNA Assays are available for a range of species including human, mouse, rat, *Drosophila*, *C. elegans*, and *Arabidopsis*. We continuously update the annotation and expand our miRNA assays collection for these species in alignment with the Sanger miRBase Registry.

Product	No. of reactions	Cat. No.
TaqMan® MicroRNA Assays		
Small-Scale (Inventoried)	50–150	4427975
Extra Small-Scale (Made-to-Order)	25–75	4440885
Small-Scale (Made-to-Order)	50–150	4440886
Medium-Scale (Made-to-Order)	750	4440887
Large-Scale (Made-to-Order)	2,900	4440888

Custom TaqMan® Small RNA Assays

Custom TaqMan® Small RNA Assays let you "create" your own TaqMan® Assay when a predesigned TaqMan® MicroRNA Assay is not offered. Simply submit your target sequence online, and we'll design and manufacture an assay for optimal performance with your target.

Product	No. of reactions	Cat. No.
Custom TaqMan® Small RNA Assays		
Extra Small-Scale	25–75	4440418
Small-Scale	50–150	4398987
Medium-Scale	750	4398988
Large-Scale	2,900	4398989

TaqMan® Non-coding RNA Assays

Specific and reproducible quantification of long non-coding RNA transcript expression levels

- Provides high sensitivity, specificity, dynamic range, and unsurpassed data quality
- Delivers faster time-to-results
- Requires no design expertise
- Works under universal cycling conditions
- Detects only non-coding RNA transcripts

TaqMan® Non-coding RNA Assays provide a highly reliable way to measure expression of long, non-coding transcripts. These assays are based on the same gold standard TaqMan® Assay technology that is used in the TaqMan® Gene Expression Assays collection. With TaqMan® Assay technology, you get the high specificity and sensitivity you need for quantification and validation studies.

Detect only the target non-coding transcript

One of the main differences between TaqMan® Gene Expression Assays and TaqMan® Non-coding RNA Assays is that the non-coding assays may detect only the target non-coding transcript. Alternatively, coding assays are designed to detect one or more coding transcripts of the same gene. TaqMan® Non-coding Assays were designed using the most up-to-date ncRNA annotations from NCBI and RNAdb. To facilitate the microarray validation workflow, we have also designed a number of assays for transcripts that are available on Invitrogen™ NCode™ Non-coding RNA Arrays.

Product	No. of 20-µL reactions	Cat. No.
TaqMan® Non-coding RNA Assay (Made-to-Order; single-tube 20X reaction mix)		
Small-Scale	360	4426961
Medium-Scale	750	4426962
Large-Scale	2,900	4426963

TaqMan® Pri-miRNA Assays

Detect the origins of microRNA expression

- Highly specific—quantitate microRNA transcription from a single genomic locus
- Fast, simple, and scalable—two-step RT-qPCR provides high-quality results
- A new perspective—measure microRNA expression at the gene level

TaqMan® Pri-miRNA Assays bring the same high sensitivity, superior specificity, and broad linear dynamic range for which all Applied Biosystems® TaqMan® Genomic Assays are renowned, to the detection of pri-miRNA tra scripts. As a result of their high sensitivity, TaqMan® Pri-miRNA Assays require minimal sample input—as little as 1 ng of total RNA can quantify expression from moderate or high-expression microRNA (miRNA) loci.

Applications

TaqMan® Pri-miRNA Assays are ideal for addressing the following key areas of miRNA expression and functionality:

- Regulation of miRNA "gene" transcription
- Regulation of miRNA biogenesis
- Mapping of pri-miRNA transcripts

Assay design and selection

Since pri-miRNA transcripts have not been exhaustively mapped, assays are designed within close proximity to each stem-loop sequence represented in the Sanger miRBase data repository. This ensures accurate measurement of the pri-miRNA transcript of interest. Additionally, each publicly available stem-loop can be matched to both a TaqMan® Pri-miRNA Assay and a TaqMan® MicroRNA Assay, enabling independent quantitation of RNA sequences produced at either end of the miRNA maturation pathway to be quantified independently.

Product	No. of 20-µL reactions	Cat. No.
TaqMan® Pri-miRNA Assays (Made-to-Order; single-tube 20X reaction mix)		
Small-Scale	360	4427012
Medium-Scale	750	4427013
Large-Scale	2,900	4427014

Megaplex™ Primer Pools

Ideal for microRNA profiling and expression analysis

- Highly specific—quantitate only the biologically active mature miRNAs
- Highly streamlined workflow reduces the number of RT (reverse transcription) reactions per sample
- Significantly reduced sample consumption, particularly with the optional preamplification step
- Comprehensive coverage is ideal for human, mouse, and rat miRNA profiling

Whether your profiling experiment requires ultimate sensitivity, broad coverage, or both, Megaplex™ Primer Pools provide the flexibility to reach your research goals. Megaplex™ Primer Pools provide comprehensive coverage of Sanger miRBase for human and rodent species, and when used with TaqMan® OpenArray® MicroRNA Panels or TaqMan® MicroRNA Arrays, offer an ideal microRNA (miRNA) profiling solution.

- Megaplex™ RT Primers are pools of novel stem-looped RT primers that streamline the profiling of hundreds of miRNA targets in a single experiment and reduce the number of RT reactions and the amount of total RNA required to generate a comprehensive miRNA expression profile
- Optional Megaplex™ PreAmp Primers significantly enhance the ability to detect low-expressed miRNAs, enabling the generation of a comprehensive expression profile from as low as 1 ng of input total RNA

Megaplex™ Primer Pools can be used with individual TaqMan® MicroRNA Assays, TaqMan® Array MicroRNA Cards, or TaqMan® OpenArray® MicroRNA Panels. The cards and panels provide all the advantages of TaqMan® MicroRNA Assays in a convenient preconfigured 384-well microfluidic card or 3,072-well OpenArray® Plate format, respectively. The content of each card or panel is matched to the respective Megaplex™ Primer Pools and contains unique TaqMan® MicroRNA Assays, reducing set-up time and experimental variability.

Requires less RNA sample

Compared to the microarray alternative, Megaplex™ Primer Pools enable significant reduction in the amount of starting material, particularly when the preamplification step is incorporated into the workflow.

Streamlined workflow

Samples can be processed from cells to data in as little as 4 hours with minimal hands-on time and expertise requirements. All reagents required for completing your profiling experiment are validated together and available from Life Technologies. The highly streamlined workflow—coupled with the high specificity, superior sensitivity, and broad linear dynamic range of TaqMan® Assays—makes this product solution ideal for miRNA profiling, especially when compared to microarrays.

Helps elucidate the role of miRNAs in human disease

MicroRNAs have been demonstrated to play pivotal roles in the etiology of a variety of common human diseases, including cancer. TaqMan® miRNA analysis tools can help you develop biomarkers for cancer, better understand cancer stem cells, and further elucidate the role of miRNAs in the development and progression of cancer.

Product	Quantity	Cat. No.
Megaplex™ Primer Pools, Human Pool A v2.1	50 reactions	4401009
Megaplex™ Primer Pools, Human Pool B v2.0	50 reactions	4401010
Megaplex™ Primer Pools, Human Pool B v3.0	50 reactions	4444749
Megaplex™ Primer Pools, Human Pool Set (Pools A & B) v2.0	50 reactions	4401091
Megaplex™ Primer Pools, Human Pool Set (Pools A & B) v3.0	50 reactions	4444750
Megaplex™ Primer Pools, Rodent Pool A v2.0	50 reactions	4401090
Megaplex™ Primer Pools, Rodent Pool B v2.0	50 reactions	4401011
Megaplex™ Primer Pools, Rodent Pool B v3.0	50 reactions	4444752
Megaplex™ Primer Pools, Rodent Pool Set (Pools A & B) v2.0	50 reactions	4401012
Megaplex™ Primer Pools, Rodent Pool Set (Pools A & B) v3.0	50 reactions	4444766
Megaplex™ RT Primers, Human Pool A v2.1	50 reactions	4399966

Product	Quantity	Cat. No.
Megaplex™ RT Primers, Human Pool B v2.0	50 reactions	4399968
Megaplex™ RT Primers, Human Pool B v3.0	50 reactions	4444281
Megaplex™ RT Primers, Human Pool Set (Pools A & B) v2.0	50 reactions	4400928
Megaplex™ RT Primers, Human Pool Set (Pools A & B) v3.0	50 reactions	4444745
Megaplex™ RT Primers, Rodent Pool A v2.0	50 reactions	4399970
Megaplex™ RT Primers, Rodent Pool B v2.0	50 reactions	4399972
Megaplex™ RT Primers, Rodent Pool B v3.0	50 reactions	4444292
Megaplex™ RT Primers, Rodent Pool Set (Pools A & B) v2.0	50 reactions	4400926
Megaplex™ RT Primers, Rodent Pool Set (Pools A & B) v3.0	50 reactions	4444746
Megaplex™ PreAmp Primers, Human Pool A v2.1	50 reactions	4399233
Megaplex™ PreAmp Primers, Human Pool B v2.0	50 reactions	4399201
Megaplex™ PreAmp Primers, Human Pool B v3.0	50 reactions	4444303
Megaplex™ PreAmp Primers, Human Pool Set (Pools A & B) v2.0	50 reactions	4400927
Megaplex™ PreAmp Primers, Human Pool Set (Pools A & B) v3.0	50 reactions	4444748
Megaplex™ PreAmp Primers, Rodent Pool A v2.0	50 reactions	4399203
Megaplex™ PreAmp Primers, Rodent Pool B v2.0	50 reactions	4399237
Megaplex™ PreAmp Primers, Rodent Pool B v3.0	50 reactions	4444308
Megaplex™ PreAmp Primers, Rodent Pool Set (Pools A & B) v2.0	50 reactions	4400925
Megaplex™ PreAmp Primers, Rodent Pool Set (Pools A & B) v3.0	50 reactions	4444747

TaqMan® MicroRNA Reverse Transcription Kit

For optimal TaqMan® MicroRNA Assay performance

The TaqMan® MicroRNA Reverse Transcription (RT) Kit provides the necessary components for optimal performance of TaqMan® MicroRNA Assays. The components of this kit are used with the RT primer included with the TaqMan® MicroRNA Assay to convert miRNA to cDNA.

Product	Quantity	Cat. No.
TaqMan® MicroRNA Reverse Transcription Kit	200 reactions	4366596
	1,000 reactions	4366597

TaqMan® Array Human and Rodent MicroRNA Cards

TaqMan® MicroRNA Assays for profiling studies on 384-well microfluidic cards

- Highly specific—quantitate only the biologically active, mature miRNAs
- Convenient workflow reduces the number of reverse transcription reactions per sample
- Dynamic range of up to 7 logs detects high and low expressors in a single experiment
- Simplify sample handling and increase sample throughput
- An ideal miRNA profiling solution for human, mouse, and rat species

TaqMan® Array MicroRNA Cards provide all the advantages of TaqMan® MicroRNA Assays in a convenient preconfigured microfluidic card, minimizing the experimental variability and effort required to run 384 TaqMan® MicroRNA Assays in parallel.

Megaplex™ RT Primers are highly multiplexed RT primers designed to convert up to 381 miRNAs to cDNA prior to real-time analysis, and are required to run the TaqMan® Array MicroRNA Card. For greater sensitivity, or when working with limited sample, Megaplex™ PreAmp Primers provide an optional preamplification step prior to real-time analysis. TaqMan® Array MicroRNA Cards and Megaplex™ Primer Pools enable a comprehensive expression profile consistent with Sanger miRBase to be created in as little as four hours, providing the ideal miRNA profiling solution using the Applied Biosystems® ViiA™ 7 or 7900HT Fast Real-Time PCR Systems.

Simplified sample handling and increased sample throughput

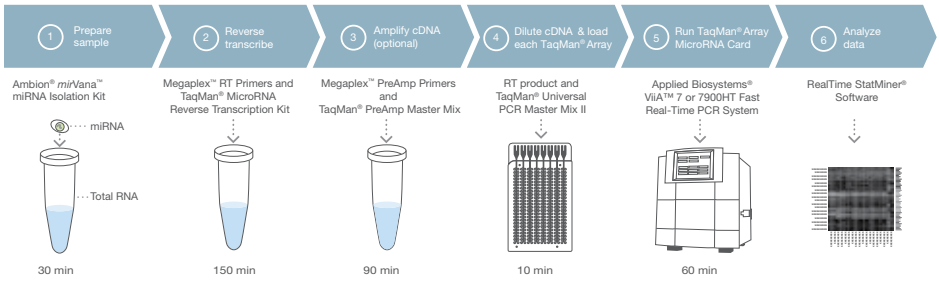
Following reverse transcription of miRNA targets using Megaplex™ RT Primers and optional preamplification using Megaplex™ PreAmp Primers, TaqMan® Universal PCR Master Mix II is simply combined with each reaction and pipetted into each of the eight sample loading ports in the TaqMan® Array MicroRNA Card, simplifying sample handling and increasing sample throughput.

Provides comprehensive coverage of Sanger miRBase

Megaplex™ RT Pools and PreAmp Primers are available for human and rodent species, and consist of preselected and matched content across a series of primer pools and TaqMan® Array Cards. The content is derived from miRBase for human and rodent species, providing close to full coverage of Sanger with the most up-to-date TaqMan® MicroRNA Assays.

Broad flexibility to meet your research needs

Choose from six workflows to best match your miRNA coverage requirements and your amount of starting material.



TaqMan® Array MicroRNA Card workflow. Get a complete human or rodent miRNA profile in as little as 4 hours, and in only 5.5 hours with the optional Megaplex™ PreAmp Primers and TaqMan® PreAmp Master Mix step.

Product	Quantity	Cat. No.
TaqMan® Array Human MicroRNA Card A v2.0	4 arrays	4398965
TaqMan® Array Human MicroRNA Card B v2.0	4 arrays	4398966
TaqMan® Array Human MicroRNA Card B v3.0	4 arrays	4444910
TaqMan® Array Human MicroRNA Card Set (A & B) v2.0	8 arrays	4400238
TaqMan® Array Human MicroRNA Card Set (A & B) v3.0	8 arrays	4444913
TaqMan® Array Rodent MicroRNA Card A v2.0	4 arrays	4398967
TaqMan® Array Rodent MicroRNA Card B v2.0	4 arrays	4398968
TaqMan® Array Rodent MicroRNA Card B v3.0	4 arrays	4444899
TaqMan® Array Rodent MicroRNA Card Set (A & B) v2.0	8 arrays	4400239
TaqMan® Array Rodent MicroRNA Card Set (A & B) v3.0	8 arrays	4444909

TaqMan® OpenArray® MicroRNA Panels

TaqMan® MicroRNA Assays for profiling studies on OpenArray® Plates

- Comprehensive coverage—generate human or rodent miRNA profiles with as little as 100 ng of total RNA
- High throughput—run up to 36 samples per 8-hour working day with minimal hands-on time and no need for robotics
- Cost-effectiveness—three samples per panel can be run, at a 33 nL reaction volume, resulting in significant reagent cost savings

TaqMan® OpenArray® MicroRNA Panels provide many of the advantages of TaqMan® MicroRNA Assays in a high-throughput plate and are ideal for medium to large miRNA profiling studies. TaqMan® OpenArray® MicroRNA Panels enable the running of hundreds of TaqMan® MicroRNA Assays in parallel per sample. This greatly simplifies the generation of a comprehensive miRNA profile for up to 36 samples in a single working day.

Provides comprehensive coverage of Sanger miRBase

Extensive coverage of the Sanger miRBase sequence repository is offered for human and rodent (mouse and rat) miRNA. The content is derived from miRBase, providing close to full coverage of Sanger with the most up-to-date TaqMan® MicroRNA Assays.

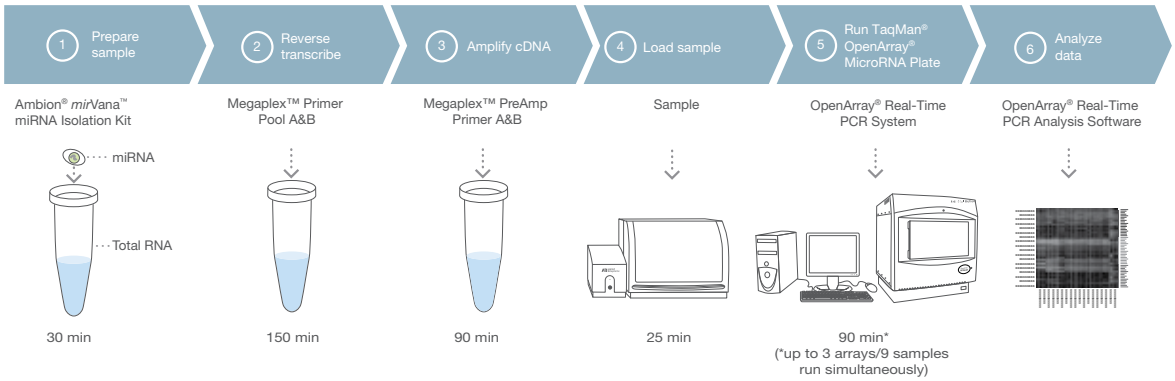
Streamlined workflow and high-throughput sample handling

The workflow is completed in 6 steps (see figure).

Profile miRNA with real-time PCR performance

A single panel generates 818 high-quality TaqMan® Assay data points using Megaplex™ Primer Pools and TaqMan® MicroRNA Assays:

- Demonstrated dynamic range four logs or more
- Minimally >90% of assays demonstrate standard deviation <0.5 using a typical tissue total RNA sample.
- Demonstrated high concordance with individual TaqMan® MicroRNA Assays



TaqMan® OpenArray® MicroRNA Panel workflow. Get a complete human or rodent miRNA profile for nine samples in about 7 hours.

Species	Megaplex™ Primer Pools	MicroRNA targets	Controls	
			+ev	-ev
Human	A	377	3	1
	B	377	3	1
	Total	754		
Rodent	A	375	5	1
	B	375	5	1
	Total	750		

Product	Quantity	Cat. No.
TaqMan® OpenArray® Human MicroRNA Panel	1 panel	4461104
TaqMan® OpenArray® Rodent MicroRNA Panel	1 panel	4461105

TaqMan® Protein Assays

Small-sample protein quantitation using real-time PCR

- Small sample size—uses 10–500-fold less sample input than traditional protein analysis methods with a typical input sample range of 5–500 cells or tissue lysate containing 1–100 ng total protein
- Sensitive detection—detect low expressing proteins that cannot be detected with other current methods
- Simple workflow—one-step lysis sample preparation, universal protocol, no washing steps
- Fast quantitation—cells to data in less than four hours
- Reproducible, well characterized assays—minimizes assay optimization, saving time and effort
- Quantitative—protein marker expression fold changes detected

TaqMan® Protein Assays help quantitate protein expression using gold standard TaqMan® 5´ nuclease chemistry with a workflow and sample quantity similar to TaqMan® Gene Expression and MicroRNA (miRNA) Assays. Because the protein expression results are obtained on the same analytical platform, these revolutionary assays enable direct correlation of mRNA and/or miRNA expression to protein expression.

TaqMan® Predesigned Protein Assays

The TaqMan® Protein Assays product family is comprised of a simple, one-step sample preparation kit for preparing cell lysates, six predesigned assays for detecting human protein markers, a supporting core reagents kit, and two control cell lysate kits. Four of the assays detect the human stem cell pluripotency markers hNANOG, hOCT3/4, hSOX2, and hLIN28. Two other assays detect the ubiquitously expressed proteins hICAM1 and hCSTB.

TaqMan® Protein Assays Open Kit for probe preparation

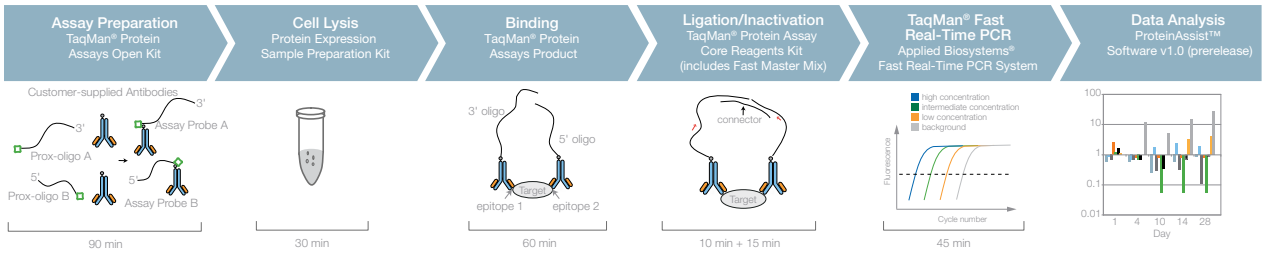
The TaqMan® Protein Assays Open Kit contains components to make TaqMan® Protein Assay probes. You supply the biotinylated antibodies and add them to streptavidin-oligonucleotides (prox-oligos) to make the two antibody-oligonucleotide assay probes utilized in binding.

Streamlined workflow

The sample preparation step consists of a gentle, one-step cell lysis using a buffered non-ionic detergent, with no further sample cleanup or purification required. The crude lysate samples can be directly mixed with the paired assay probes from any Protein Assay Kit for the target binding step. The entire experimental process, from sample prep to data output, can be completed in about 3.5 hours (see figure).

Data analysis

Data obtained from TaqMan® Protein Assays can be analyzed using our free ProteinAssist™ Software package.



TaqMan® Protein Assays workflow.

Product	Quantity	Cat. No.
TaqMan® Protein Assays Open Kit		
TaqMan® Protein Assays Open Kit	4,000 reactions	4453745
TaqMan® Protein Assays Oligo Probe Kit	4,000 reactions	4448549
TaqMan® Protein Assays Buffer Kit	4,000 reactions	4448571
TaqMan® Protein Assays (Predesigned)		
TaqMan® Protein Assay Kit (hCSTB)	100 reactions	4405465
TaqMan® Protein Assay Kit (hICAM1)	100 reactions	4405471

Product	Quantity	Cat. No.
TaqMan® Protein Assay Kit (hOCT3/4)	100 reactions	4405489
TaqMan® Protein Assay Kit (hNANOG)	100 reactions	4405483
TaqMan® Protein Assay Kit (hSOX2)	100 reactions	4405495
TaqMan® Protein Assay Kit (hLIN28)	100 reactions	4405477
Sample preparation kits		
Protein Quant Sample Lysis Kit	25 mL	4448536
Protein Expression Sample Prep Kit	1,000 reactions	4405443
Lysate control kits		
Protein Expression Lysate Control Kit (Raji)	100 reactions	4405448
Protein Expression Lysate Control Kit (NTERA2)	100 reactions	4405454
Core reagent kits		
TaqMan® Protein Assays Core Reagents Kit with Master Mix	100 reactions	4405501
	500 reactions	4448591
TaqMan® Protein Assays Core Reagents Base Kit	100 reactions	4405460
	500 reactions	4448592
TaqMan® Protein Assays Fast Master Mix	100 reactions	4400088
	500 reactions	4448616

DataAssist™ Software

For rapid and accurate quantitation of relative gene expression

DataAssist™ Software is an easy-to-use data analysis tool that utilizes the Comparative C_t($\Delta\Delta C_t$) method to rapidly and accurately quantitate relative gene expression across a large number of genes and samples.

The analysis workflow in the DataAssist™ software is simple and straightforward. You can import raw data from up to hundreds of plates or TaqMan® Arrays, and change analysis settings including selection of normalization controls and method (single or multiple control genes). It provides instant responses and fast calculations, allows normalization using multiple reference genes, and provides analysis results in content-rich tables and scalable graphic charts that are easily exported.

The license for using this software is limited to data generated from Applied Biosystems® real-time PCR instrumentation.

To download free DataAssist™ Software, go to www.appliedbiosystems.com/dataassist

ProteinAssist™ Software

For use with TaqMan® Protein Assays

- Multi-plate RQ analysis using a "Study" concept
- Friendly user interface for fast, easy plate setup to define samples names, assays, and input quantities
- Novel algorithms for calculating RQ using dilution curves of reference and test samples

ProteinAssist™ Software is a free software package used for analyzing data obtained from TaqMan® Protein Assays. It performs relative quantification calculations with TaqMan® Protein Assay C_t data. TaqMan® Protein Assays enable detection and relative quantification of protein targets using an adapted form of PLA™, a proximity ligation assay technology, in combination with real-time PCR.

The multi-plate RQ analysis enables:

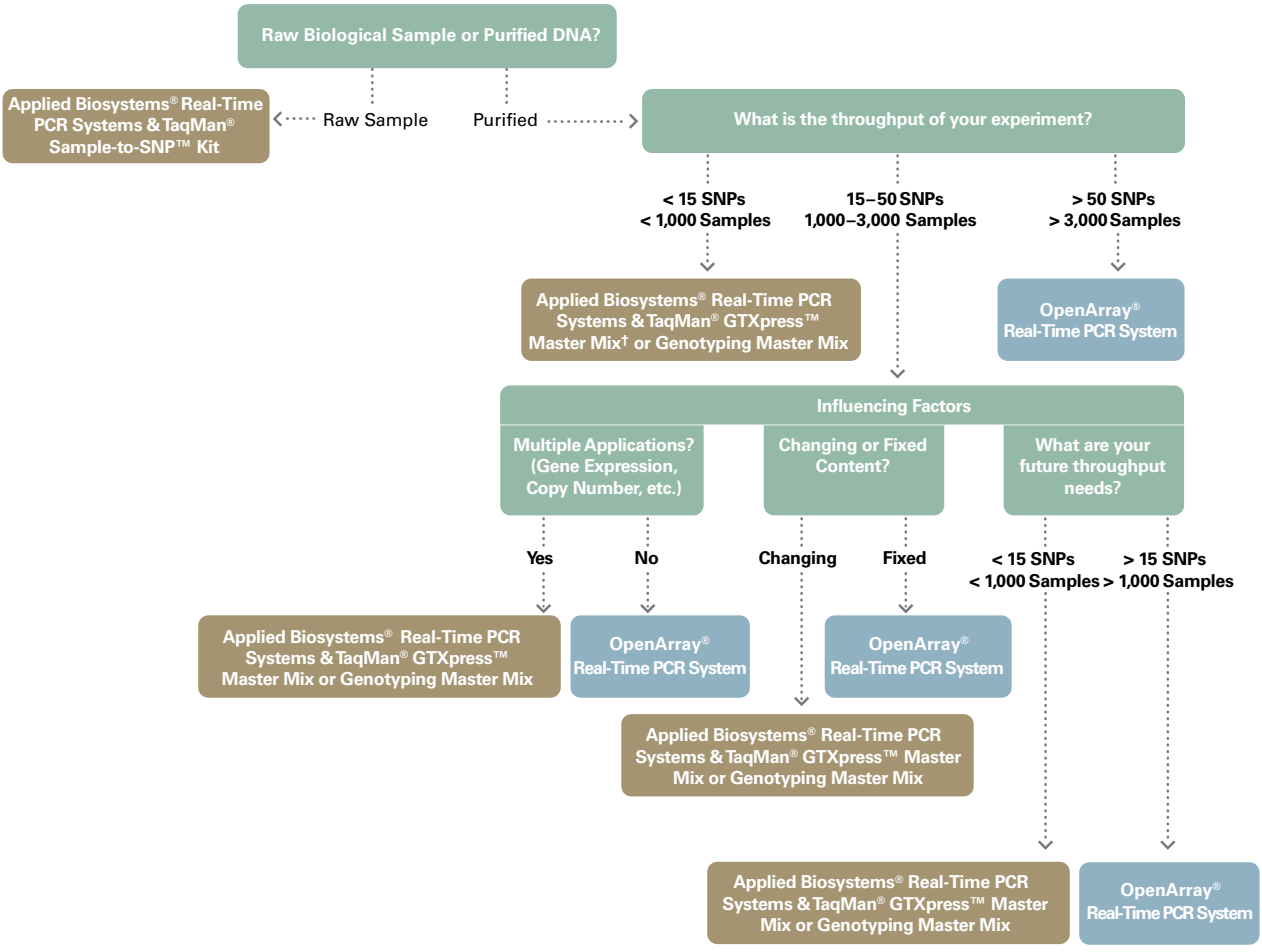
- Analysis of one to hundreds of 96- or 384-well plates as a single "Study"
- Analysis of multiple assays and/or samples per plate and "Study"
- Reporting of fold-change results in graph or heat map format

To download free ProteinAssist™ Software, go to www.appliedbiosystems.com/proteinassist.

TaqMan® SNP genotyping selection guide

The world's largest collection with the most flexible throughput formats for SNP analysis

With 4.5 million predesigned SNP assays available and a robust custom design pipeline, Life Technologies offers high-performance TaqMan® SNP Genotyping Assays together with best-in-class TaqMan® reagent solutions. And with the TaqMan® Sample-to-SNP™ Kit and OpenArray® Real-Time PCR System, we continue to provide SNP detection capabilities and high-confidence results with the ability to screen up to tens of thousands of samples at lower costs. To find out which technology or products fit your needs best, explore our selection guide below.



TaqMan® SNP Genotyping Assays

Highly flexible technology for SNP detection

- Large selection of SNP assays to advance your screening, association, candidate region, candidate gene, or fine-mapping studies
- Easy-to-use, single-tube format
- Robust assay design provides highly accurate, reproducible, and reliable results
- Easy assay ordering and a simple workflow enable quick results

TaqMan® SNP Genotyping Assays provide fast, accurate genotyping data and the simplest workflow available anywhere. A highly flexible technology for SNP detection, TaqMan® Assays are based on proven TaqMan® probe and primer chemistry. Designed for use on Applied Biosystems® real-time PCR systems, these assays produce high-confidence results in a wide variety of genotyping applications, including screening, candidate gene and candidate region studies, and fine-mapping studies.

TaqMan® Assays accelerate the pace of discovery by eliminating timely experimental design and optimization and are available as either off-the-shelf, predesigned, or custom assays. Content-rich marker-selection tools simplify study design and facilitate selection from a library of human assays, which includes over 4.5 million genome-wide human assays (of which 3.5 million are HapMap SNP-based assays, 160,000 validated assays, and over 70,000 coding region assays) and 10,000 mouse assays. Additionally, custom assays are also available for any genome. Simply submit your target SNP sequences through our secure, confidential online ordering system.

Type of assays	Species	Description
TaqMan® Predesigned SNP Genotyping Assays	Human	Select from our industry leading collection of over 4.5 million ready-to-order assays, including: • 160,000 validated assays with 20 million associated genotypes • ~70,000 coding SNP assays for the detection of SNPs within coding regions, including many putative functional SNPs • Over 4.5 million predesigned assays for high-density, genome-wide marker coverage
	Mouse	• More than 10,000 mouse assay designs available • Small-, medium-, and large-scale reaction sizes • Assays for use in applications such as speed congenics, genetic mapping, and genetic monitoring

Product	Fill volume	Number of 5-µL reactions	Reporter dye label	Cat. No.
TaqMan® Predesigned SNP Genotyping Assays, Human	187.5 µL, 40X	1,500	VIC®/FAM™	4351379
	625 µL, 40X	5,000	VIC®/FAM™	4351376
	750 µL, 80X	12,000	VIC®/FAM™	4351374
TaqMan® Predesigned SNP Genotyping Assays, Mouse	187.5 µL, 40X	1,500	VIC®/FAM™	4351384
	625 µL, 40X	5,000	VIC®/FAM™	4351382
	750 µL, 80X	12,000	VIC®/FAM™	4351380

We offer multiple ordering tools to support your SNP assay selection. For gene-based searches, go to http://snp.appliedbiosystems.com. Search features included are gene symbol, rs number, Entrez gene name, Entrez gene identifiers, and AB Assay ID.

Custom TaqMan® SNP Genotyping Assays

- Available for any organism
- Supports SNPs, multiple nucleotide polymorphisms (MNPs), and insertions/deletions (In/Dels)
- Secure and confidential ordering system
- Functional test on 20 unique DNA samples is performed for all human genotyping assays
- Simple TaqMan® Assay workflow reduces the chance of sample contamination and sample loss

Custom projects

Both custom plating and custom oligos/assays are available for this product line. Please speak with your sales representative for more information.

Product	Fill volume	# of 5-µL reactions	Reporter dye labels	Cat. No.
Custom TaqMan® SNP Genotyping Assays, Human	187.5 µL, 40X	1,500	VIC®/FAM™	4331349
	625 µL, 40X	5,000	VIC®/FAM™	4332072
	750 µL, 80X	12,000	VIC®/FAM™	4332073
Custom TaqMan® SNP Genotyping Assays, Non-human	187.5 µL, 40X	1,500	VIC®/FAM™	4332077
	625 µL, 40X	5,000	VIC®/FAM™	4332075
	750 µL, 80X	12,000	VIC®/FAM™	4332076

Order custom assays at www.appliedbiosystems.com/cadt.

TaqMan® Drug Metabolism Genotyping Assays

Individual assays for your drug metabolism studies

- Study single nucleotide polymorphisms (SNPs), multiple nucleotide polymorphisms (MNPs), and insertions/deletions (In/Dels) on one platform
- Work confidently—our bioinformatics mapping, design, and *in silico* QC means you will study the right SNP
- Get started immediately with ready-to-use assays optimized for Applied Biosystems® real-time PCR platforms

Life Technologies' collection of TaqMan® Drug Metabolism Genotyping Assays for basic and clinical researchers provides 2,700 unique assays to detect polymorphisms in 221 genes that code for various drug metabolism enzymes (DMEs) and drug transporters. These polymorphisms have been associated with certain diseases, such as cancer, and have been shown to significantly impact drug efficacy. Where possible all assays have been mapped to the common public allele nomenclature. All TaqMan® DME Genotyping Assays have proven performance across 180 unique DNA samples.

Product	# of 5-µL reactions	Reporter dye labels	Cat. No.
TaqMan® Drug Metabolism Genotyping Assays	750 reactions	VIC®/FAM™	4362691

To order, go to www.appliedbiosystems.com.

GeneAssist™ Copy Number Assay Workflow Builder

The GeneAssist™ Copy Number Assay Workflow Builder allows you to search for a predesigned TaqMan® Copy Number Assay by chromosome location, gene ID, variation ID, or other criteria. Predesigned assays can be added to the Shopping List to start the ordering process.

Design a Custom TaqMan® Copy Number Assay by entering your premasked sequence and selecting the desired target sites. The Custom Copy Number Assay tool will then design the assays. Assay designs may be compared to available reference assay sequences for compatibility in duplex PCR. Custom Plus TaqMan® Copy Number Assays can be designed using the tool for target regions of interest for which a predesigned assay is not available. Please note that the Custom Plus Assay design option must be used to check for genome specificity of assay designs. When the design job is complete, you will be notified to proceed with the ordering process.

For more information, go to http://www5.appliedbiosystems.com/tools/cnv/.

TaqMan® Copy Number Assays

- Simplest method available to study copy number variation
- Predesigned TaqMan® Copy Number Assays for human and mouse copy number analysis
- Assays available for common vector marker and reporter genes
- Custom Plus TaqMan® Copy Number Assays for user-defined human and mouse genomic targets
- Custom assays for other targets of interest
- Reference assays for unique human and mouse genomic sequences
- Measure distinct copy number changes quantitatively with high specificity and reproducibility

TaqMan® Copy Number Assays are designed for analysis of copy number variations (CNVs) and smaller regions in human and mouse genomes. CNV is an important polymorphism associated with diseases such as cancer, immune diseases, and neurological disorders as well as drug metabolism. TaqMan® Copy Number Assays combine TaqMan® Assay chemistry with Applied Biosystems® real-time PCR instruments to enable the generation of specific, reproducible, and easy-to-interpret copy number results. Ideally suited for validation studies and large sample screens, TaqMan® Copy Number Assays provide a flexible, targeted approach to copy number analysis.

Predesigned TaqMan® Copy Number Assays

The human predesigned assay collection includes over 1.6 million assays targeting gene exons and introns, extragenic regions, and CNV sequences from the Database of Genomic Variants (DGV). The mouse collection includes over 180,000 assays targeting gene exons. Assays to common vector marker and reporter genes are also available for transgenic studies.

Custom Plus TaqMan® Copy Number Assays

Custom Plus TaqMan® Copy Number Assays are the optimal solution for studying CNV in human and mouse genomic regions of interest for which a predesigned assay is not available. Custom Plus assays undergo thorough bioinformatic analysis and have the quality of predesigned TaqMan® Copy Number Assays. They can be generated for high-quality genomic targets of interest using the GeneAssist™ Copy Number Assay Tool.

Custom TaqMan® Copy Number Assays

Custom TaqMan® Copy Number Assays are an option for additional targets of interest. Users can submit their own premasked custom target sequences for assay design or primer/probe pair sequences for assay formulation.
Continued on next page.

Comparison of TaqMan® Copy Number Assay products.

Feature	Predesigned TaqMan® Copy Number Assays	Custom Plus TaqMan® Copy Number Assays	Custom TaqMan® Copy Number Assays
Designed with copy number–specific algorithm optimized for performance	•	•	•
Availability limited to human and mouse assays	•	•	
Contains TaqMan® FAM™ dye–labeled MGB probe and two unlabeled PCR primers	•	•	•
Targets undergo SNP and repetitive sequence masking	•	•	
Genome specificity check	•	•	
Reference assay compatibility check	•	• (optional)	• (optional)
Assay sequences provided			•
Assay context sequence and genome location provided	•	•	

Dependable method

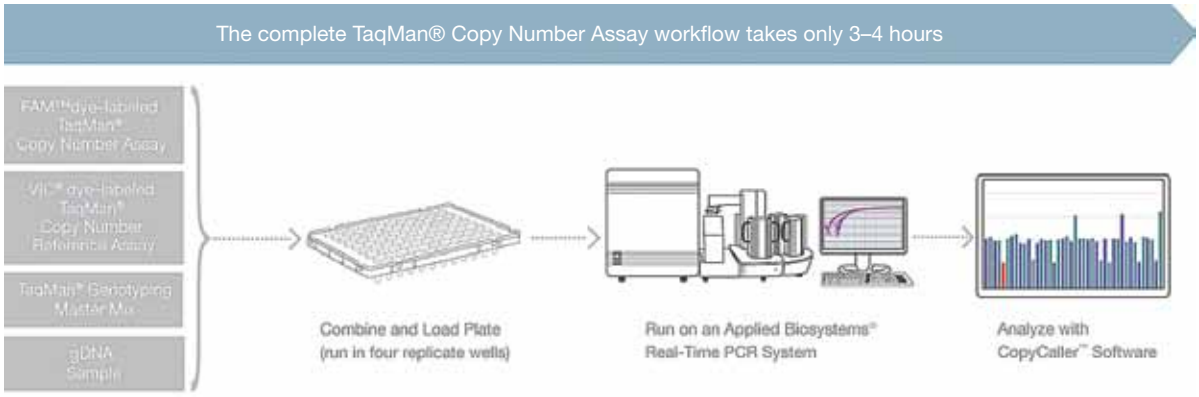
TaqMan® Copy Number Assays consist of a TaqMan® minor groove binding (MGB) probe labeled with FAM™ dye and unlabeled PCR primers. TaqMan® Copy Number Assays are run simultaneously with a choice of TaqMan® Copy Number Reference Assays (VIC® dye-labeled TAMRA™ probes) in a duplex real-time polymerase chain reaction. The copy number assay detects the target gene or genomic sequence of interest, and the reference assay detects a sequence that is known to be present in two copies in the diploid genome. Relative quantitation analysis is performed by CopyCaller™ Software, using either a known calibrator sample or no calibrator sample method.

Simplest workflow available

TaqMan® Copy Number Assays have the simplest workflow of all currently available CNV analysis methods (see figure). The test assay (FAM™ dye-labeled), the reference assay (VIC® dye-labeled), your sample DNA, and TaqMan® Master Mix are combined and then run on an Applied Biosystems® real-time PCR system using the standard TaqMan® Genotyping Assay protocol. On average, set-up to primary analysis takes only 3–4 hours (including an approximately 2 hr PCR run).

Custom projects

Both custom plating and custom oligos/assays are available for this product line. Please speak with your sales representative for more information.



TaqMan® Copy Number Assay workflow.

Product	Conc.	# of rxns in 96-well format	# of rxns in 384-well format	Cat. No.
TaqMan® Copy Number Assay, Small	20X	360	720	4400291
TaqMan® Copy Number Assay, Medium	20X	750	1,500	4400292
TaqMan® Copy Number Assay, Large	60X	2,900	5,800	4400293
Custom Plus TaqMan® Copy Number Assay, Small	20X	360	720	4442487
Custom Plus TaqMan® Copy Number Assay, Medium	20X	750	1,500	4442520
Custom Plus TaqMan® Copy Number Assay, Large	60X	2,900	5,800	4442488
Custom TaqMan® Copy Number Assay, Small	20X	360	720	4400294
Custom TaqMan® Co py Number Assay, Medium	20X	750	1,500	4400295
Custom TaqMan® Copy Number Assay, Large	60X	2,900	5,800	4400296
Human Reference Assays				
TaqMan® Copy Number Reference Assay, RNase P	20X (1 tube)	750	1,500	4403326
	20X (4 tubes)	3,000	6,000	4403328
TaqMan® Copy Number Reference Assay, TERT	20X (1 tube)	750	1,500	4403316
	20X (4 tubes)	3,000	6,000	4403315
Mouse Reference Assays				
TaqMan® Copy Number Reference Assay, Mouse, Tfrc	20X (1 tube)	750	1,500	4458366
	20X (4 tubes)	3,000	6,000	4458367
TaqMan® Copy Number Reference Assay, Mouse, Tert	20X (1 tube)	750	1,500	4458368
	20X (4 tubes)	3,000	6,000	4458369

TaqMan® OpenArray® Genotyping Plates

TaqMan® Genotyping Assays in a high-throughput format

- Simple workflow
- High throughput
- Flexible formats

The OpenArray® Real-Time PCR System is a dual-use platform that supports TaqMan® Genotyping Assays formatted on TaqMan® OpenArray® Genotyping Plates in addition to real-time PCR applications. The system utilizes nanoliter fluidic technology in conjunction with gold standard TaqMan® chemistry to enable mid-density, high-throughput workflows while reducing the cost per data point.

Simple workflow

TaqMan® OpenArray® Genotyping Plates contain your selected TaqMan® SNP Genotyping Assays preloaded and dried down in the through-holes you specify. Simply mix your sample and TaqMan® OpenArray® Genotyping Master Mix, load onto the genotyping plate, seal, cycle, and image. The OpenArray® Real-Time PCR System's simple workflow enables one person to validate or screen up to 256 SNPs across >1,500 samples in one day without the use of robotics. One person can therefore generate data for over 70,000 genotypes per day.

High throughput and high performance

In a single day, 24 TaqMan® OpenArray® Genotyping Plates providing 36,864 genotype reactions can be run, screening up to 1,728 DNA samples without the use of robotics. This system is ideal for screening and validating complex disease associations, marker-assisted breeding, or other studies requiring large sample sizes, and provides high performance and reproducibility (Table 1).

Flexible formats

Each TaqMan® OpenArray ® Genotyping Plate contains 3,072 through-holes arranged in 48 subarrays of 64 through-holes each. The OpenArray® AccuFill™ System can precisely load one, two, or three samples onto each subarray. This results in six plate format options (Table 2).
Continued on next page.

Table 1. Observed performance of the OpenArray® Real-Time PCR System using human cell line DNA and 896 TaqMan® SNP Genotyping Assays.

Feature	Performance*
Assay conversion rate from an Applied Biosystems® 7900HT System	>95%
Concordance with assays run on an Applied Biosystems® 7900HT System	99.7%
Call rate	>95%

* Individual performance is dependent on end user's sample integrity and purity.

Table 2. TaqMan® OpenArray® Genotyping Plate format options.

TaqMan® OpenArray Genotyping Plate format	Number of samples/plate	Minimum order	Total number of samples/ minimum order
16 assays	144 samples	10	1,440
32 assays	96 samples	10	960
64 assays	48 samples	20	960
128 assays	24 samples	40	960
192 assays	16 samples	60	960
256 assays	12 samples	80	960

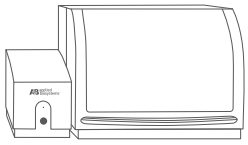
1. Select Assays or Panels & Order Products

Visit **www.appliedbiosystems.com** to select assays and OpenArray® Plate formats for custom plates, or to select from pre-designed OpenArray® Pathway Panels or TaqMan® OpenArray® Digital PCR Plates. Digital PCR plates are pre-treated to accept your primers and samples in your lab. All other OpenArray® Plates are delivered with assays dried down in the plate through-holes.

2. Add Sample & Master Mix

For gene expression and genotyping applications, mix cDNA or DNA samples with Master Mix in 384-Well Sample Plates. For digital PCR applications, mix primers, samples and Master Mix in 384-Well Sample Plates.

3. Load Samples

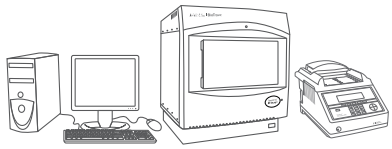


Load sample mixes onto an OpenArray® Plate with the AccuFill™ System.

4. Insert Into Case & Seal

Insert the OpenArray® Plate into the OpenArray® case filled with immersion fluid, and seal with glue.

5. Cycle, Image & Analyze Data



For genotyping applications, cycle in the Dual Flat Block GeneAmp® PCR System 9700 and transfer to the OpenArray® Real-Time PCR Instrument for imaging. For real-time and digital PCR applications, cycle and image on the OpenArray® Real-Time PCR Instrument and analyze data.

TaqMan® OpenArray® Genotyping Assay workflow.

TaqMan® OpenArray® Genotyping Kits	Quantity	Cat. No.
TaqMan® OpenArray® Genotyping Kit, Custom Format 16 Includes 10 TaqMan® OpenArray® Plates + 1 TaqMan® OpenArray® Accessories Kit	1 kit	4413546
TaqMan® OpenArray® Genotyping Kit, Custom Format 32 Includes 10 TaqMan® OpenArray® Plates + 1 TaqMan® OpenArray® Accessories Kit	1 kit	4413548
TaqMan® OpenArray® Genotyping Kit, Custom Format 64 Includes 10 TaqMan® OpenArray® Plates + 1 TaqMan® OpenArray® Accessories Kit	1 kit	4413550
TaqMan® OpenArray® Genotyping Kit, Custom Format 128 Includes 10 TaqMan® OpenArray® Plates + 1 TaqMan® OpenArray® Accessories Kit	1 kit	413551
TaqMan® OpenArray® Genotyping Kit, Custom Format 192 Includes 10 TaqMan® OpenArray® Plates + 1 TaqMan® OpenArray® Accessories Kit	1 kit	4413553
TaqMan® OpenArray® Genotyping Kit, Custom Format 256 Includes 10 TaqMan® OpenArray® Plates + 1 TaqMan® OpenArray® Accessories Kit	1 kit	4413554
TaqMan® OpenArray® Genotyping Training Plate	1 plate	4453991
TaqMan® OpenArray® Genotyping Master Mix	1 kit	4404846
OpenArray® 384-Well Sample Plates	10 plates	4406947
OpenArray® Practice Kit	1 kit	4453989
TaqMan® OpenArray® Genotyping Accessories Kit	1 kit	4404572

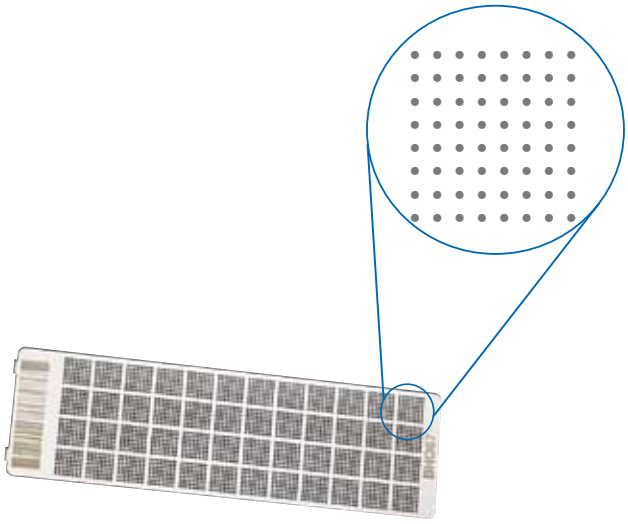
TaqMan® OpenArray® Digital PCR Kits

Absolute target quantification without reference standards or endogenous controls

- Accurate and sensitive—detect and count individual molecules to quantify viral load, gDNA, cDNA, plasmids, or next-generation sequencing libraries
- Fast—produce up to 144 digital PCR answers in a three-hour run
- Flexible—use your existing assays and capacity to test from one to 48 assay/sample dilutions per plate
- Wide dynamic range—by accommodating as few as 64 data points per replicate group, a dilution series can easily be loaded into the TaqMan® OpenArray® Digital PCR Plate, greatly expanding the range of sample concentrations that can be analyzed to produce a digital answer
- Intuitive and economical—software includes a Poisson calculator to design your individual digital PCR experiment, minimizing optimization time and sample usage

Digital PCR is a new approach to nucleic acid detection and quantification. It is a method of absolute quantification and rare allele detection that differs from conventional qPCR, in that it directly counts the number of target molecules rather than relying on reference standards or endogenous controls.

In digital PCR, each sample is partitioned into many individual real-time PCR reactions, some of which contain the target molecule (positive reactions), while the others do not (negative reactions). Following real-time PCR analysis, the ratio of positive reaction answers to negative reaction answers generates an absolute answer for the exact number of target molecules in the sample, without reference to standards or endogenous controls.



TaqMan® OpenArray® Digital PCR Plate design.

The OpenArray® Real-Time PCR System allows you to run over 9,000 digital PCRs simultaneously to generate more than 36,000 data points in a single day without the use of robotics. TaqMan® OpenArray® Digital PCR Kits eliminate the need to refer to endogenous controls or standards and deliver world-class digital PCR results in a flexible, easy-to-use format.

Simple workflow

TaqMan® OpenArray® Digital PCR Plates are delivered pretreated with hydrophilic and hydrophobic coatings to accept your own TaqMan® Assays. Simply mix assays along with your sample and TaqMan® OpenArray® Digital PCR Master Mix, load onto the TaqMan® OpenArray® Plate, seal, cycle, and image (see figure , facing page).

High throughput and high performance

The OpenArray® Real-Time PCR System provides the highest sample throughput. In a single day, 12 TaqMan® OpenArray® Digital PCR Plates, providing 36,864 digital PCR reactions, can be run without the use of robotics. The OpenArray® System is ideal for single-cell research, library quantification for next-generation sequencing, quantification of viral reference standards, and pharmaceutical applications of digital PCR, and provides high performance and reproducibility.

Flexible formats

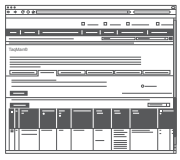
Each TaqMan® OpenArray® Digital PCR Plate contains 3,072 through-holes arranged in 48 subarrays of 64 through-holes each (see figure). The OpenArray® AccuFill™ System can precisely load one sample/assay dilution onto each subarray.

OpenArray® Digital PCR Software

OpenArray® Digital PCR Software allows researchers to analyze and manage data quickly and easily.

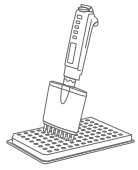
- Helps design your studies—includes Poisson calculator for use in designing digital PCR experiments
- Assists in OpenArray® workflow—automatically produces plate setup files from OpenArray® Plate bar code
- Intuitively presents results—produces heat-map plate views and graphical outputs that represent number of copies per sample dilution, while simple copy and paste functions enable easy export and presentation of results

1. Select Assays or Panels & Order Products



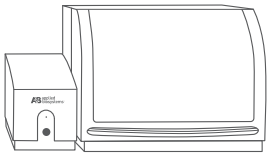
Visit **www.appliedbiosystems.com** to select assays and OpenArray® Plate formats for custom plates, or to select from pre-designed OpenArray® Pathway Panels or TaqMan® OpenArray® Digital PCR Plates. Digital PCR plates are pre-treated to accept your primers and samples in your lab. All other OpenArray® Plates are delivered with assays dried down in the plate through-holes.

2. Add Sample & Master Mix



For gene expression and genotyping applications, mix cDNA or DNA samples with Master Mix in 384-Well Sample Plates. For digital PCR applications, mix primers, samples and Master Mix in 384-Well Sample Plates.

3. Load Samples

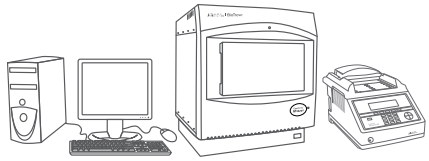


Load sample mixes onto an OpenArray® Plate with the AccuFill™ System.

4. Insert Into Case & Seal

Insert the OpenArray® Plate into the OpenArray® case filled with immersion fluid, and seal with glue.

5. Cycle, Image & Analyze Data



For genotyping applications, cycle in the Dual Flat Block GeneAmp® PCR System 9700 and transfer to the OpenArray® Real-Time PCR Instrument for imaging. For real-time and digital PCR applications, cycle and image on the OpenArray® Real-Time PCR Instrument and analyze data.

TaqMan® OpenArray® Digital PCR workflow.

Product	Quantity	Cat. No.
TaqMan® OpenArray® Digital PCR Kit	10 pack	4458070
Includes 10 OpenArray® Digital PCR Plates, 2X TaqMan® OpenArray® Digital PCR Master Mix (5 mL), OpenArray® Real-Time PCR Accessories Kit (enough to run 10 plates)		
TaqMan® OpenArray® Digital PCR Kit	3 pack	4460585
Includes 3 OpenArray® Digital PCR Plates, 2X TaqMan® OpenArray® Digital PCR Master Mix (1.5 mL)		
OpenArray® Real-Time PCR Accessories Kit	1 kit	4453975
Includes 10 OpenArray® Real-Time PCR Cases, 10 OpenArray® Immersion Fluids, 2 OpenArray® Case Sealing Glues		
OpenArray® Digital PCR Software 1.0	1 unit	4459279

TaqMan® Mutation Detection Assays

Detect rare mutations within genomic DNA samples

- High specificity—mutant allele detection is based on a modified allele-specific primer, while the wild type background is suppressed by the proprietary MGB blocker
- Superior sensitivity—assays can detect 0.1% mutant molecules in a background of wild type DNA
- Wide dynamic range and excellent PCR efficiency—assays demonstrate 7 logs of dynamic range and an average efficiency of 100 ± 10%
- Fast, simple workflow—like TaqMan® Assays, requires typically three hours from sample to results, with minimum hands-on time

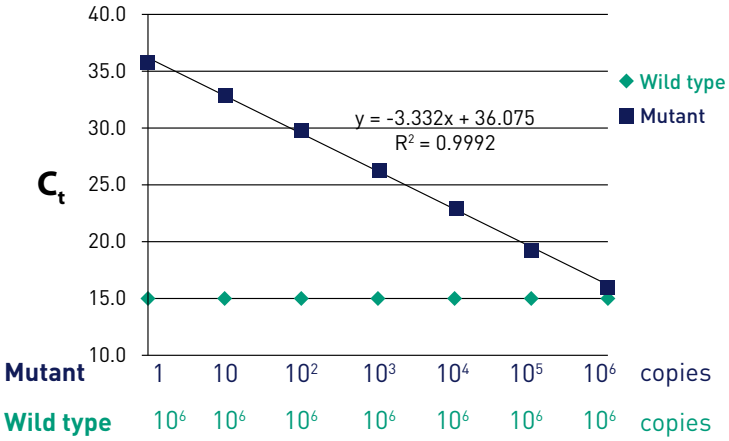
TaqMan® Mutation Detection Assays are designed to detect and measure specific DNA mutations against a background of wild type genomic DNA (gDNA). These assays are powered by competitive allele-specific TaqMan® PCR technology, known as castPCR™. castPCR™ technology is highly specific and can detect rare amounts of mutations within gDNA samples from human cancer cells.

Instrument and sample type compatibility

TaqMan® Mutation Detection Assays are compatible with the Applied Biosystems® ViiA™ 7, 7900HT, 7500, 7500 Fast, and StepOnePlus™ Real-Time PCR Systems. The assays can be used with gDNA extracted from FFPE samples, cell lines, and fresh frozen samples.

Mutation Detector™ Software allows users to detect and quantitate key mutations in *KRAS*, *BRAF*, and *EGFR* genes. This is a research use only software provided free of charge to customers who are using TaqMan® Mutation Detection Assays for mutation analysis.

Product	Cat. No.
TaqMan® Mutation Detection Assays	4465804
TaqMan® Mutation Detection Reference Assays	4465807
TaqMan® EGFR Exon 19 Deletions Assay	4465805



Mutation detection for an individual sample is determined by the ΔC_t between a reference assay and a mutation assay. The reference assay C_t represents the quantity of the target gene, and the mutation assay C_t represents the quantity of the mutation sequence.

TaqMan® Genotyper™ Software

For accurate genotyping analysis

- Accurate—new genotyping algorithm improves accuracy in automated genotype calling
- Efficient—provides multiplate data analysis for high-throughput workflows
- Flexible—allows for editing or removal of plates or individual data points
- Versatile—allows the use of controls and reference panels for accurate genotype calling
- Comprehensive—includes quality control tools for troubleshooting your experiments

TaqMan® Genotyper™ Software is a free SNP genotyping data analysis tool for use with TaqMan® SNP Genotyping Assays (Predesigned, Custom, & DME) in combination with 48-, 96-, and 384-well microtiter plates, and OpenArray® Genotyping Plates. The software showcases a state-of-the-art genotype-calling algorithm, an intuitive user interface, and enhanced multi-plate analysis features to meet the requirements of emerging markets and future research.

To download free TaqMan® Genotyper™ Software, go to www.appliedbiosystems.com/taqmangenotyper.

CopyCaller™ Software

For accurate copy number variation analysis

CopyCaller™ Software enables you to perform relative quantitation analysis of genomic DNA targets using the real-time PCR data from predesigned, Custom Plus, or Custom TaqMan® Copy Number Assay experiments. It can also be used for analysis of common vector marker and reporter gene targets. The software and associated copy number assays help detect and measure copy number variation of specific sequences in the human and mouse genomes.

To download CopyCaller™ Software, go to www6.appliedbiosystems.com/support/software/copycaller.

Custom TaqMan® Assay Design Tool

When you want to design a TaqMan® Assay

The Custom TaqMan® Assay Design Tool allows you to order Custom TaqMan® SNP Genotyping Assays, Custom Plus TaqMan® RNA Assays, and Custom TaqMan® Gene Expression Assays by entering sequences and then submitting for assay design. Upon notification of successful assay design, simply add the desired custom assays to your shopping basket.

To use the Custom TaqMan® Assay Design Tool, go to www.appliedbiosystems.com/cadt.

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nucleic acid purification and analysis

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Overview of nucleic acid purification

Purification kits and systems designed for your success

Life Technologies nucleic acid purification products are optimized to provide maximum yield, purity, and integrity from a variety of sample types in several different format options. The following chapter contains products and technologies to isolate RNA, miRNA, plasmid DNA, and gDNA. In addition, we review our instruments for automated purification. For complete details on the entire portfolio, go to www.invitrogen.com.

Overview of RNA purification

Life Technologies offers a complete portfolio of products to stabilize, isolate, and analyze RNA throughout your research, and is now brought to you under the RNA expert brand, Ambion®. The following table is an overview of this extensive portfolio, but for a complete listing, go to www.invitrogen.com/rnapreps.

Product	Quantity	Cat. No.	Recover from tissue	Recover from cells	Transcriptome analysis	Microarray	qRT-PCR	cDNA synthesis	Northern blotting	Automation-compatible	Viral RNA	RNA enrichment
Total RNA												
PureLink® RNA Mini Kit	50 preps	12183018A	•	•			•	•	•			
RNAqueous® Micro Kit	50 preps	AM1931	•	•			•		•			
PureLink® Total RNA Blood Kit	50 preps	K156001		•		•	•	•	•			
MagMAX™-96 Total RNA Isolation Kit	96 reactions	AM1830	•	•		•	•	•	•	•		
TRIzol® Reagent	200 mL	15596018	•	•		•	•	•	•			
TRIzol® Plus RNA Purification System	50 preps	12183555	•	•		•	•	•	•			
TRIzol® LS Reagent	200 mL	10296028		•		•	•	•	•		•	
Plant RNA Isolation Reagent	100 mL	12322012	•	•		•	•	•	•			
Dynabeads® mRNA DIRECT™ Kit	5 mL	610-11	•	•			•	•	•		•	
mRNA Catcher™ PLUS (1 plate)	96 preps	K157002	•	•	•	•	•	•	•	•		
Micro-FastTrack™ 2.0 mRNA Isolation Kit	20 preps	K152002	•	•	•	•	•	•	•			
FastTrack® 2.0 mRNA Isolation Kit	6 preps	K159302	•	•	•	•	•	•	•			
MicroRNA												
*mir*Vana™ miRNA Isolation Kit	40 purifications	AM1560	•	•	•						•	•
PureLink® miRNA Isolation Kit	25 preps	K157001	•	•	•	•	•	•	•			•

Product	Quantity	Cat. No.	Recover from tissue	Recover from cells	Transcriptome analysis	Microarray	qRT-PCR	cDNA synthesis	Northern blotting	Automation-compatible	Viral RNA	RNA enrichment
MicroRNA, cont.												
MagMAX™-96 for Microarrays Total RNA Isolation Kit	96 reactions	AM1839	•	•		•	•		•	•		
Transcriptome RNA												
RiboMinus™ Transcriptome Isolation Kit (Human/Mouse)	6 preps	K155002		•	•	•	•		•			•
Formalin-fixed, paraffin-embedded (FFPE)												
RecoverAll™ Total Nucleic Acid Isolation Kit	40 preps	AM1975	•		•		•	•	•			
MagMAX™ FFPE Total Nucleic Acid Isolation Kit	96 preps	4463365	•							•		
MagMAX™ FFPE DNA Isolation Kit	96 preps	4463578	•							•		
Laser capture microdissection (LCM)												
PureLink® RNA Micro Scale Kit	50 preps	12183016	•	•			•	•	•			
RNAqueous® Micro Kit	50 preps	AM1931	•	•			•		•			
qPCR direct from cells												
TaqMan® Gene Expression Cells-to-Ct™ Kit	100 reactions	AM1728		•			•					
Ambion® Single Cell-to-Ct™ Kit	50 reactions	4458237		•			•					
TaqMan® MicroRNA Cells-to-Ct™ Kit	100 reactions	4391848		•			•					
Power SYBR® Green Cells-to-Ct™ Kit	40 reactions/ 100 PCRs	4402953		•			•					
Stabilized Blood-to-Ct™ Nucleic Acid Preparation Kit for qPCR (compatible with either PAXgene® or Tempus™ Blood RNA Tubes)	50 reactions	4449079		•			•					

For more information about RNA isolation kits, go to www.invitrogen.com/rnapreps.
For information about instruments for nucleic acid purification, see pages 99–101.

PureLink® RNA Mini Kit

Simplified total RNA purification from cells and tissues

- Purification complete typically in less than 20 minutes
- High-quality, pure RNA ready for all downstream applications
- High yield from a single prep—up to 1,000 µg of purified RNA

The PureLink® RNA Mini Kit is designed for rapid purification of total RNA from a wide range of cells and tissue types including animal, plant, yeast, bacteria, and blood (Table1). Using a unique silica-based spin-column system, the PureLink® RNA Mini Kit yields high-quality RNA in significantly greater quantities than other commercial kits (Figure 1, Table 2), without the need for hazardous reagents such as phenol.

PureLink® technology combines guanidine isothiocyanate lysis with the speed, purity, and ease of use of silica-membrane purification. The safe and easy procedure can be completed typically in less than 20 minutes without the need for hazardous phenol/chloroform extraction or alcohol precipitation. High-quality purified RNA can be obtained from mini- to midi-prep amounts of starting material with minimal genomic DNA contamination. Purified RNA is eluted in RNase-free water and is ready to be used in downstream applications, such as RT-PCR, qRT-PCR, northern blotting, cDNA synthesis, and nuclease protection assays.

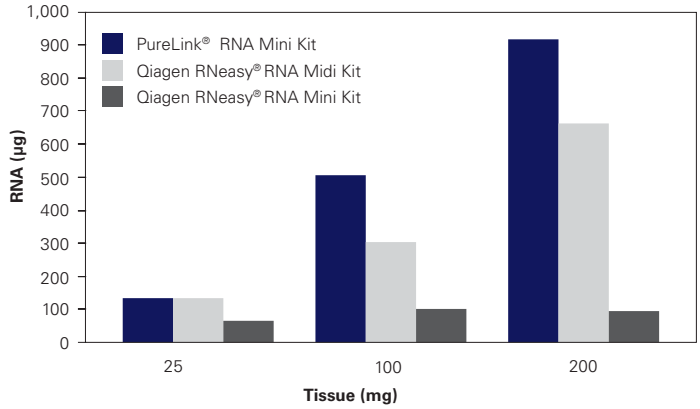


Figure 1. The PureLink® RNA Mini Kit produces high yields from a range of starting material. Total RNA was purified using 25 mg, 100 mg, and 200 mg of rat liver according to manufacturers' instructions. In each case, the PureLink® RNA Mini Kit provided equal or greater yields than the Qiagen RNeasy® kits.

Table 1. Sample sizes compatible with the PureLink® RNA Mini Kit.

Source	Amount
Animal and plant cells	Up to 10^8 cells
Animal tissue	Up to 200 mg
Plant tissue	Up to 250 mg
Whole blood	Up to 0.2 mL
Yeast cells	Up to 5 x 10^8 cells
Bacterial cells	Up to 10^9 cells

Table 2. The PureLink® RNA Mini Kit outperforms the Qiagen RNeasy® kits.

Specification	PureLink® RNA Mini Kit	RNeasy® Micro Kit	RNeasy® Mini Kit	RNeasy® Midi Kit
Number of preps/kit	50	50	50	50
Sample size	Up to 200 mg tissue, up to 5 x 10^7 cells	<5 mg tissue, <5 x 10^5 cells	0.5 to 30 mg tissue, up to 10^7 cells	20 to 250 mg tissue, 5 x 10^6 to 1 x 10^8 cells
Binding capacity	1,000 µg	45 µg	100 µg	1,000 µg
Protocol time	20 min	30–40 min	20 min	1 hr
Elution volume	30–100 µL	10–14 µL	30–100 µL	300–500 µL

Product	Quantity	Cat. No.
PureLink® RNA Mini Kit	50 preps	12183018A
	10 preps	12183020

For more information, go to www.invitrogen.com/totalrna.

MagMAX™ purification systems

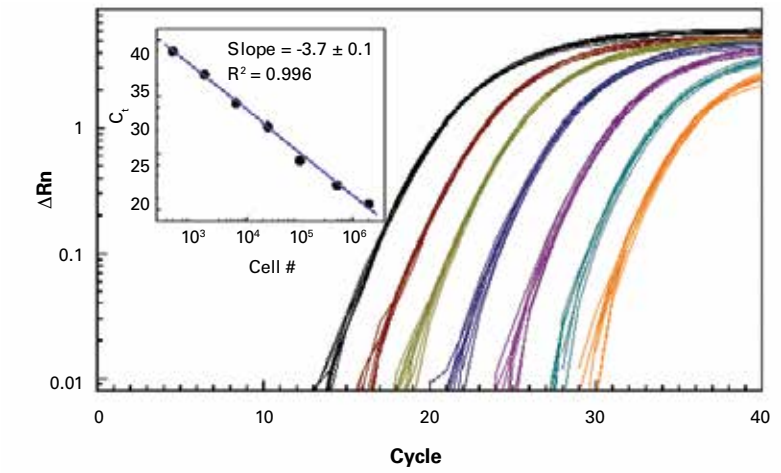
Maximum output, minimum time

- Flexible throughput options from 1 to 96 samples at a time
- Increased productivity from the significant time savings vs. column-based purification
- Consistent, high yields due to solution-phase binding and washing kinetics

Magnetic beads offer many benefits compared to other technologies for isolating RNA. Beads bind RNA more efficiently than glass-fiber filters, resulting in higher and more consistent RNA yields. Additionally, because filters are not used, there is no risk of filter clogging due to cellular particulates in samples. Since only a small volume of magnetic beads is needed for high-efficiency binding, the bound RNA can be eluted in just 20–50 µL of nuclease-free water, concentrating RNA from large, dilute samples.

The MagMAX™ kits accommodate diverse biological samples and a broad range of cell and tissue input amounts (see figure, and table on next page). The MagMAX™-96 kits are optimized for high-throughput isolation of RNA from 25 cells to 2 x 10^6 cells, as well as small plant and mammalian tissue samples (up to 10 mg). The protocol is fully amenable to automation, and detailed guidelines for general automation are included with the kits.

Continued on next page.



Recovery of total RNA from a wide range of cell numbers. RNA was isolated from 100 cells to 2 x 10^6 cells (K562) in 8 replicate wells using the MagMAX™-96 Total RNA Isolation Kit. Equivalent volumes of the recovered RNA (4% of eluted volume) were used for qRT-PCR (10 µL reaction) targeting human RNA Polymerase II mRNA. The CV among C_t values for the 8 replicate samples of each cell number input was less than 3%.

MagMAX™ Kits: for manual or automated nucleic acid isolation using magnetic particles.
Gene expression—cells and tissue

	MagMAX™-96 Total RNA Isolation Kit	MagMAX™-96 for Microarrays Total RNA Isolation Kit	MagMAX™ FFPE Total Nucleic Acid Isolation Kit
Cells	25–2 x 10^6	5 x 10^7	
Tissue	10 mg animal; 5 mg plant	Up to 100 mg	Up to two 10 µm sections per well
Features	• Ambion® TURBO DNase™ Enzyme Kit included for removal of gDNA • Compatible with plant tissues when used with Ambion® Plant RNA Isolation Acid • Plate format	• Includes organic extraction for stringent downstream applications, such as microarray analysis • Best for difficult samples or large volumes • Also isolates miRNAs with alternative protocol • Plate format	• Eliminates deparaffinization step • Plate format and automation-friendly • RNA and/or DNA typically in under 3 hr (automated) or 3.5 hr (manual)

Pathogen detection—viruses, bacteria, cell-free media, and blood

	MagMAX™-96 Viral RNA Isolation Kit	MagMAX™ Total Nucleic Acid Isolation Kit	MagMAX™-96 Blood RNA Isolation Kit	MagMAX™ for Stabilized Blood Tubes
Viral (RNA and DNA)	50–400 µL	175 µL liquid; 300 mg solid	50 µL	
Bacterial (RNA and DNA)		175 µL liquid; 300 mg solid		
Features	• Ideal when working with low viral concentrations • Also isolates DNA • Kits available in 50-reaction tubes or 96-well plate formats	• Lyses difficult pathogens by mechanical disruption with zirconia beads • Compatible with a variety of sample matrices • Also isolates gDNA • Tube format	• For detection of pathogens (virus and some bacteria) in blood & milk • Start with as little as 50 µL whole blood • Plate format	• Optimized for Tempus™ or PAXgene® Blood RNA Tubes only • Manual 96-well plate and tube format supported • Can be partially automated on the MagMAX™ Express-96 Deep Well Magnetic Particle Processor

Product	Quantity	Cat. No.
MagMAX™-96 Total RNA Isolation Kit	96 reactions	AM1830
MagMAX™ for Stabilized Blood Tubes (compatible with PAXgene® Blood RNA Tubes)	96 reactions	4451894
MagMAX™ for Stabilized Blood Tubes (compatible with Tempus™ Blood RNA Tubes)	96 reactions	4451893
MagMAX™ FFPE Total Nucleic Acid Isolation Kit	96 reactions	4463365
MagMAX™-96 for Microarrays Total RNA Isolation Kit	96 reactions	AM1839
MagMAX™-96 Viral RNA Isolation Kit	50 reactions	AM1939
	96 reactions	AM1836
MagMAX™-96 AI/ND Viral RNA Isolation Kit	4 x 96 reactions	AM1835
MagMAX™ Total Nucleic Acid Isolation Kit	100 reactions	AM1840
MagMAX™-96 Blood RNA Isolation Kit	96 reactions	AM1837

For more information, go to www.invitrogen.com/magmax.

TRIzol® reagents

Reliably purify RNA from multiple sample sources

- Superior lysis capability, even with difficult sample types
- Optimized formulations and protocols for tissues, cells, serum, virus, and bacteria

Referenced in thousands of citations, TRIzol® reagent is a trusted reagent for preparing high-quality, intact RNA from any starting material. TRIzol® reagents are ready-to-use monophasic solutions of phenol and guanidine isothiocyanate, suitable for isolating total RNA, DNA, and proteins. Isolation procedures are based upon improvements to the single-step RNA isolation method developed by Chomczynski and Sacchi [1,2] and are completed typically in less than one hour.

Table 1. The TRIzol® family of reagents.

Product	Sample type	Protocol
TRIzol® Reagent	Tissues and cells	1 mL of reagent for 100 mg tissue or 10^7 cells
TRIzol® LS	Fluid samples such as serum and virus preparations	0.75 mL of reagent for 5 x 10^6 cells
TRIzol® Max™ Bacterial RNA Isolation Kit	Bacterial samples	0.2 mL of Max™ Bacterial Enhancement Reagent and 1.0 mL TRIzol® for 10^8 bacterial cells

TRIzol® Plus RNA Purification System

Combining unparalleled lysis with the highest purity

- TRIzol® reagent sample lysis
- 1,000-µg column-binding capacity of the PureLink® RNA Mini Kits helps provide unmatched yields and purity

The TRIzol® Plus RNA Purification System couples the legendary lysis capabilities of TRIzol® Reagent with the ease and time-saving speed of the PureLink® RNA Mini Kit, resulting in high RNA integrity and yield. The purified total RNA is suitable for use in sensitive studies, including microarray analysis or qRT-PCR.

Table 2. TRIzol® Plus RNA Purification System specifications and advantages.

Specification	TRIzol® Plus RNA Purification System	Qiagen RNeasy® Lipid Tissue Mini Kit
Kit contents	100 mL TRIzol® reagent + 50 spin columns	50 mL QIAzol® reagent + 50 spin columns
Sample size	Up to 200 mg tissue or 10^8 cells	Up to 30 mg tissue or 10^7 cells
Sample types	All sample types, especially difficult samples	Fatty tissues (brain, adipose)
Binding capacity	1,000 µg	100 µg
Processing time	45 min	45 min
Elution volume (dependent on sample size)	Minimum 30 µL	30–100 µL

Product	Quantity	Cat. No.
TRIzol® Reagent (for tissues or cells)	100 mL	15596026
	200 mL	15596018
TRIzol® LS Reagent (for liquid samples, serum, and virus preparations)	100 mL	10296010
	200 mL	10296028
TRIzol® Max™ Bacterial RNA Isolation Kit	1 kit (100 mL)	16096020
	1 kit (200 mL)	16096040
TRIzol® Plus RNA Purification System	50 preps	12183555

TRIzol® and TRIzol® LS Reagents include the indicated amount of reagent. The TRIzol® Max™ Bacterial RNA Isolation Kit contains TRIzol® Reagent and Max Bacterial Enhancement Reagent. The TRIzol® Plus RNA Purification System includes a PureLink® Micro-to-Midi™ Total RNA Purification System and 100 mL of TRIzol® Reagent.

References

1.Chomczynski P and Sacchi N (1987) *Anal Biochem* 162:156.
2.Sewall A and McRae S (1998) *Focus*® 20:36.

For more information on TRIzol® products, go to www.invitrogen.com/trizol.

RiboMinus™ Transcriptome Isolation Kit (Human/Mouse)

Transcriptome enrichment without ribosomal RNA

- Whole transcriptome analysis with no rRNA contamination
- Complete collection of transcribed elements
- Ideal for enhanced microarray sensitivity

The RiboMinus™ Transcriptome Isolation Kit includes a novel purification system that enriches the spectrum of RNA transcripts by depleting large ribosomal RNA (rRNA) molecules in a total RNA sample. The kit uses 5´-biotin-labeled probes specific for large rRNA to remove >95% of the 18S and 28S rRNA molecules from human or mouse total RNA. The rRNA/probe complex is removed from samples with streptavidin-coated magnetic beads, which increases the representation of RNA transcript species, including non-poly(A)-tailed mRNA, fragmented mRNA, or other regulatory RNAs that cannot be purified via poly(A) selection methods.

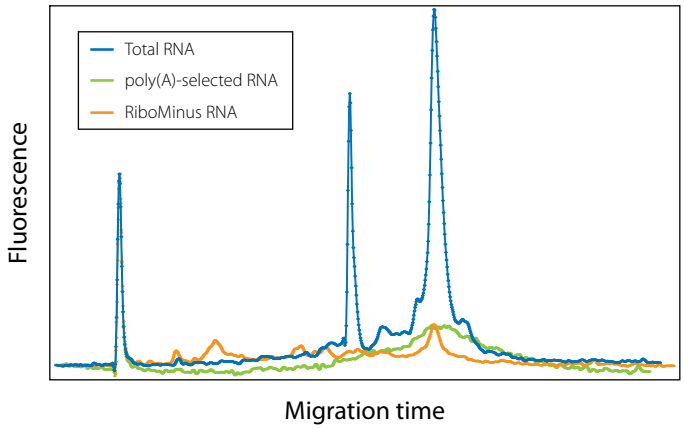


Figure 2. RiboMinus™ Transcriptome Isolation Kit efficiently removes >95% of the 18S and 28S rRNA. 2100 Bioanalyzer analysis of total RNA, RiboMinus™ RNA, and oligo(dT)-selected RNA with a RNA 600 Nano LabChip®. Total RNA was purified from HeLa cells using the PureLink® Micro-to-Midi™ Total RNA Purification System. Total RNA (10 µg) was enriched using the RiboMinus™ Transcriptome Isolation Kit.

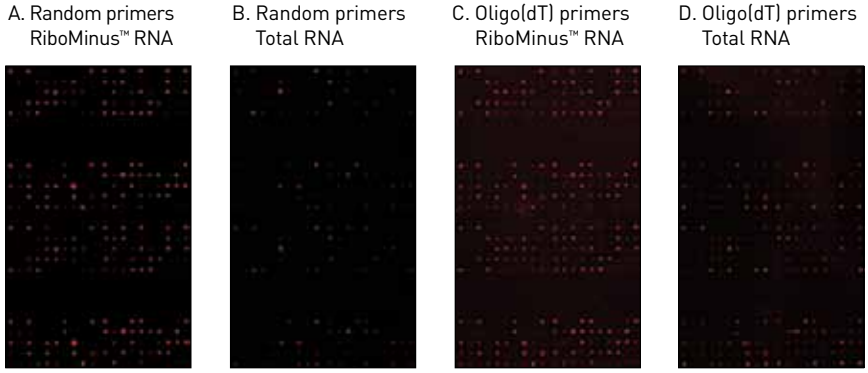


Figure 1. Improved microarray spotting intensity using the RiboMinus™ Kit. Arrays were labeled with 0.5 µg of RiboMinus™ or total RNA samples. The results with mRNA enriched with the RiboMinus™ Kit yielded better overall spot intensity and a higher signal-to-background ratio than using total RNA. **A**. Random priming of mRNA isolated using the RiboMinus™ Transcriptome Isolation Kit. **B**. Random priming of total RNA. **C**. Oligo(dT) priming of mRNA isolated using the RiboMinus™ Transcriptome Isolation Kit. **D**. Oligo(dT) priming of total RNA.

Product	Quantity	Cat. No.
RiboMinus™ Transcriptome Isolation Kit (Human/Mouse)	6 preps	K155002
RiboMinus™ Transcriptome Isolation Kit (Yeast)	12 preps	K155002
RiboMinus™ Transcriptome Isolation Kit (Bacteria)	12 preps	K155003

The RiboMinus™ Isolation Kits include RiboMinus™ Magnetic Beads; RiboMinus™ human/mouse, bacteria, or yeast probe; and hybridization buffer.

For more information on the RiboMinus™ Transcriptome Isolation Kit, go to www.invitrogen.com/rnapreps.

MicroRNA isolation kits

Efficient recovery of miRNA and other small RNAs

- Quantitative recovery of small RNA (<200 nt)
- Maintenance of representative amounts of small RNA (eliminating experimental bias)
- A choice of kits optimized for use with a variety of sample types

MicroRNAs (miRNAs) are small, highly conserved RNA molecules, which act as key regulators of development, cell proliferation, differentiation, and the cell cycle. Many standard glass-fiber filter methods were developed to recover mRNA and are not very efficient in recovering smaller RNAs. Life Technologies offers a variety of kits for the quantitative recovery of small RNAs from a variety of sample types (see tables, this page and page 81).

MicroRNA preparation kits: quantitative, recovery of small RNAs from a variety of sample types.

	TaqMan® MicroRNA Cells-to-C$_T$™ Kit	*mir*Vana™ miRNA Isolation Kit	*mir*Vana™ PARIS™ Kit	RecoverAll™ Total Nucleic Acid Isolation Kit for FFPE	MagMAX™-96 for Microarrays Total RNA Isolation Kit
Method	Lysate containing RNA	Purification			
Product—total RNA, including small RNA and miRNA	•	•	•	•	•
Product—DNA				•	
Product—protein			•		
Technology	Cells-to-C$_T$™ Kit lysis, with optional DNase treatment	Rapid acid-phenol/chloroform extraction and glass-fiber filter purification	Cell Disruption Buffer combined with acid-phenol/chloroform extraction and glass-fiber filter purification	Cell Disruption Buffer combined with acid-phenol/chloroform extraction and glass-fiber filter purification	Cell Disruption Buffer combined with acid-phenol/chloroform extraction and glass-fiber filter purification
Sample input amounts	10 to 100,000 cells	10^3–10^7 cells or 0.5–250 mg tissue	100–10^7 cultured cells or up to 100 mg tissue	Up to four 20 µm sections	Up to 5 x 10^6 cells or up to 100 mg tissue
Features	• qPCR results directly from cells without RNA purification • Simple protocols • Superior results when used with TaqMan® MicroRNA assays	• Ideal for miRNA profiling experiments and other gene expression applications • Fast, easy isolation of small RNA from cultured cells and most tissues (including tissues with high levels of ribonucleases)	• Simple, 30 minute procedure • Ideal for correlating mRNA, miRNA, and/or siRNA with protein levels • Optional small RNA enrichment procedure can increase sensitivity in downstream analyses	• Optimized for isolation of total nucleic acids, including miRNAs, from FFPE tissue • For real-time RT-PCR and PCR, mutation screening, and microarray analyses	• Highly consistent results from experiment to experiment • Walk away—integrate with established robotic platforms • Modified protocol to recover small RNAs

Product	Quantity	Cat. No.
TaqMan® MicroRNA Cells-to-C$_T$™ Kit	100 lysis reactions/500 PCRs	4391848
	400 lysis reactions/2000 PCRs	4391996
*mir*Vana™ miRNA Isolation Kit	Up to 40 purifications	AM1560
*mir*Vana™ PARIS™ Kit	Up to 40 purifications	AM1556
RecoverAll™ Total Nucleic Acid Isolation Kit for FFPE	40 purifications	AM1975
MagMAX™-96 for Microarrays Total RNA Isolation Kit	96 purifications	AM1839

For more information about microRNA isolation kits, go to www.invitrogen.com/rnapreps.

*mir*Vana™ miRNA Isolation Kits

- Efficient isolation of small RNA
- Enrich for small RNA <200 nt to increase sensitivity in downstream analyses
- Simple 30-minute procedure
- Ideal for miRNA, siRNA, shRNA, and snRNA analysis
- Compatible with virtually all cell and tissue types

The *mir*Vana™ miRNA Isolation Kit uses a rapid procedure to isolate small RNAs, such as microRNA (miRNA), small interfering RNA (siRNA), and small nuclear RNA (snRNA), from tissues and cells. The fast and efficient glass-fiber filter–based method isolates total RNA ranging in size from kilobases down to 10-mers. The kit also provides reagents and a procedure to enrich the population of RNAs that are 200 bases and smaller, which enhances the sensitivity of small RNA detection by solution hybridization, northern analysis, and other methods. In addition, the *mir*Vana™ PARIS™ Kit provides a unique and versatile procedure that permits quantitative recovery of native proteins and all RNA species from the same sample.

Features of the *mir*Vana™ kits.

Application	*mir*Vana™ miRNA Isolation Kit	*mir*Vana™ PARIS™ Kit	PARIS™ Kit
Isolation of high-quality total RNA	Yes	Yes	Yes
Quantitative recovery of protein	No	Yes	Yes
Quantitative recovery of small RNA	Yes	Yes	No
Small RNA enrichment procedure	Yes	Yes	No
Nuclear/cytoplasmic fractionation	No	No	Yes*
Compatible with RNA*later*® Solution	Yes	Yes	Yes
Compatible with cultured cells	Yes	Yes	Yes
Compatible with tissues	All	Most	Most

* The PARIS™ fractionation procedure is compatible with fresh cultured cells but not with RNA*later*®-treated cells, frozen cell pellets, or tissues.

Product	Quantity	Cat. No.
*mir*Vana™ miRNA Isolation Kit	Up to 40 purifications	AM1560
*mir*Vana™ PARIS™ Kit	Up to 40 purifications	AM1556

For more information about microRNA isolation kits, go to www.invitrogen.com/mirvana.

Cells-to-C$_T$™ kits

Consistency in qRT-PCR

- Validated solutions allow for consistent results, every time
- Simple and convenient kits that eliminate RNA purification
- Robust performance for accuracy, reproducibility, and sensitivity

Cells-to-C$_T$™ kits enable you to quickly and easily process 1–100,000 cultured cells into real-time PCR results. A breakthrough cell lysis and RNA stabilization technology eliminates the need for RNA purification. Because samples can be processed directly in culture plates (96- or 384-well), sample handling and the potential for sample loss or transfer error are minimized, resulting in higher sensitivity and reproducibility. Cells-to-C$_T$™ kits, however, don't stop at sample preparation. The lysis technology is integrated into a complete workflow that includes reverse transcription reagents (see page 141) and high-performance TaqMan® or SYBR® Green–based PCR master mixes (see page 18).

For more information, go to www.invitrogen.com/cellstoct.

Features of the Cells-to-C$_T$™ kits.

Product	Quantity	Cat. No.	Time-to-results	Cell input amount	Features
TaqMan® Gene Expression Cells-to-C$_T$™ Kit	• 40 lysis rxns/ 100 PCRs • 100 lysis rxns /500 PCRs • 400 lysis rxns/ 2,000 PCRs	4399002 AM1728 AM1729	Sample prep = 7 min RT = 1 hr Std real-time PCR = 1.5 hr	10 to 100,000 cells	Ideal for siRNA-induced gene knockdown screening, duplexing, and standard gene expression applications
Single Cell-to-C$_T$™ Kit	• 50 rxns • 100 rxns	4458237 4458236	7 min	1 to 10 cells	• Preoptimized workflow for real-time RT-PCR from single cells • Maximum sensitivity for single-cell analysis • Better performance and reproducibility than alternatives
TaqMan® Fast Cells-to-C$_T$™ Kit	• 100 lysis rxns/ 500 PCRs	4399003	Sample prep = 7 min RT = 1 hr Fast real-time PCR = 35 min	10 to 100,000 cells	Combine with Fast cycling chemistry and instruments for the fastest results available (40% less time than traditional RNA isolation and standard PCR)
TaqMan® PreAmp Cells-to-C$_T$™ Kit	• 40 lysis rxns/ 40 preamp rxns/ 100 PCRs	4387299	Sample prep = 7 min RT = 1 hr Preamp and std real-time PCR = 2.5 hr	10 to 100,000 cells	• Preamplification reagents (included) make it ideal for limited samples • 60-fold more PCR reactions from precious samples
TaqMan® MicroRNA Cells-to-C$_T$™ Kit	• 100 lysis rxns/ 500 PCRs • 400 lysis rxns/ 2,000 PCRs	4391848 4391996	Sample prep = 10 min RT = 1 hr Std real-time PCR = 1.5 hr	10 to 100,000 cells	Superior results when used with TaqMan® MicroRNA Assays
Power SYBR® Green Cells-to-C$_T$™ Kit	• 40 lysis rxns/ 100 PCRs • 100 lysis rxns/ 500 PCRs • 400 lysis rxns/ 2,000 PCRs	4402953 4402954 4402955	Sample prep = 7 min RT = 1 hr Std real-time PCR = 1.5 hr	10 to 100,000 cells	Validated with a broad range of primer sets
Fast SYBR® Green Cells-to-C$_T$™ Kit	• 100 lysis rxns/ 500 PCRs • 400 lysis rxns/ 2,000 PCRs	4402956 4402957	Sample prep = 7 min RT = 1 hr Fast real-time PCR = 35 min	10 to 100,000 cells	Combine with Fast cycling chemistry and instruments for the fastest results available (40% less time than traditional RNA isolation and standard PCR)
Stabilized Blood-to-C$_T$™ Kit, for PAXgene® or Tempus™ blood tubes	50 rxns	4449079	Sample prep ≤1 hr	500 µL stabilized blood sample	Detect total RNA (including miRNA) from stabilized blood samples (either PAXgene® or Tempus™ blood RNA tubes) without purification
Stabilized Blood-to-C$_T$™ Kit, for PAXgene® blood tubes	200 rxns	4449082			
Stabilized Blood-to-C$_T$™ Kit, for Tempus™ blood tubes	200 rxns	4449080			

Ambion® RNA Essentials

Specially designed to meet the stringent requirements of RNA research

- Nuclease inhibitors and decontaminants
- Nuclease-free water, buffers, tips, and tubes
- Specimen collection and RNA stabilization solutions
- Transcription products and RNA ladders (see pages 111–113)

RNA can be a difficult molecule to work with and is readily degraded by RNases that are found in a variety of environmental sources. To ensure success, steps must be taken to minimize nuclease contamination in RNA purification laboratories. Lab surfaces should be decontaminated with RNase*Zap*® solution, and samples should be protected by adding RNase inactivators such as RNA*later*® solution. Equally important, RNase-free pipette tips, microcentrifuge tubes, and conical tubes should be used at all steps. Life Technologies has a complete portfolio of Essentials to support your work with precious RNA.

Selected RNA Essentials from Life Technologies.

Function	Product	Description	Quantity	Cat. No.
Surface decontaminants	RNase*Zap*® solution	Glass and plastic surfaces	250 mL	AM9780
			6 x 250 mL	AM9782
	RNase *AWAY*® Reagent	Labware decontamination	250 mL	10328011
	Electro*Zap*™ solution	Electrodes	250 mL	AM9785
Ribonuclease inhibitors	RNaseOUT™	Recombinant ribonuclease inhibitor	5,000 units	10777019
	SUPERase•In™ RNase Inhibitor (20 U/µL)	Inhibits most common RNases, including RNase A, B, C, 1 and T1	2,500 units	AM2694
	ANTI-RNase inhibitor (15–30 U/µL)	Inhibits neutral pancreatic RNase A–type enzymes	2,500 units	AM2690
			10,000 units	AM2692
	RNA*secure*™ Reagent	Nonenzymatic inhibition of RNases in solutions	10 mL	AM7006
Nuclease-free water	Nuclease-Free Water (not DEPC-treated)		4 x 50 mL	AM9937
			100 mL	AM9938
	DEPC-Treated Water		5 x 100 mL	AM9916
Nuclease-free buffers and reagents	1 M Tris, pH 8.0		500 mL	AM9856
	TE, pH 8.0		500 mL	AM9849
	PBS 10X, pH 7.4		1 L	AM9625
	Salmon Sperm DNA (sheared, 10 mg/mL)		10 x 10 mg	AM9680
	Acid-Phenol:Chloroform, pH 4.5 (with IAA, 125:24:1)		100 mL	AM9720
	Glycogen (5 mg/mL)		5 x 1 mL	AM9510
	GlycoBlue™ (15 mg/mL)		5 x 0.3 mL	AM9516
	Proteinase K Solution (20 mg/mL)		5 x 1.25 mL	AM2548

Selected RNA Essentials from Life Technologies, cont.

Function	Product	Description	Quantity	Cat. No.
RNase-free plastics	RNase-Free Tips, 1,000-µL size	Certified, nuclease-free	Ten 100-ct racks	AM12660
	RNase-Free Tips, 200-µL size		Ten 8 x 12 racks	AM12650
	Barrier (Filter) Tips, 1,000-µL size		Ten 100-ct racks	AM12665
	Barrier (Filter) Tips, 200-µL size		Ten 8 x 12 racks	AM12655
	Barrier (Filter) Tips, 20-µL size		Ten 8 x 12 racks	AM12645
	Nonstick RNase-Free Microfuge Tubes		250 x 2.0 mL	AM12475
	Nonstick RNase-Free Microfuge Tubes		500 x 0.5 mL	AM12350
	50-mL Conical Tubes (racked)		200 x 50 mL	AM12501
Specimen collection stabilization solutions	RNA*later*® solution	Aqueous, nontoxic tissue storage reagent that rapidly permeates tissues to stabilize and protect cellular RNA	100 mL	AM7020
	RNA*later*®-ICE	For thawing frozen tissues to prevent RNA degradation during thawing	25 mL	AM7030
Transcription products	MEGAscript® T7 Kit		25 reactions	AM1333
	mMESSAGE mMACHINE® T7 Kit		25 reactions	AM1344M
	mMESSAGE mMACHINE® T7 ULTRA Kit		10 reactions	AM1345
	mMESSAGE mMACHINE® SP6 Kit		25 reactions	AM1340M5X

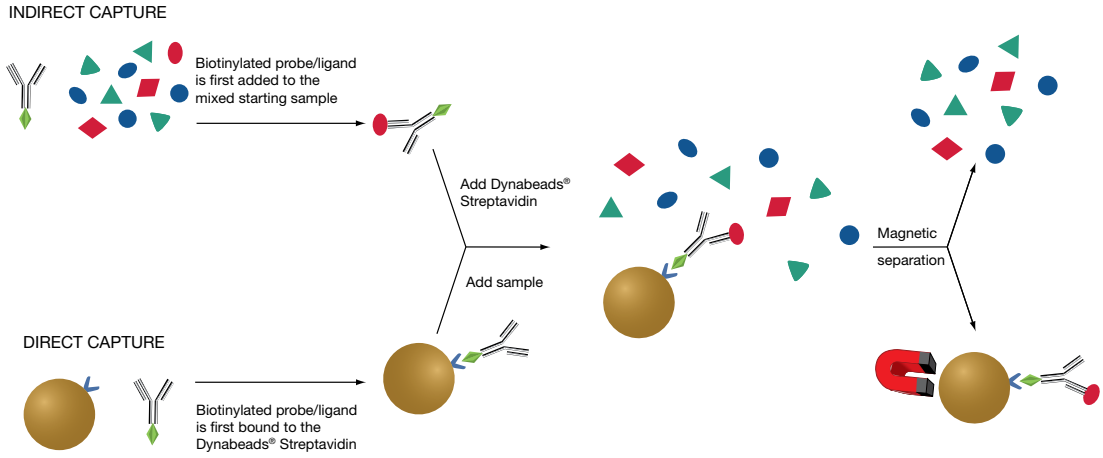
For more information or a complete list of RNA Essentials, go to www.invitrogen.com/ambion.

Sequence-specific RNA/DNA purification

Dynabeads® technology

- Products for mRNA, gDNA, and biotinylated molecules
- Protocol easily scaled to suit specific sample sizes

Invitrogen™ streptavidin-coupled Dynabeads® magnetic beads are a robust and versatile tool that can be used to target and capture specific RNA or DNA sequences and then pull them directly out of solution. The monosized, superparamagnetic Dynabeads® magnetic beads provide an efficient, solid-phase alternative to nitrocellulose and provide you with a superior level of product quality and data consistency. Excellent near–liquid phase reaction kinetics allow for extremely fast protocols. The inherent ease of magnetic handling means that downstream manipulations and buffer changes are as simple as concentrating the bead-bound target at the tube wall with a magnet and then discarding the supernatant. These beads are compatible with an extremely broad range of sample types, including most bodily fluids, crude lysates of plant, animal, and microbial origin, and purified total RNA or DNA. Since these Dynabeads® magnetic beads will only interact with specifically targeted RNA or DNA molecules, upstream purification of total RNA or DNA is usually unnecessary.



Direct and indirect approach for magnetic separation. In direct capture, the target-specific ligand is bound to the Dynabeads® magnetic beads and then added to the sample. For some applications, this enables reuse of the beads, thereby reducing costs. In indirect capture, the ligand is first allowed to bind to the target prior to addition of Dynabeads® magnetic beads. This can be beneficial when the concentration of the target is low, the specific affinity is weak, or the binding kinetics are slow.

Product	Quantity	Cat. No.
DynaMag™-15 magnet	each	123-01D
DynaMag™-50 magnet	each	123-02D
DynaMag™-Spin magnet	each	123-20D
DynaMag™-2 magnet	each	123-21D
Dynabeads® MyOne™ Streptavidin C1	2 mL	650-01
	10 mL	650-02
Dynabeads® M-270 Streptavidin	2 mL	653-05
	10 mL	653-06
Dynabeads® Streptavidin Trial Kit	4 x 1 mL	658-01D

For more information, go to www.invitrogen.com/dynabeads.

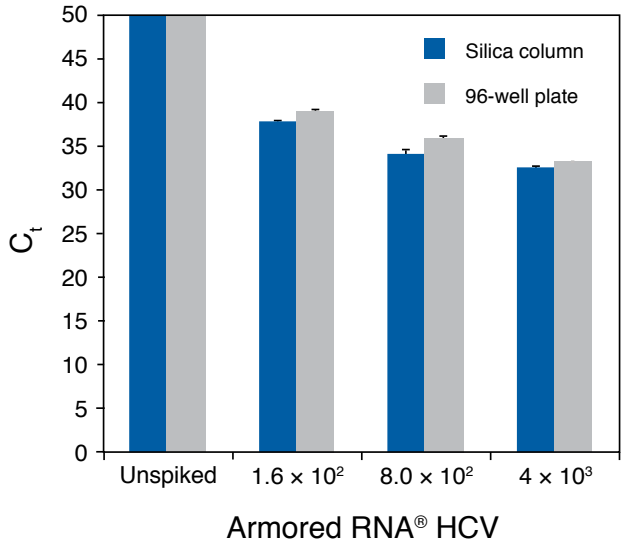
PureLink® viral RNA/DNA kits

Flexibility meets sensitivity for virus extraction research

- Highly concentrated viral nucleic acid yields for greater sensitivity
- Flexible system for purification of RNA and DNA
- Fast and easy purification with excellent reproducibility

The PureLink® Viral RNA/DNA Mini Kit is a nucleic acid purification system designed for fast and easy isolation of viral RNA or DNA from cell-free samples such as serum, plasma, and cerebrospinal fluid. Starting sample volumes can be as high as 500 µL and elution volumes as low as 10 µL, allowing for a 50-fold concentration of nucleic acids. This higher target concentration provides greater sensitivity and accuracy in downstream detection procedures such as qPCR and end-point analysis.

Specially formulated buffers enable the purification of RNA or DNA, allowing one system to be used for all your virus extraction samples. In addition, the kit provides sufficient quantities of lysis buffer to address large starting sample volumes. Equally high sensitivity is retained when switching from the spin column to the 96-well plate, making the PureLink® *Pro* 96 Viral RNA/DNA Purification Kit the most flexible and sensitive viral extraction kits available.



Comparison of sensitivity of the PureLink® *Pro* 96 Viral RNA/DNA Purification Kit with spin-column purification. Various concentrations of Armored RNA® HCV were prepared in human plasma, and 200 µL plasma samples were used for viral RNA purification using both the PureLink® *Pro* 96 Viral RNA/DNA Purification Kit and the PureLink® Viral RNA/DNA Mini Kit (spin-column purification system). After purification, Armored RNA® HCV-specific primers were used for qRT-PCR using the SuperScript® III Platinum® One-Step Quantitative RT-PCR Kit. Recovery of Armored RNA® was nearly identical between the PureLink® *Pro* 96 Viral RNA/DNA Purification Kit and the PureLink® Viral RNA/DNA Mini Kit.

Product	Quantity	Cat. No.
PureLink® Viral RNA/DNA Mini Kit	50 preps	12280050
PureLink® *Pro* 96 Viral RNA/DNA Purification Kit	4 x 96 preps	122800-96A

PureLink® Viral RNA/DNA Kits contain spin columns in collection tubes, collection and recovery tubes, lyophilized carrier RNA, proteinase K, lysis buffer, wash buffer, and RNase-free water.

For more information on the PureLink® viral RNA/DNA kits, go to www.invitrogen.com/viral.

Overview of plasmid DNA purification kits

Life Technologies offers an extensive portfolio of plasmid DNA purification products to prepare for downstream applications such as PCR, cloning, or transfection. The PureLink® and ChargeSwitch® families of products come in a variety of formats and scales, providing the plasmid purity and quantity that you need. The following table includes a selection of the mini, midi, and maxi kits Life Technologies provides for isolating plasmid DNA. To learn more about our automated nucleic acid purification systems, see pages 99–101. *Continued on next page.*

Overview of plasmid DNA purification kits, cont.

Selected mini, midi, and maxi kits for isolating plasmid DNA from Life Technologies.

Product	Quantity	Cat. No.	Grade	Format
Miniprep plasmid DNA purification kits				
PureLink® HiPure Plasmid Miniprep Kit	100 preps	K210003	Transfection	Gravity-flow column
PureLink® Quick Plasmid Miniprep Kit	250 preps	K210011	Molecular biology	Spin/vacuum column
PureLink® *Pro* Quick96 Plasmid Purification Kit	4 x 96 preps	K211004A	Molecular biology	Spin/vacuum column
PureLink® HQ Mini Plasmid DNA Purification Kit	100 preps	K210001	Transfection	Spin column
PureLink® 96 HQ Mini Plasmid DNA Purification Kit	4 x 96 preps	K210096	Transfection	Vacuum/centrifuge
ChargeSwitch® Pro Filter Plasmid Miniprep Kit	100 preps	CS31103	Transfection	Spin column
ChargeSwitch® Plasmid ER Mini Kit	50 preps	CS10100	Transfection	Magnetic beads
ChargeSwitch® NoSpin Plasmid Micro Kit	960 preps	CS1020110	Molecular biology	Magnetic beads
Midiprep plasmid DNA purification kits				
PureLink® HiPure Plasmid Midiprep Kit	50 preps	K210005	Transfection	Gravity-flow column
PureLink® HiPure Plasmid Filter Midiprep Kit	25 preps	K210014	Transfection	Gravity-flow column
ChargeSwitch® Pro Filter Plasmid Midi Kit	25 preps	CS31104	Transfection	Magnetic beads
Large-scale plasmid (>400 µg) DNA purification kits				
PureLink® HiPure Plasmid Maxiprep Kit	25 preps	K210007	Transfection	Gravity-flow column
PureLink® HiPure Plasmid Filter Maxiprep Kit	25 preps	K210017	Transfection	Gravity-flow column
PureLink® HiPure FP (Filter and Precipitator) Maxiprep Kit	25 preps	K210027	Transfection	Gravity-flow column
PureLink® HiPure Plasmid Megaprep Kit	4 preps	K210008	Transfection	Vacuum-assisted cartridge
PureLink® HiPure Plasmid Gigaprep Kit	2 preps	K210009	Transfection	Vacuum-assisted cartridge
ChargeSwitch® Pro Filter Plasmid Maxi Kit	10 preps	CS31106	Transfection	Magnetic beads
BenchPro® 2100 Plasmid Purification System	1 instrument	MC1001	Transfection	Positive air–driven fluidics card that contains a novel anion-exchange membrane
BenchPro® 2100 Plasmid Purification Card and Reagent Tray Kit	4 preps	MC2001		
Accessories				
EveryPrep™ Universal Vacuum Manifold	1 manifold	K211101		

Advantage	Protocol time	Culture volume	Yield
UltraPure™ purification resulting in extremely low endotoxin levels. Also integrates column-based workflow with double nucleic acid binding for high yields.	90–120 min	1–3 mL	Up to 30 µg
Quick, easy, cost-effective protocol	<25 min	1–5 mL	Up to 40 µg
High-throughput processing flexibility with protocols for vacuum/centrifuge	45 min	1.5 mL	Up to 20 µg
High yields, high purity	<30 min	1–5 mL	Up to 60 µg
High-throughput processing. Transfection-grade plasmid purity.	40 min	1–3 mL	Up to 10 µg
High-quality plasmid DNA using ChargeSwitch® in a column format	<20 min	1–5 mL	Up to 25 µg
High yields, high purity	~10 min	1–5 mL	Up to 20 µg
Centrifuge-free, high-throughput protocol	15 min	0.5–1 mL	Up to 5 µg
UltraPure™ purification resulting in extremely low endotoxin levels. Also integrates column-based workflow with double nucleic acid binding for high yields.	120 min	25–100 mL	100 to 350 µg
UltraPure™ purification resulting in extremely low endotoxin levels. Also integrates column-based workflow with double nucleic acid binding for high yields.	90 min	25–100 mL	100 to 350 µg
Fastest maxi plasmid prep on the market. Transfection-grade purification.	30 min	25 mL	200 µg
UltraPure™ Purification resulting in extremely low endotoxin levels. Also integrates column-based workflow with double nucleic acid binding for high yields	120 min	100–500 mL	500–850 µg
UltraPure™ purification resulting in extremely low endotoxin levels. Also integrates column-based workflow with double nucleic acid binding for high yields.	90 min	25–500 mL	500–850 µg
UltraPure™ purification resulting in extremely low endotoxin levels. Also integrates column-based workflow with double nucleic acid binding for high yields.	60 min	25–500 mL	500–850 µg
Fastest large-scale plasmid prep in the market. UltraPure™ purification resulting in extremely low endotoxin levels. Also integrates column-based workflow with double nucleic acid binding for high yields.	2–3 hr	500 mL–2.5 L	1.5–2.5 mg
UltraPure™ purification resulting in extremely low endotoxin levels. Also integrates column-based workflow with double nucleic acid binding for high yields.	2–3 hr	2.5–5 L	7.5–10 mg
Fastest maxi plasmid prep on the market. Transfection-grade purification.	30 min	100 mL	Up to 800 µg
Fully automated system that requires less than 5 min to set up	>5 min hands-on; 80 min automated run time	125 mL	Up to 1 mg

For a complete listing of products and kit sizes, go to www.invitrogen.com/plasmidpurification.

PureLink® HiPure plasmid purification kits

Pure, transfection-grade plasmid DNA

- High plasmid yields
- Low endotoxin levels

The PureLink® HiPure plasmid purification kits are designed to isolate plasmid DNA of the highest purity at the scale you need. In as little as one hour, DNA is pure enough for transfecting without the need for additional steps to remove contaminants. Phenol, chloroform, and cesium chloride are eliminated, minimizing exposure to and disposal of hazardous materials.

Characteristics of PureLink® plasmid kits.

Product	Quantity	Cat. No.	Protocol speed	Lysate clearing method	DNA binding step	Precipitation method
Miniprep plasmid: up to 30 µg from 1–3 mL of culture						
PureLink® HiPure Plasmid Miniprep Kit	25 preps	K210002	Standard	Centrifugation	Gravity-flow column	Centrifugation
	100 preps	K210003				
Midiprep plasmid: 100–350 µg from 25–100 mL of culture						
PureLink® HiPure Plasmid Midiprep Kit	25 preps	K210004	Standard	Centrifugation	Gravity-flow column	Centrifugation
	50 preps	K210005				
PureLink® HiPure Plasmid Filter Midiprep Kit	25 preps	K210014	Fast	Lysate filter (no centrifugation)	Gravity-flow column	Centrifugation
Maxiprep plasmid: 500–850 µg from 100–500 mL of culture						
PureLink® HiPure Plasmid Maxiprep Kit	10 preps	K210006	Standard	Centrifugation	Gravity-flow column	Centrifugation
	25 preps	K210007				
PureLink® HiPureFilter Plasmid Maxiprep Kit	10 preps	K210016	Fast	Lysate filter (no centrifugation)	Gravity-flow column	Centrifugation
	25 preps	K210017				
PureLink® HiPure Plasmid FP Maxiprep Kit	10 preps	K210026	Ultra-fast	Lysate filter (no centrifugation)	Gravity-flow column	HiPure precipitator
	25 preps	K210027				
Megaprep plasmid: 1.5–2.5 mg from 500 mL–2.5 L of culture						
PureLink® HiPure Plasmid Megaprep Kit	4 preps	K210008	Ultra-fast	Vacuum-assisted	Vacuum-assisted column	Centrifugation
Gigaprep plasmid: 7.5–10 mg from 2.5–5 L of culture						
PureLink® HiPure Plasmid Gigaprep Kit	2 preps	K210009	Ultrafast	Vacuum-assisted	Vacuum-assisted column	Centrifugation

For more information on PureLink® HiPure Plasmid Purification Kits, go to www.invitrogen.com/plasmidpurification.

PureLink® Quick Plasmid Miniprep Kit

Prepare up to 40 µg of sequencing-grade plasmid DNA using traditional silica

- Familiar silica-membrane, spin-column format
- Ready-to-use plasmid DNA typically in <30 minutes
- Flexible formats work with vacuum and centrifugation platforms

For use in standard molecular biology applications, the PureLink® Quick Plasmid Miniprep Kits offer rapid isolation of plasmid DNA from 1–5 mL cultures using the PureLink® spin columns. These columns use a unique silica membrane to achieve high yields (up to 40 µg) following a simple bind-wash-elute procedure. The plasmid DNA is eluted in water or TE, free of RNA, proteins, and other contaminants.

Product	Quantity	Cat. No.
PureLink® Quick Plasmid Miniprep Kit	50 preps	K210010
	250 preps	K210011

PureLink® Quick Plasmid DNA Miniprep Kits include resuspension buffer, RNase A, lysis buffer, precipitation buffer, wash buffer, TE buffer, spin columns, and wash and recovery tubes.

For more information on PureLink® Quick Plasmid Miniprep Kits, go to www.invitrogen.com/plasmidpurification.

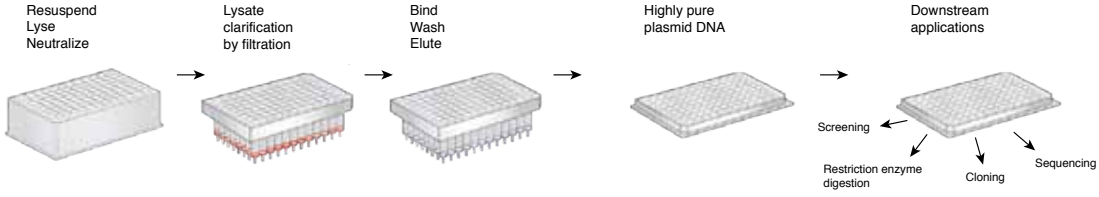
PureLink® *Pro* Quick96 Plasmid Purification Kit

Purify up to 20 µg of molecular biology–grade plasmid DNA in a 96-well format

- Allows for consistently high yields of high-purity plasmid DNA
- Experience ease of use and better performance due to improved plate design and protocol
- Increase processing flexibility with protocols for centrifuge, vacuum manifold, or automation

The PureLink® *Pro* Quick96 Plasmid Purification Kit combines advanced silica-plate extraction chemistry with an optimized 96-well plate design for manual or automated processing of plasmid DNA from *E. coli*. The PureLink® *Pro* Quick96 Plasmid Purification Kit uses two 96-well plates, a filtration plate (clear O-ring), and a binding plate (red O-ring), to purify plasmid DNA. Bacterial cells are subjected to alkaline lysis and are pelleted by centrifugation. The bacterial lysate is then applied to the Quick96 Filtration Plate for clearing, and collected in the Quick96 Binding Plate, where the plasmid DNA binds to the silica membrane. After washing to remove any contaminants, purified DNA is eluted and ready for use in downstream applications. The entire procedure can typically be completed in 45 minutes.

The PureLink® *Pro* Quick96 Plasmid Purification Kit allows rapid and reproducible purification of up to 20 µg of plasmid DNA from *E. coli* strains grown in up to 5 mL LB, 3 mL 2X YT, or 3 mL TB. In an automated platform using 1.5 mL of *E. coli* culture grown overnight in a square-well block, typical yields are 5–15 µg plasmid DNA, with an average of 9.4 µg. The resulting plasmid DNA is supercoiled with no detectable genomic DNA or RNA. The PureLink® *Pro* Quick96 Plasmid Purification Kit provides higher yields of high-quality plasmid DNA for use in common downstream applications such as automated sequencing, PCR, restriction enzyme digestion, and cloning.



From *E. coli* culture to purified plasmid DNA in less than 45 minutes.

Product	Quantity	Cat. No.
PureLink® *Pro* Quick96 Plasmid Purification Kit	4 x 96 reactions	K211004A

PureLink® *Pro* Quick96 Plasmid Purification Kit includes resuspension buffer, lysis buffer, neutralization buffer, RNase A, wash buffers, elution buffer, square-well blocks, filtration plates, binding plates, wash plates, and elution plates.

For more information on The PureLink® *Pro* Quick96 Plasmid Kit, go to www.invitrogen.com/plasmidpurification.

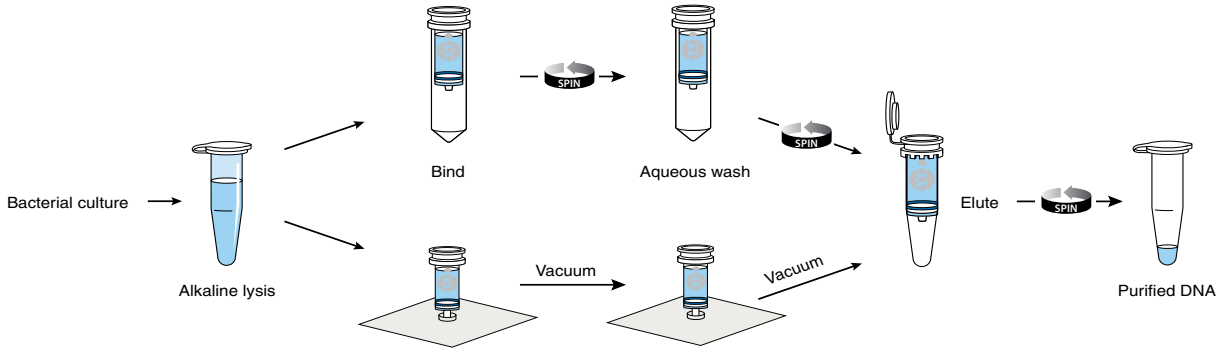
ChargeSwitch® plasmid kits

Quick, easy methods for high-quality plasmid DNA

- High-yield and high-purity plasmid
- Fast and easy prep, facilitated with simultaneous lysate clearing and binding step
- 100% ethanol-free, guanidinium-free, organics-free, aqueous protocol to enhance downstream application success

The ChargeSwitch® kits generate high-quality plasmid DNA without the use of harsh or potentially interfering reagents, for better success in downstream applications. Purification is by a simple ion-exchange mechanism: at low pH values (<6.5) the ChargeSwitch® surface ligand is positively charged and binds plasmid DNA, and at higher pH (>8.5) the charge is neutralized and the bound nucleic acid is eluted. The ChargeSwitch® protocols are fast and easy and can be adapted for use with vacuum manifolds.

- The ChargeSwitch® Pro Filter Plasmid Mini Kit is ideal for purifying plasmid DNA from 1–5 mL overnight bacterial cultures, yielding up to 25 µg of plasmid DNA, typically in <30 min
- The ChargeSwitch® Pro Filter Plasmid Midi Kit is ideal for purifying plasmid DNA from 25–100 mL overnight bacterial cultures, yielding up to 350 µg of plasmid DNA, typically in 30 min
- The ChargeSwitch® Pro Filter Plasmid Maxi Kit is ideal for purifying plasmid DNA from 100 mL overnight bacterial cultures, yielding up to 800 µg of plasmid DNA, typically in <30 min



The easy-to-follow ChargeSwitch®-Pro Plasmid Miniprep Kit process.

Product	Quantity	Cat. No.
ChargeSwitch® Pro Filter Plasmid Mini Kit	100 preps	CS31103
ChargeSwitch® Pro Filter Plasmid Midi Kit	25 preps	CS31104
ChargeSwitch® Pro Filter Plasmid Maxi Kit	10 preps	CS31106
	25 preps	CS31107
ChargeSwitch®-Pro Plasmid Miniprep Kit	10 preps	CS30010
	50 preps	CS30050
	250 preps	CS30250

For more information on the ChargeSwitch® plasmid kits, go to www.invitrogen.com/chargeswitch.

Plasmid DNA Purification Service

We'll take care of your plasmid DNA purification, so you can take care of your research

- High-quality customized purification
- Stringent quality control ensures reliable results
- Ready-to-use plasmid DNA in about 7 days

The Plasmid DNA Purification Service from Life Technologies saves you time and resources by doing the plasmid purification work for you. We will produce ultrapure, low-endotoxin plasmid DNA at any scale—from 1 mg up to 100 mg—for your research or preclinical applications. Purified DNA undergoes rigorous testing to ensure that you receive the quantity and quality of DNA you require.

For more information, go to www.invitrogen.com/customservices and select "Molecular Biology Services" from the menu on the left side of the page.

Overview of genomic DNA purification kits

Life Technologies offers a range of genomic DNA purification kits for sensitive, scalable purification from an expansive set of starting materials to maximize process efficiency and downstream performance. The table below lists many of our genomic DNA purification kits.

Selected genomic DNA purification kits from Life Technologies.

Product	Quantity	Cat. No.	Average yield
Tissue and cells			
PureLink® Genomic DNA Mini Kit	250 preps	K182002	Varies
PureLink® Pro 96 Genomic DNA Purification Kit	4 x 96 preps	K182104A	
MagMAX™ DNA Multi-Sample Kit	50 preps	4413020	Varies with sample type
MagMAX™-96 DNA Multi-Sample Kit	96 preps	4413021	
ChargeSwitch® gDNA Micro Tissue Kit	50 preps	CS11203	Up to 5 µg
ChargeSwitch® gDNA Mini Tissue Kit	25 preps	CS11204	Up to 30 µg
ChargeSwitch® Direct 96 gDNA Kit	1 plate	CS11205	50 ng/well (no elution)
	10 plates	CS11206	
	96 preps (8-well format)	CS11209	
ChargeSwitch® EasyPlex™ 8-Well gDNA Kit	96 preps	CS11211	50 ng/well (no elution)
ChargeSwitch® EasyPlex™ gDNA Kit, 96-well plate	1 plate	CS11207	
DNAzol® Reagent	100 mL	10503027	Varies with starting amount
Bacteria			
ChargeSwitch® gDNA Mini Bacteria Kit	50 preps	CS11301	Up to 12 µg
Forensic samples			
ChargeSwitch® Forensic Kit	100 preps	CS11200	Varies with sample type
ChargeSwitch® gDNA Normalized Buccal Cell Kit	50 preps	CS11020	1–3 ng/µL eluate
ChargeSwitch® gDNA Buccal Cell Kit	50 preps	CS11021	Up to 6 µg
Viruses			
PureLink® Viral RNA/DNA Mini Kit	50 preps	12280050	Varies with sample type
ChargeSwitch® EasyPlex™ Viral RNA/DNA Kit	96 preps	CS1228101	No elution **Blood and Serum**
Plant tissue			
PureLink® Genomic Plant DNA Purification Kit	50 preps	K183001	7 µg
ChargeSwitch® gDNA Plant Kit	96 preps	CS18000	7 µg
Plant DNAzol® Reagent	100 mL	10978021	Varies with starting amount
Blood and serum			
PureLink® Genomic DNA Mini Kit	250 preps	K182002	Varies
PureLink® Pro 96 Genomic DNA Purification Kit	4 x 96 preps	K182104A	
ChargeSwitch® gDNA 50–100 µL Blood Kit	50 preps	CS11000	Up to 3 µg
ChargeSwitch® gDNA 0.2–1 mL Serum Kit	50 preps	CS11040	Up to 200 ng
ChargeSwitch® Direct 96 gDNA Kit	1 plate	CS11205	50 ng (no elution)
ChargeSwitch® EasyPlex™ 8-well gDNA Kit	96 preps	CS11211	50 ng (no elution)
ChargeSwitch® EasyPlex™ gDNA Kit, 96-well plate	1 plate	CS11207	
GeneCatcher™ gDNA 0.3–1 mL Blood Kit	96 preps	CS21101	6–30 µg
GeneCatcher™ gDNA 3–10 mL Blood Kit	200 mL	CS21110	60–300 µg
DNAzol® BD Reagent	100 mL	10974020	Varies with starting amount
Dynabeads® SILANE Genomic DNA	96 preps	370-12D	10 µg
Dynabeads® DNA DIRECT™ Blood Kit	100 preps	631-02	1–5 µg DNA/100 µL blood

Sample size	Advantage	Automation compatible
Up to 1 mL blood (200 µL blood 96-well), 5 x 10^6 cells, 25 mg tissue, bacteria, blood spots, swabs	Robust performance in familiar spin-column format; High-throughput/automation options	iPrep™
Varies with sample	Magnetic bead–based purification for high yield and purity	No
		MagMAX™
3–5 mg tissue	High-quality DNA, fast protocol	iPrep™
25 mg tissue	High-quality DNA, fast protocol	iPrep™
2 x 10^4 cells	No sample transfer that can lead to error or contamination—direct amplification in the same well as DNA binding without elution; high-throughput/automation options	iPrep™
2 x 10^4 cells	GMP for standardized testing labs; nonmagnetic beads; high-throughput/automation options	iPrep™
Scalable	Organic isolation of genomic DNA from blood (mammalian tissues, cells)	No
0.5 mL culture or a colony	Gram-positive and Gram-negative bacteria (magnetic beads)	iPrep™
Varies with sample	Optimized for STR analysis and for poor or environmentally degraded samples (magnetic beads); high-throughput/automation options	iPrep™
1 swab or cells pelleted from mouthwash		iPrep™
1 swab or cells pelleted from mouthwash		iPrep™
10–500 µL	Viral RNA or DNA from cell-free samples (spin column); high-throughput/automation options	iPrep™
30 µL serum	For viral detection/genotyping from 30 µL of cell-free sample; high-throughput/automation options	iPrep™
100 mg tissue	Chloroplasts and low-abundance DNA plant samples (spin column)	iPrep™
100 mg tissue	Plant leaf or seed samples (magnetic beads)	iPrep™
Scalable	Organic isolation of genomic DNA from plant material	No
Up to 1 mL blood (200 µL blood 96-well), 5 x 10^6 cells, 25 mg tissue, bacteria, blood spots, swabs	Robust performance in familiar spin-column format; high-throughput/automation options	iPrep™
50–100 µL blood	For small-volume blood processing	iPrep™
0.2–1 mL serum	Purification from serum samples	No
10 µL blood or 2 x 10^4 cells	No sample transfer that can lead to error or contamination—direct amplification in the same well as DNA binding without elution; high-throughput/automation options	iPrep™
10 µL blood or 2 x 10^4 cells, swabs	GMP for standardized testing labs; nonmagnetic beads; high-throughput/automation options	iPrep™
0.3–1 mL blood	Efficient extraction from large volumes of blood	iPrep™
3–10 mL blood		
Scalable	Isolation of genomic DNA from blood	No
350 µL blood	Highly predictable binding per mg of beads; linear range of DNA yield relative to WBC count	Yes
10 µL sample up to 500 µL sample	Fast protocol, sample lysis to PCR in one tube, scalable, high throughput–compatible	No

PureLink® genomic DNA kits

For high-yield genomic DNA from cells and tissues

- Optimized columns, plate membranes and buffer formulations for enhanced DNA binding and release
- Compatible with a large range of sample types and input amounts
- Flexible format can be processed manually or on automated platforms

The PureLink® Genomic DNA Kits enable high-yield, high-purity DNA extractions from a wide variety of sample types, including blood, tissues, cells, bacteria, swabs, and blood spots, in a familiar silica spin-column or semi-skirted 96-well plate format. An optimized protocol is outlined in the product manual for each different sample type.

PureLink® *Pro* 96 Genomic DNA Kit

The PureLink® *Pro* 96 Genomic DNA Kit is ideal for high-throughput applications. Sample processing is performed with either a vacuum manifold or centrifugation at just 2,100 x *g*, allowing more flexibility in centrifuge choice. The semi-skirted plate design is compatible with most vacuum manifolds on robotic workstations.

PureLink® Genomic DNA Mini Kit

The PureLink® Genomic DNA Mini Kit is optimized to bind and release DNA efficiently. Samples are lysed in an optimized buffer formulated to enhance proteinase K activity and minimize protein contamination. The chaotropic salt binding buffer allows the highest DNA binding of any column method. Powerful wash buffers remove all traces of protein and salt. DNA is eluted in a low-salt buffer to allow for pH stabilization of the DNA in storage.

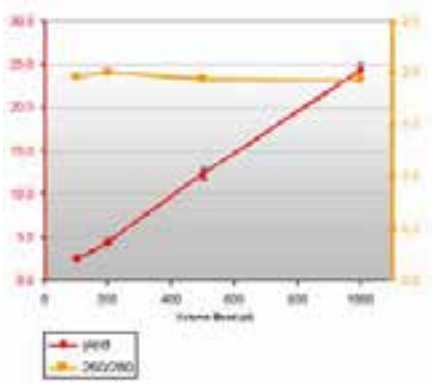


Figure 1. High yields of pure DNA from up to 1 mL of blood using the spin column–based PureLink® Kit. DNA was extracted from the indicated blood volumes using the PureLink® Genomic DNA Kit. Purified gDNA was analyzed on a 1% E-gel® gel. DNA yields increased linearly with volume. In addition, A_{260}/A_{280} ratios were consistently 1.9 for all the samples and replicates tested, indicating that all inhibitors are completely removed.

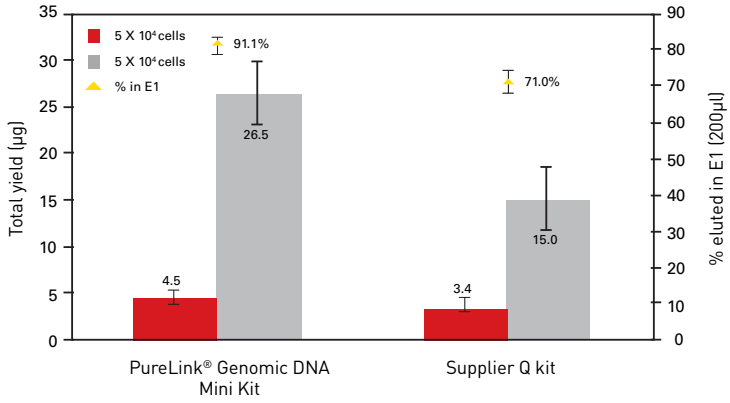


Figure 2. Higher, more concentrated yields achieved with the PureLink® Mini Kit. DNA was extracted from the indicated amount of 293 cells using the PureLink® Genomic DNA Mini Kit and Supplier Q's kit. DNA concentration was measured using spectrophotometry (A_{260}) after two 200-µL elutions. The percentage of yield eluted in the first elution is indicated by the yellow triangles. Significantly higher yields of DNA were obtained using the PureLink® Genomic DNA Mini Kit compared to the competition (p-value = 0.014). In addition, a greater percentage of DNA was recovered in the first elution using the PureLink® kit compared to Supplier Q's kit.

Product	Quantity	Cat. No.
PureLink® Genomic DNA Mini Kit	10 preps	K182000
	50 preps	K182001
	250 preps	K182002
PureLink® *Pro* 96 Genomic DNA Purification Kit	4 x 96 preps	K182104A
PureLink® Genomic Plant DNA Purification Kit	50 preps	K183001

PureLink® Genomic DNA Mini Kits include genomic spin columns, collection tubes, digestion buffer, lysis/binding buffer, wash buffers, elution buffer, proteinase K, and RNase A. PureLink® *Pro* 96 Genomic DNA Kits include genomic binding plates, wash plates, deep well plates, foil tape, digestion buffer, lysis/binding buffer, wash buffers, elution buffer, proteinase K, and RNase A. PureLink® Genomic Plant DNA Purification Kits include genomic spin columns, collection tubes, wash tubes, resuspension/precipitation/binding buffers, wash buffers, elution buffer, SDS, and RNase A.

ChargeSwitch® gDNA Micro and Mini Tissue Kits

Ultrasensitive DNA purification from tissue

- Fast protocol for producing high-quality gDNA
- Reliable purification across multiple tissue types
- No enzymatic inhibitors for improved downstream performance

ChargeSwitch® kits use a magnetic bead–based technology to isolate genomic DNA without the need for hazardous chemicals, centrifugation, or vacuum manifolds.

The ChargeSwitch® gDNA Micro Tissue Kit enables purification of high-quality DNA from the smallest laser microdissected tissue samples and mouse ear clips. Using this kit, purification from a sample of mouse ear clips or 3–5 mg of tissue will typically yield up to 5 µg of pure genomic DNA. The ChargeSwitch® gDNA Mini Tissue Kit purifies high-quality DNA from mini tissue samples. This kit provides a high level of DNA purity, making it ideal for small tissue genotyping and general PCR-based research. Both of these kits avoid the use of enzymatic inhibitors, enhancing performance in downstream applications.

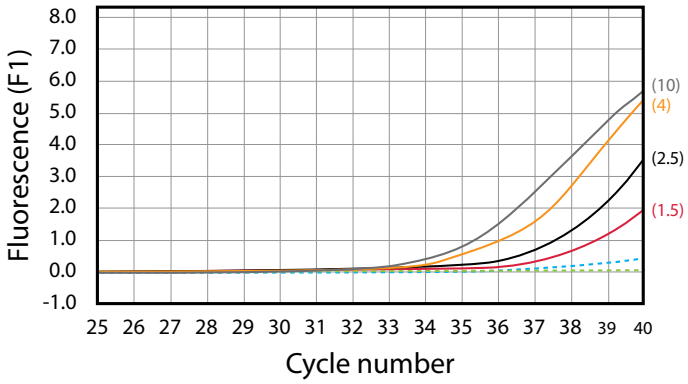


Figure 1. The ChargeSwitch® gDNA Micro Tissue Kit provides ultrasensitive purification from laser capture microdissected samples with a relative sensitivity of 1.5 cells. Diluted aliquots from 50 (black and red) and 20 (orange and gray) pooled, single sperm cells were amplified by PCR for the amelogenin gene.

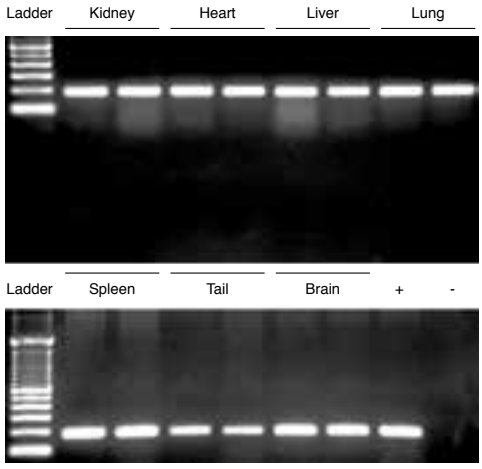


Figure 2. High-efficiency PCR processing using the ChargeSwitch® gDNA Mini Tissue Kit. 186-bp PCR products from PCR amplification of mouse ß-globulin gene from different mouse tissue types in duplicate. PCR products were resolved on a 1.8% agarose gel and visualized with ethidium bromide. Positive control (+): 50 ng genomic mouse DNA control; negative control (–): buffer.

Product	Quantity	Cat. No.
ChargeSwitch® gDNA Micro Tissue Kit	50 preps	CS11203
ChargeSwitch® gDNA Mini Tissue Kit	25 preps	CS11204

ChargeSwitch® gDNA Micro and Mini Tissue Kits are supplied with lysis buffer, magnetic beads, proteinase K, RNase A, purification buffer, wash buffer, and elution buffer.

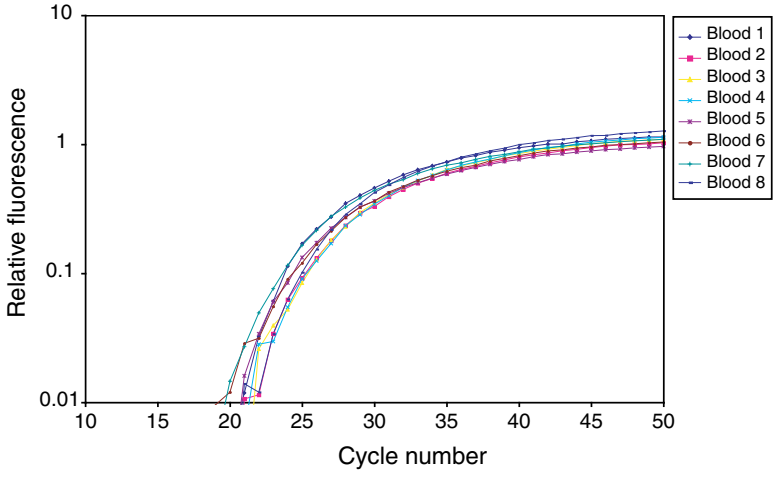
For more information and a complete list of available ChargeSwitch® products, go to www.invitrogen.com/chargeswitch.

ChargeSwitch® Direct 96 gDNA Kit

Isolation to amplification in one well

- Obtain pure gDNA without PCR inhibitors
- Save precious material using small samples
- Minimize contamination and sample loss

The ChargeSwitch® Direct 96 gDNA Kit gives you results faster than ever by going directly from genomic DNA (gDNA) isolation to PCR in one well. There are no beads, membranes, centrifugation, or vacuum manifold steps to deal with—not even a transfer between tubes or plates. The ChargeSwitch® Direct 96 gDNA plates and tubes are coated with the unique ChargeSwitch® surface. At low pH, the surface is positively charged and binds the negatively charged nucleic acid backbone, allowing proteins and other contaminants to be washed away. The purified DNA can then be subjected directly to multiplex PCR, qPCR, STR analysis, or sequencing in the same wells. The ChargeSwitch® Direct 96 gDNA Kit is automation-compatible, and the semi-skirted plates fit into most thermal and real-time cyclers.



Real-time PCR performance of gDNA isolated using the ChargeSwitch® Direct 96 gDNA Kit. Real-time PCR using gDNA isolated from eight different 10-µL human blood samples and amplified directly in the ChargeSwitch® Direct gDNA Plate. PCR was performed using the Certified LUX™ Primer Set for the 18S rRNA gene and Platinum® Quantitative PCR SuperMix-UDG. The eight samples have comparable threshold cycles (C_t values).

Product	Quantity	Cat. No.
ChargeSwitch® Direct 96 gDNA Kit	96 preps (1 plate)	CS11205
	960 preps (10 plates)	CS11206
ChargeSwitch® Direct 8-Well gDNA Kit	96 preps (8 x 12 strips)	CS11209

ChargeSwitch® Direct 96 gDNA Kits contain plate binding and lysis buffer, wash buffers, elution buffer, plate(s), and plate seal(s).

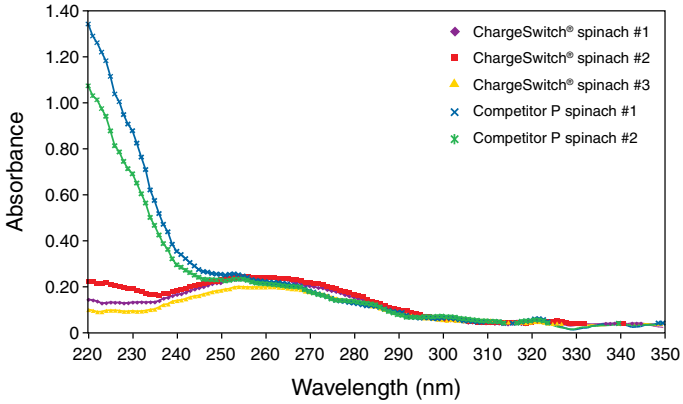
For more information on the ChargeSwitch® kits, go to www.invitrogen.com/chargeswitch.

ChargeSwitch® gDNA plant, forensic, and buccal cell kits

Reliability for genomic DNA purification from specialized samples

- Higher-purity DNA extraction
- Simple, fast process for increasing throughput
- Successful removal of inhibitors without organic solvents

ChargeSwitch® technology, a major advance in DNA purification, is based on a unique, ionizable nucleic acid–binding ligand whose charge can be switched based on the pH of the surrounding medium. Purification is achieved through a simple three-step procedure in aqueous buffers and avoids the use of guanidine, ethanol, and other troublesome reagents. ChargeSwitch® gDNA kits have been adapted for use with plant, forensic, and buccal samples.



Higher-purity DNA obtained using the ChargeSwitch® Kit vs. silica-based kits. DNA samples were isolated from spinach using the ChargeSwitch® gDNA Plant Kit and Competitor P's silica magnetic bead kit. UV spectral analysis of the gDNA shows traces of impurities (high absorbance at 220–230 nm is indicative of guanidine salt contamination) in samples purified with Competitor P's kit.

ChargeSwitch® gDNA plant kit

The ChargeSwitch® gDNA Plant Kit provides the most innovative and successful method for plant DNA extraction—even when working with difficult plant samples. DNA extracted using the ChargeSwitch® method is of higher purity than silica-prepared DNA, which is typically contaminated with guanidine salts. DNA purity is so high compared to other methods that the ChargeSwitch® Kit is routinely used for the most sensitive applications, including genetically modified organism (GMO) screening.

ChargeSwitch® gDNA forensic kits

The ChargeSwitch® Forensic DNA Purification Kit affords high sensitivity and robustness for poor or environmentally degraded samples. This kit is fully validated for single tandem repeat (STR) profiling and is compatible with high-throughput protocols. Use the iPrep™ ChargeSwitch® Forensic Kit when using the iPrep™ Purification System to process your forensic samples.

ChargeSwitch® gDNA buccal cell kits

The ChargeSwitch® gDNA Buccal Cell Kit maximizes the yield of high quality genomic DNA from buccal samples for rapid buccal swab processing. The ChargeSwitch® gDNA Normalized Buccal Cell Kit purifies high-quality genomic DNA from buccal samples at a ready normalized concentration by eliminating the need for lengthy quantitation and dilution steps. This rapid single-tube protocol is easily adapted for high-throughput automation. Both kits purify high-quality DNA that is ideal for STR analysis.

Product	Quantity	Cat. No.
ChargeSwitch® gDNA Plant Kit	96 preps	CS18000
	960 preps	CS1800010
iPrep™ ChargeSwitch® Forensic DNA Purification Kit	52 preps	IS10002
ChargeSwitch® Forensic DNA Purification Kit	100 preps	CS11200
ChargeSwitch® gDNA Buccal Cell Kit	50 preps	CS11021
	960 preps	CS1102110
ChargeSwitch® gDNA Normalized Buccal Cell Kit	50 preps	CS11020
	960 preps	CS1102010

For more information and a complete list of available ChargeSwitch® products, go to www.invitrogen.com/chargeswitch.

MagMAX™ Express magnetic particle processors

Maximum output, minimum time

- Superior nucleic acid recovery and cross-contamination control
- Two configurations for higher or lower throughput
- Compatible with magnetic bead–based sample preparation kits
- Easy to use with preloaded programs
- Reliable service and support from Life Technologies

The MagMAX™ Express processors are the premier automated platforms to meet your throughput needs. They seamlessly incorporate the rapid, reliable, and cost-efficient magnetic bead–based extraction of nucleic acids that you expect from MagMAX™ technology. The MagMAX™ Express Magnetic Particle Processor (Figure 1) utilizes 24 permanent rods (in an array of 2 rows of 12 rods), and the MagMAX™ Express-96 magnetic particle processors (Figure 2) utilize 96 permanent rods to collect magnetic beads from solution and releases the beads into the well containing reagents for the next step of isolation. The effectiveness of bead collection and transfer leads to superior washing, elution efficiency, and rapid processing. Increase performance, throughput, and consistency, all while freeing up lab personnel to handle other activities. The instrument is designed to run without a computer but can be attached to and controlled by a PC (not included). Preloaded software scripts support all MagMAX™ kits. Software can be user-edited and user-programmed to support non–Life Technologies magnetic bead chemistries. Superior support and on-site field service is available.

For more information about MagMAX™ kits, see pages 76–77.

Figure 1. MagMAX™ Express Magnetic Particle Processor.

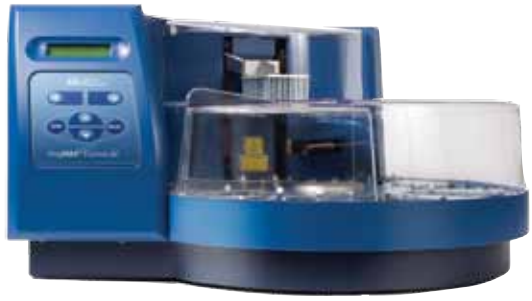


Figure 2. MagMAX™ Express-96 Magnetic Particle Processor.

Product	Quantity	Cat. No.
MagMAX™ Express Magnetic Particle Processor	1 each	4400074
MagMAX™ Express-96 Deep Well Magnetic Particle Processor	1 each	4400077
MagMAX™ Express-96 Standard Magnetic Particle Processor	1 each	4400076

For more information on the MagMAX™ purification system, go to www.invitrogen.com/magmax.

iPrep™ Purification Instrument

Rigorous standardization of your nucleic acid purification process

- Purify up to 13 samples in as little as 18 minutes per run
- Minimize setup variation with prefilled cartridge reagents
- Superior performance in sensitive downstream applications

The iPrep™ Purification Instrument provides a rapid and affordable route into automated DNA purification. It minimizes user-to-user variability, enables reproducibility, and provides the sensitivity and purity required to meet the rigorous standards of your demanding research applications. The iPrep™ Purification Instrument gives you the flexibility to process up to 192 samples per 8-hour workday, permitting a fast turnaround that is crucial to your time-sensitive experiments. Thanks to prefilled reagent cartridges, setting up runs on the iPrep™ instrument for DNA purification is quick, easy, and intuitive.

The easy-to-use iPrep™ Purification Instrument. To isolate nucleic acids using the iPrep™ Purification Instrument, simply insert the iPrep™ Card and select a protocol. Then load the cartridges and tips, add your samples, close the instrument, and press "Start."

The iPrep™ Purification Instrument yields greater amounts of highly pure DNA. Mean yield and purity of DNA purified from various rat tissue samples using either the iPrep™ Purification Instrument or a 6-prep system from Competitor Q.*

	DNA yield (µg)		A_{260}/A_{280} ratio	
Tissue type	iPrep™ system	Competitor Q	iPrep™ system	Competitor Q
Lung	11.9	6.5	1.84	1.82
Heart	7.0	4.6	1.85	1.87
Kidney	11.9	5.9	1.82	1.83
Liver	11.4	5.5	1.83	1.88

* Values are the means of three replicate assays; PCR efficiencies were shown to be 100% with all samples.

Product	Quantity	Cat. No.
iPrep™ Purification Instrument	1 unit	IS10000
Reagents for iPrep™ Purification Instrument		
iPrep™ PureLink® gDNA Blood Kit	52 preps	IS10005
iPrep™ ChargeSwitch® Forensic Kit	52 preps	IS10002
iPrep™ ChargeSwitch® Buccal Cell Kit	52 preps	IS10003
iPrep™ ChargeSwitch® gDNA Tissue Kit	52 preps	IS10004
iPrep™ gDNA Tissue Card	1 card	IS10013
iPrep™ gDNA Forensic Card	1 card	IS10011
iPrep™ PureLink® gDNA Blood Card	1 card	IS10012

For more information on the iPrep™ Purification Instrument, go to www.invitrogen.com/iprep.

BenchPro® 2100 Plasmid Purification System

Plasmid purification automation, typically with less than 5 minutes of hands-on time

- Transfection-grade plasmid with 125 mL of starting culture
- Consistency and reliability through prefilled and sealed reagents
- Minimize tedious hands-on work; requires typically less than 5 minutes to set up

The BenchPro® 2100 Plasmid Purification System provides a complete walk-away solution for high-quality purification of your large-scale plasmid DNA. Going straight from culture to purified plasmid, it simplifies the traditional 22-step process of manual plasmid purification to four easy set-up steps:

1. Grow 125 mL of bacterial culture in LB medium with appropriate antibiotic
2. Assemble the Reagent Tray and Cell Liner on the Waste Tray, and add your culture to the Cell Liner
3. Load the BenchPro® 2100 Plasmid Purification Card into one of the instrument slots
4. Run the purification protocol

The BenchPro® 2100 delivers up to 1 mg of plasmid DNA from 125 mL of culture that is ready for applications such as transfection of mammalian cells, automated and manual sequencing, PCR amplification, cloning, transformation, and *in vitro* transcription.

Figure 1. BenchPro® 2100 Plasmid Purification System.

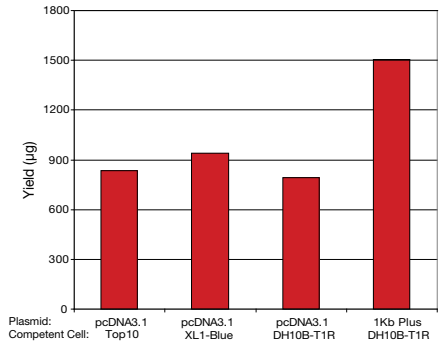


Figure 2. The BenchPro® 2100 Plasmid Purification System is compatible with a wide range of plasmids and cells. This graph shows the average DNA yields of plasmids purified from 125 mL of overnight *E. coli* culture grown in LB medium (OD 2.0–2.4) using the BenchPro® 2100 instrument.

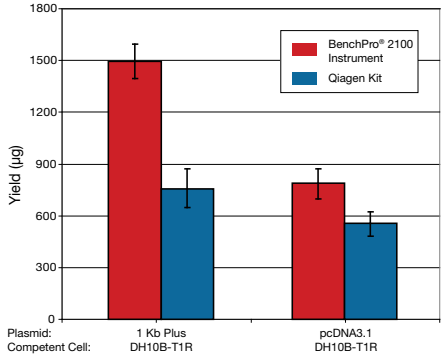


Figure 3. Obtain higher yields using the BenchPro® 2100 Plasmid Purification System. This graph shows the average DNA yields from 125 mL of *E. coli* culture (OD 2.0–2.4) of 1 Kb Plus (19 kb) and pcDNA3.1 (6.2 kb) plasmids purified using the BenchPro® instrument or the Qiagen® HiSpeed® Plasmid Maxi Kit.

Product	Quantity	Cat. No.
BenchPro® 2100 Plasmid Processing Station	1 each	MC1001
BenchPro® 2100 Plasmid Purification Card and Reagent Tray Kit	1 kit (4 cards)	MC2001
BenchPro® 2100 Piercing Device	1 each	MC3001
BenchPro® 2100 Waste Tray	1 kit (2 trays)	MC4001

For more information on the BenchPro® 2100 Plasmid Processing Station, go to www.invitrogen.com/benchpro2100.

Overview of DNA clean-up and gel extraction kits

Whether isolating a specific size of DNA from complex PCR mixtures or recovering bands from agarose gels, Life Technologies has a clean-up solution that will meet your needs.

Selected DNA clean-up and gel extraction kits from Life Technologies.

			Specifications											
	Quantity	Cat. No.	Time: ≤10 min	Scalable elution volume	No guanidine HCl	No ethanol precipitation No alcohol exposure	Size selection possible	Size: 90 bp to 12 kb	Size: 300 bp to 40 kb	Primer removal: >98%	DNA recovery: >80%	Spin column	Magnetic bead	Adapted for 96-well
PureLink® PCR Purification Kit	50 preps 250 preps	K310001 K310002					●	●	●	●	●	●		●
PureLink® PCR Micro Kit	50 preps 250 preps	K310050 K310250	●				●	●		●	●	●		
ChargeSwitch®-Pro PCR Clean-up Kit	10 preps 50 preps 250 preps	CS32010 CS32050 CS32250	●		●	●		●		●	●	●		
ChargeSwitch® PCR Clean-up Kit	100 preps 960 preps	CS12000 CS1200010	●	●	●	●	●	●		●	●		●	●
PureLink® Quick Gel Extraction and PCR Purification Combo Kit	50 preps	K220001					NA			NA	●	●		
PureLink® Quick Gel Extraction Kit	50 preps 250 preps	K210012 K210025					NA	●		NA	●	●		

The table above lists many of our DNA clean-up kits, but for a complete listing and more information, please go to www.invitrogen.com.

PureLink® PCR purification kits

Purify PCR products from complex mixtures of DNA

- Efficient DNA fragment purification and removal of by-products without gel purification
- 15-minute protocol
- Single-column or 96-well plate formats

PureLink® PCR purification kits provide rapid and efficient removal of short primers, dNTPs, enzymes, short-failed PCR products, and salts from PCRs. Two proprietary buffers are supplied with each PureLink® kit. Use the Binding Buffer for routine purifications of double-stranded DNA fragments from 100 bp to 12 kb. Use Binding Buffer HC for removal of primer-dimers and short-failed PCR products (<300 bp).

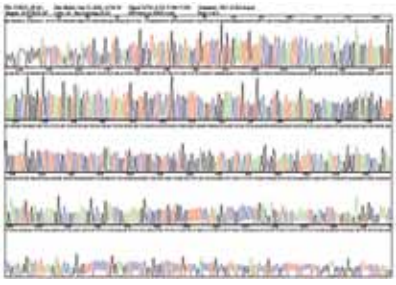
Product	Quantity	Cat. No.
PureLink® PCR Purification Kit	50 preps	K310001
	250 preps	K310002
PureLink® *Pro* 96 PCR Purification Kit	4 plates (4 x 96 rxns)	K310096A
PureLink® PCR Micro Kit	50 preps	K310050
	250 preps	K310250

ChargeSwitch®-Pro PCR Clean-Up Kit

Simple and rapid clean-up of PCR products

- Complete kit for fast, hassle-free setup
- Up to 20% fewer handling steps than competitors' protocols
- Up to 500 µL of PCR product in one column

The ChargeSwitch®-Pro PCR Clean-Up Kit is a fast and effective method of removing primers, unincorporated dNTPs, enzymes, and salts. Unlike most spin-column methods, the ChargeSwitch®-Pro PCR Clean-Up Kit does not use ethanol or chaotropic salts, which means the recovered DNA is of the purest quality and can improve the success of downstream applications such as DNA sequencing, cloning, or PCR. The ChargeSwitch® PCR Clean-Up Kit is also available as a high-throughput option in a magnetic bead format.



Purification kit	No. of bases ≥Phred 20
ChargeSwitch®-Pro PCR Clean-Up Kit	876
QIAquick® PCR Purification Kit	872

Sequencing results from amplicons purified using the ChargeSwitch®-Pro PCR Clean-Up Kit. The ChargeSwitch®-Pro PCR Clean-Up Kit and QIAquick® PCR Purification Kit were used according to the manufacturers' directions. Purified 2-kb sample (100 ng) from each purification was used in BigDye® Terminator sequencing reactions with the T7 primer. **A**. Electropherogram of a sample purified with the ChargeSwitch®-Pro kit. **B**. Phred 20 scores averaged from triplicate samples.

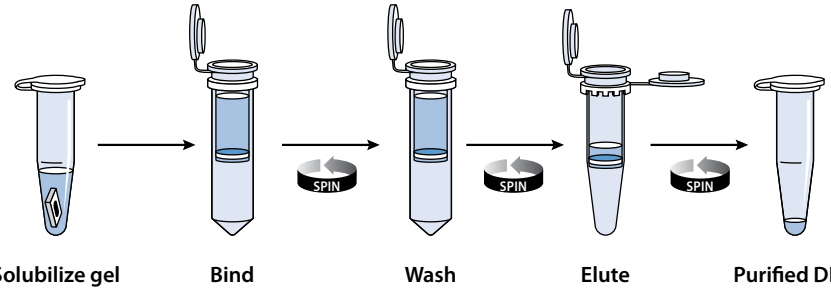
Product	Quantity	Cat. No.
ChargeSwitch®-Pro PCR Clean-Up Kit (spin columns)	10 preps	CS32010
	50 preps	CS32050
	250 preps	CS32250
ChargeSwitch® PCR Clean-Up Kit (magnetic beads)	100 preps	CS12000
	960 preps	CS1200010

PureLink® Quick Gel Extraction Kit

Low-cost gel extraction of DNA samples

- Purify DNA fragments from 40 bp to 10 kb
- Extract and isolate typically in less than 30 minutes
- Obtain DNA free of proteins, dye, and agarose

The PureLink® Quick Gel Extraction Kit is designed to purify DNA fragments from agarose gels. The simple procedure uses a silica-based spin cartridge to purify and isolate DNA that is then ready to use in a variety of applications, including DNA sequencing, PCR, *in vitro* transcription, restriction mapping, cloning, and labeling.



The PureLink® Quick Gel Extraction Kit is quick and easy to use.

Product	Quantity	Cat. No.
PureLink® Quick Gel Extraction Kit	50 preps	K210012
	250 preps	K210025

Nucleic acid gel selection guide

Life Technologies offers many precast gels with different percentages and formats to cover a wide range of nucleic acid separations. It is important to use the correct gel percentage and well format to get the best results for your application. In addition, a variety of nucleic acid gel stains are available. This section provides information to help you match your application with the correct gel.

Gels for nucleic acid separation

E-Gel® gels are self-contained, bufferless, precast agarose gels for separating nucleic acids in the range of 20 bp to 10 kb. They're ideal for analyzing PCR products, restriction digests, and plasmid preparations. Novex® TBE gels are polyacrylamide gels for separating nucleic acids in the range of 10–3,000 bp. These gels are ideal for analyzing oligos or miRNAs, or whenever the highest possible resolution is needed.

Gels for purification

E-Gel® CloneWell™ SYBR® Safe Gels are unique double-comb gels for simultaneous nucleic acid separation and band isolation. They make cloning experiments fast, safe, and efficient.

Choosing the right gel percentage

In general, the size of the molecules being separated should dictate the agarose percentage you choose. Use a lower-percentage gel to resolve larger molecules and a higher-percentage gel to resolve smaller ones. As a general rule, molecules should migrate through 70% of the length of the gel for the best resolution.

Table 1. Choosing a gel type for your appliation.

	Gel % available	Separation range	Shelf life	Average run time	Applications
E-Gel® gel	0.8%, 1.2%, 2%, 4%	20 to 10,000 bp	6 months	10–30 mins	Subcloning and analysis of plasmid DNA, restriction digests, and PCR products
TBE	6%, 8%, 10%, 20%, 4–20%, 4–12%	10 to 3,000 bp	2 months	45 min	Separating double-stranded DNA, oligos, and PCR products
TBE-urea	6%, 10%, 15%	20 to 800 bases	1–2 months	65 min	Separating single-stranded DNA, RNA, and oligos
DNA retardation	6%	30 to 3,000 bp	2 months	90 min	Separating products of gel-shift assays

Safe, sensitive DNA stains

A variety of nucleic acid stains for effective analysis.

SYBR® Safe Stain—a smarter, safer alternative to ethidium bromide

SYBR® Gold Stain—our most sensitive stain for any gel system

SYBR® Green I Stain—ultrasensitive stain for double-stranded DNA, with low background

SYBR® Green II Stain—highly sensitive RNA or ssDNA detection

Table 2. Nucleic acid gel stains selection guide.

Stain	dsDNA	ssDNA	RNA	Safety
SYBR® Safe	◐	○	○	●
SYBR® Gold	●	●	●	○
SYBR® Green I	◐	○	○	○
SYBR® Green II	○	◐	◐	○

Scale: Good ○ Better ◐ Best ●

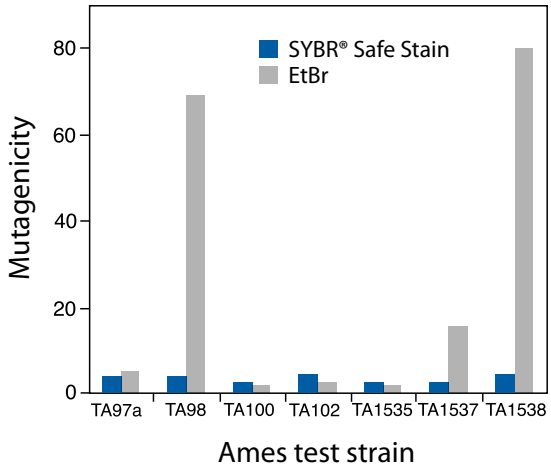
To learn more about nucleic acid separation products, go to www.invitrogen.com/nagels.

SYBR® Safe DNA Gel Stains

Sensitive staining with reduced mutagenicity

- Little or no mutagenicity or carcinogenicity in mammalian cell lines
- As sensitive as ethidium bromide and up to 400 times the sensitivity of colorimetric gel stains
- Cast directly in the gel or use as a post-stain

SYBR® Safe DNA Gel Stain provides sensitive DNA and RNA detection with substantially reduced mutagenicity, making it much safer to use than ethidium bromide. Available as a convenient, ready-to-use solution, it can be detected with a standard UV transilluminator, a visible-light transilluminator, or a laser scanner. According to U.S. Federal Regulations, SYBR® Safe DNA Gel Stain is not considered hazardous waste.



Ames test confirms SYBR® Safe DNA Gel Stain is nonmutagenic in all tested conditions. An increase in revertants of more than two-fold (strains TA97a, TA98, TA100, and TA102) or three-fold (strains TA1535, TA1537, and TA1538) over background indicates a positive result for mutagenicity. Testing was performed by Covance Laboratories, Inc., an independent testing laboratory in Vienna, Virginia.

Product	Quantity	Cat. No.
SYBR® Safe DNA Gel Stain Starter Kit*	1 kit	S33110
SYBR® Safe DNA Gel Stain in DMSO	400 µL	S33102
SYBR® Safe DNA Gel Stain in 0.5X TBE	1 L	S33100
SYBR® Safe DNA Gel Stain in 1X TAE	1 L	S33111
SYBR® Gold Nucleic Acid Gel Stain	500 µL	S11494
SYBR® Green I Nucleic Acid Gel Stain, (10,000X in DMSO)	500 µL	S7563
	1 mL	S7567
SYBR® Green II RNA Gel Stain, (10,000X in DMSO)	500 µL	S7564
	1 mL	S7568
SYBR® Safe Photographic Filter	1 filter	S37100
UltraPure™ 10 mg/mL Ethidium Bromide	10 mL	15585011

* The SYBR® Safe DNA Gel Stain Starter Kit contains 1 L SYBR® Safe DNA Gel Stain and a SYBR® Safe Photographic Filter.

For more information on SYBR® Safe Nucleic Acid Gel Stains, go to www.invitrogen.com/sybrsafe.

E-Gel® precast agarose electrophoresis system

Fast, bufferless agarose electrophoresis

- Quick, convenient, and easy electrophoresis
- Ready-to-use cassettes precast with agarose, electrodes, and in-gel stain
- Ideal for analyzing PCR products, restriction digests, and plasmid preparations

E-Gel® precast gels are self-contained agarose gels designed for fast, convenient, and easy electrophoresis. Each E-Gel® gel is ready to use with agarose, electrodes, and nucleic acid stain packaged inside a dry, disposable, UV-transparent cassette. There are no gels to pour, no buffer to make, no staining/destaining required, and no gel boxes to assemble. Just load your samples and run. E-Gel® gels offer excellent resolution and clarity in as little as 10 minutes and are ideal for analyzing PCR products, restriction digests, and plasmid preparations. See page 109 for ordering sizes, formats, and percentages.

Choose the E-Gel® precast gels that best suit your experimental needs.

Product	Description
E-Gel® EX gels	Single row of 10 sample wells and one marker well
E-Gel® CloneWell™ gels	Two rows of 8 sample wells and one marker well
E-Gel® gels	Single row of 12 sample wells, or two rows with 8 sample wells and one marker well each; spaced for compatibility with 8-channel pipettors for easy multi-sample loading
Clear E-Gel® gels	1.2% agarose without gel stain, in a 12-well, single-comb E-Gel® format for flexible downstream applications, including post-staining with SYBR® Green I or II, SYBR® Gold, or Vistra Green™ dyes, and analyzing fragments prelabeled with fluorescein or Texas Red® dye
E-Gel® *48* Gels	48 sample wells and 4 marker wells compatible with multichannel pipettors or liquid-handling robots for increased throughput
E-Gel® *96* Gels	96 sample wells and 8 marker wells in a unique staggered-well format; compatible with multichannel pipettors and 8-, 12-, and 96-pin liquid-handling robots for high-throughput electrophoresis

E-Gel® precast gel & iBase™ Power System

E-Gel® Safe Imager™ Transilluminator

Complete E-Gel® electrophoresis system

E-Gel® EX Gels

Our fastest, most sensitive, and most flexible precast agarose gels

- Complete separation in just ten minutes
- Ultrasensitive detection of DNA
- Run RNA and DNA samples on the same gel
- Easy access to the gel

E-Gel® EX Gels are the fastest resolving gels in the E-Gel® product line and offer complete resolution of DNA samples typically in just ten minutes. These gels can also be used for a quick check of the integrity of RNA samples before proceeding with downstream applications. The gel cassette is designed to be opened, so that the gel inside can be accessed readily for excision of specific bands or for transfer to a membrane for Southern blot analysis. E-Gel® EX gels were developed for ultimate sensitivity and demonstrate over 5-fold greater sensitivity than comparable gels containing ethidium bromide. The superior sensitivity allows you to use less of your valuable sample.

Product	Quantity	Cat. No.
E-Gel® EX Gel Starter Kit, 1%*	1 kit	G6511STEU (EU adaptor) G6511STUK (UK adaptor)
E-Gel® EX Gel Starter Kit, 2%*	1 kit	G6512STEU (EU adaptor) G6512STUK (UK adaptor)
E-Gel® EX Gel, 1%, 10-Pak	10 gels	G4010-01
E-Gel® EX Gel, 1%, 20-Pak	20 gels	G4020-01
E-Gel® EX Gel, 2%, 10-Pak	10 gels	G4010-02
E-Gel® EX Gel, 2%, 20-Pak	20 gels	G4020-02
E-Gel® EX Gel, 4%, 10-Pak	10 gels	G4010-04

*The E-Gel® EX Gel Starter Kits include an E-Gel® iBase™ Power System, an E-Gel® Safe Imager™ Real-Time Transilluminator, E-Gel® 1 Kb Plus DNA Ladder, gel knife, and a pack of 10 gels containing either 1% or 2% E-Gel® EX Gels.

E-Gel® CloneWell™ SYBR® Safe Gels and E-Gel® iBase™ Power System

Gel-extract your DNA band in three easy steps

- View bands in real time and minimize DNA damage
- Get improved cloning efficiencies
- Eliminate runover using the integrated powerbase with automatic shut-off

E-Gel® CloneWell™ SYBR® Safe Gels are double-comb gels with a twist. Load your sample into the top well and run until your band migrates into the bottom well. Then, easily remove your pure DNA band with a pipette, and you're ready to clone. There are no additional purification kits or steps required. Use the E-Gel® iBase™ Power System to run E-Gel® CloneWell™ SYBR® Safe Gels. SYBR® Safe DNA Gel Stain is incorporated into the gel itself, allowing you to visually monitor migration on a blue-light transilluminator, such as the E-Gel® Safe Imager™ transilluminator. Sensitivity obtained using this transilluminator is comparable to that obtained with a standard UV transilluminator. However, unlike UV transilluminators, the Safe Imager™ transilluminator does not produce UV light and does not require UV protective equipment during use.

Figure 1. E-Gel® nucleic acid analysis—snap, load, run!

Self-contained power system

The E-Gel® iBase™ Power System is compatible with all low-throughput E-Gel® precast gels and is a self-contained device with a built-in power supply. Simply connect the AC adaptor provided and plug it into an electrical outlet. View program selection and running time on the easy-to-read LCD display. Preset programs are available for various gel types, or you can manually set your own run times. A reverse function allows you to change the field direction, allowing for complete capture of your DNA. In addition, the E-Gel® iBase™ Power System includes an automatic shut-off feature, so you won't overrun your gel. The E-Gel® iBase™ Power System fits on the E-Gel® Safe Imager™ Real-Time Transilluminator so you can safely watch the DNA bands migrate through the gel. This system maximizes safety to the user and improves cloning efficiency over ethidium bromide/UV methods.

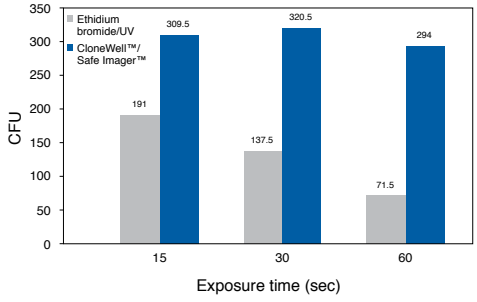


Figure 2. The CloneWell™ system provides higher cloning efficiencies. When compared to traditional ethidium bromide gel extraction methods, the CloneWell™ method results in more CFUs (colony forming units).

Product	Quantity	Cat. No.
E-Gel® CloneWell™ SYBR® Safe Gels and the iBase™ Starter Kit*	1 base and 18 gels	G6400STEU (EU adaptor) G6400STUK (UK adaptor)
E-Gel® iBase™/E-Gel® Safe Imager™ Combo†	1 unit	G4665EU (EU adaptor) G6465UK (UK adaptor)
E-Gel® CloneWell™ 0.8% SYBR® Safe Gels, 18-Pak‡	18 gels	G661808
E-Gel® iBase™ Power System	1 unit	G6400EU (EU adaptor) G6400UK (UK adaptor)
E-Gel® Safe Imager™ Real-Time Transilluminator§	1 unit	G6500
Safe Imager™ 2.0 Blue-Light Transilluminator§	1 unit	G6600

* The E-Gel® CloneWell™ SYBR® Safe Gels and the iBase™ Starter Kit include an iBase™ Power System and 18 E-Gel® CloneWell™ SYBR® Safe Gels.
† The E-Gel® iBase™/E-Gel® Safe Imager™ Combo includes an iBase™ Power System and an E-Gel® Safe Imager™ Real-Time Transilluminator.
‡ See page 109 for additional formats and percentages.
§ The E-Gel® iBase™ Power System is compatible with the E-Gel® Safe Imager™ Real-Time Transilluminator, and the E-Gel® PowerBase™ v.4 is compatible with the Safe Imager™ 2.0 Blue-Light Transilluminator.

For more information and a full product listing of the E-Gel® CloneWell™ system, go to www.invitrogen.com/nagel.

E-Gel® equipment and high-throughput system

E-Base™ integrated power supplies

E-Base™ Integrated Devices are specially designed for running E-Gel® *48*, E-Gel® *96*, E-PAGE™ *48*, and E-PAGE™ *96* gels. Each device features a small footprint for space saving and a built-in power supply. Two types of bases are offered:

1. The Mother E-Base™ device comes with a power cord that can be connected directly to an electrical outlet and is used for electrophoresis of one gel.
2. The Daughter E-Base™ device connects to the Mother E-Base™ device and to other Daughter E-Base™ devices for the simultaneous electrophoresis of two or more gels. The Daughter E-Base™ device does not have a power cord and cannot be used without a Mother E-Base™ device.

E-Holder™ platform

The E-Holder™ platform is designed with a spring-loaded mechanism to ensure that the E-Gel® *96* and E-Gel® *48* gel cassettes stay firmly in place during robotic loading. This allows reproducible placement and loading from one gel to the next without having to make adjustments to the robot loading software. In addition, the E-Holder™ platform allows you to load samples on one E-Gel® *96* or E-Gel® *48* gel while other gels are running in the Mother or Daughter E-Base™ combination.

E-Gel® Opener

The E-Gel® Opener is a simple device specifically designed to quickly and efficiently open an E-Gel® cassette. This allows you to purify DNA fragments from the gel, transfer samples onto a membrane for Southern blot analysis, or post-stain Clear E-Gel® gels. The E-Gel® Opener is safe and easy to use. Simply place the E-Gel® cassette into the E-Gel® Opener and turn the knob to tighten. The E-Gel® Opener uses two steel blades to safely and quickly pop open the E-Gel® cassette without harming your gel. In only a few minutes and with minimal effort, your E-Gel® cassette will be ready for subsequent procedures. For maximum durability, the E-Gel® Opener is made of anodized aluminum.

E-Editor™ 2.0 Software

E-Editor™ 2.0 Software is user-friendly and Windows® software–compatible and quickly arranges and displays your E-PAGE™ *96* System results. This software takes the digital image from your E-PAGE™ *96* gel and reconfigures the staggered lanes into a side-by-side format for easy comparison, analysis, and documentation. Just open up your image (.tif, .jpg, or .bmp) in the E-Editor™ 2.0 program, align the lanes, and arrange. Save the reconfigured image, or copy and paste selected lanes into other applications for further analysis. E-Editor™ 2.0 Software is available free of charge with the purchase of E-PAGE™ *96* gels and related equipment.

The E-Editor™ 2.0 Software and manual are available free of charge with the purchase of E-PAGE™ *96* gels and related equipment. To download, go to www.invitrogen.com/epage.

Product	Quantity	Cat. No.
Mother E-Base™ Integrated Power Supply	1 base	EB-M03EU (EU adaptor) EB-M03UK (UK adaptor)
Daughter E-Base™ Integrated Power Supply	1 base	EB-D03EU (EU adaptor) EB-D03UK (UK adaptor)
E-Holder™	1 holder	EH-03
E-Editor™ 2.0 Software and manual	1 copy	Downloadable
E-Gel® Opener	1 opener	G5300-01
E-Gel® Opener Replacement Blades	10 blades	G5350-10

E-Gel® gels are available in a variety of well formats and agarose percentages.

Product % agarose	Resolution range	Run length	Run time	Quantity	Cat. No.
Double-comb starter paks					
E-Gel® CloneWell™ 0.8% SYBR® Safe Gels	100 bp–6 kb	2.9 cm	14 to 36 min	1 kit*	G6500STEU (EU adaptor) G6500STUK (UK adaptor)
E-Gel® SizeSelect™ 2.0% Agarose	50 bp–2 kb	2.9 cm	8.5 to 20.5 min	1 kit†	G6612STEU (EU adaptor) G6612STUK (UK adaptor)
Double-comb gels					
E-Gel® CloneWell™ 0.8% SYBR® Safe Gels	100 bp–6 kb	2.9 cm	14 to 36 min	18 gels	G6618-08
E-Gel® SizeSelect™ 2.0% Agarose	50 bp–2 kb	2.9 cm	8.5 to 20.5 min	10 gels	G6610-02
Single-comb starter paks					
E-Gel® EX Starter Kit, 1.0%	100 bp–5 kb	5.8 cm	10 min	1 kit‡	G6511ST
E-Gel® EX Starter Kit, 2.0%	50 bp–2 kb	5.8 cm	10 min	1 kit‡	G6512ST
Single-comb gels					
E-Gel® EX 1.0% Gels	100 bp–5 kb	5.8 cm	10 min	10 gels	G4010-01
				20 gels	G4020-01
E-Gel® EX 2.0% Gels	50 bp–2 kb	5.8 cm	10 min	10 gels	G4010-02
				20 gels	G4020-02
E-Gel® EX 4.0% Gels	10 bp–400 bp	5.8 cm	15 min	10 gels	G4010-04
Single-comb starter paks					
E-Gel® gels with SYBR® safe gels					
1.2%	100 bp–5 kb	5.8 cm	30 min	1 kit§	G620601EU (EU adaptor) G620601UK (UK adaptor)
2%	100 bp–2 kb	5.8 cm	30 min	1 kit§	G620602EU (EU adaptor) G620602UK (UK adaptor)
E-Gel® with ethidium bromide gels					
0.8%	800 bp–10 kb	5.8 cm	30 min	1 kit§	G600008EU (EU adaptor) G600008UK (UK adaptor)
1.2%	100 bp–5 kb	5.8 cm	30 min	1 kit§	G600001EU (EU adaptor) G600001UK (UK adaptor)
2%	100 bp–2 kb	5.8 cm	30 min	1 kit§	G600002EU (EU adaptor) G600002UK (UK adaptor)
4%	20 bp–500 bp	5.8 cm	30 min	1 kit§	G600004EU (EU adaptor) G600004UK (UK adaptor)
Single-comb 18-paks					
E-Gel® gels with SYBR® Safe stain					
1.2%	100 bp–5 kb	5.8 cm	30 min	18 gels	G521801
2%	100 bp–2 kb	5.8 cm	30 min	18 gels	G521802
E-Gel® gels with ethidium bromide					
0.8%	800 bp–10 kb	5.8 cm	30 min	18 gels	G501808
1.2%	100 bp–5 kb	5.8 cm	30 min	18 gels	G501801
2%	100 bp–2 kb	5.8 cm	30 min	18 gels	G501802
4%	20 bp–500 bp	5.8 cm	30 min	18 gels	G501804
Double-comb 18-paks					
0.8%	1kb–10 kb	2.9 cm	15 min	18 gels	G601808
2%	100 bp–2 kb	2.9 cm	15 min	18 gels	G601802
E-Gel® *48* gels with ethidium bromide					
E-Gel® *48*, 1%	400 bp–10 kb	3.2 cm	20 min	8 gels	G800801
E-Gel® *48*, 2%	100 bp–2 kb	3.2 cm	20 min	8 gels	G800802
E-Gel® *48*, 4%	10 bp–400 bp	3.2 cm	20 min	8 gels	G800804
E-Gel® *96* gels					
E-Gel® *96* gels with SYBR® Safe stain					
E-Gel® *96*, 2%	100 bp–2 kb	1.6 cm	12 min	8 gels	G720802
E-Gel® *96* gels with ethidium bromide					
E-Gel® *96*, 1%	1 kb–10 kb	1.6 cm	12 min	8 gels	G700801
E-Gel® *96*, 2%	100 bp–2 kb	1.6 cm	12 min	8 gels	G700802

* The E-Gel® CloneWell Starter Pack includes an E-Gel® iBase™ Power System, an E-Gel® Safe Imager™ Real-Time Transilluminator, an amber filter, Safe Imager™ viewing glasses, 18 E-Gel® CloneWell™ gels, and E-Gel® High Range DNA Marker.
† The E-Gel® SizeSelect™ 2% Starter Kit includes 10 E-Gel® SizeSelect™ 2% agarose gels, an E-Gel® iBase™ Power System, an E-Gel® Safe Imager™ Real-Time Transilluminator, and a 50 bp ladder.
‡ E-Gel® EX Starter Kits include an E-Gel® iBase™ Power System, an E-Gel® Safe Imager™ Real-Time Transilluminator, E-Gel® 1 Kb Plus DNA Ladder, gel knife, and a pack of 10 gels containing either 1% or 2% E-Gel® EX gels.
§ E-Gel® Starter Paks include six E-Gel® gels, the E-Gel® PowerBase™ v.4, and a manual.

DNA ladders for E-Gel® precast gels

Preselected DNA ladders for analyzing results on E-Gel® gels

A wide selection of DNA ladders ranging from 10 bp to 12 kb are available for separating bands and accurately estimating the size and quantity of DNA on a variety of E-Gel® formats. The table below indicates the best ladders to use with various E-Gel® formats and agarose percentages.

DNA ladder selection guide for E-Gel® gels.

% agarose	Suggested markers	Quantity	Cat. No.
Single-comb E-Gel® gels			
0.8%	E-Gel® 1 Kb Plus DNA Ladder	100 apps	10488-090
	500 bp DNA Ladder	100 µg	10594018
	High DNA Mass Ladder	50 apps	10496016
1.2%	E-Gel® 1 Kb Plus DNA Ladder	100 apps	10488-090
	100 bp DNA Ladder	50 µg	15628019
	1 Kb Plus DNA Ladder	250 µg	10787018
	High DNA Mass Ladder	50 apps	10496016
2%	E-Gel® 1 Kb Plus DNA Ladder	100 apps	10488-090
	50 bp DNA Ladder	50 µg	10416014
	25 bp DNA Ladder	50 µg	10597011
	1 Kb Plus DNA Ladder	250 µg	10787018
4%	E-Gel® 1 Kb Plus DNA Ladder	100 apps	10488-090
	10 bp DNA Ladder	50 µg	108210115
	25 bp DNA Ladder	50 µg	10597011
	50 bp DNA Ladder	50 µg	10416014
Double-comb E-Gel® gels			
0.8%	High DNA Mass Ladder	50 apps	10496016
	Low DNA Mass Ladder	50 apps	10068013
2%	E-Gel® 1 Kb Plus DNA Ladder	100 apps	10488-090
	1 Kb Plus DNA Ladder	250 µg	10787018
	50 bp DNA Ladder	50 µg	10488085
	Low DNA Mass Ladder	50 apps	10068013
E-Gel® *48* gels			
1%	E-Gel® High Range DNA Ladder	100 apps	12352019
2%, 4%	E-Gel® Low Range Quantitative DNA Ladder	100 apps	12373031
E-Gel® *96* gels			
1%	E-Gel® High Range DNA Ladder	100 apps	12352019
2%	E-Gel® Low Range Quantitative DNA Ladder	100 apps	12373031

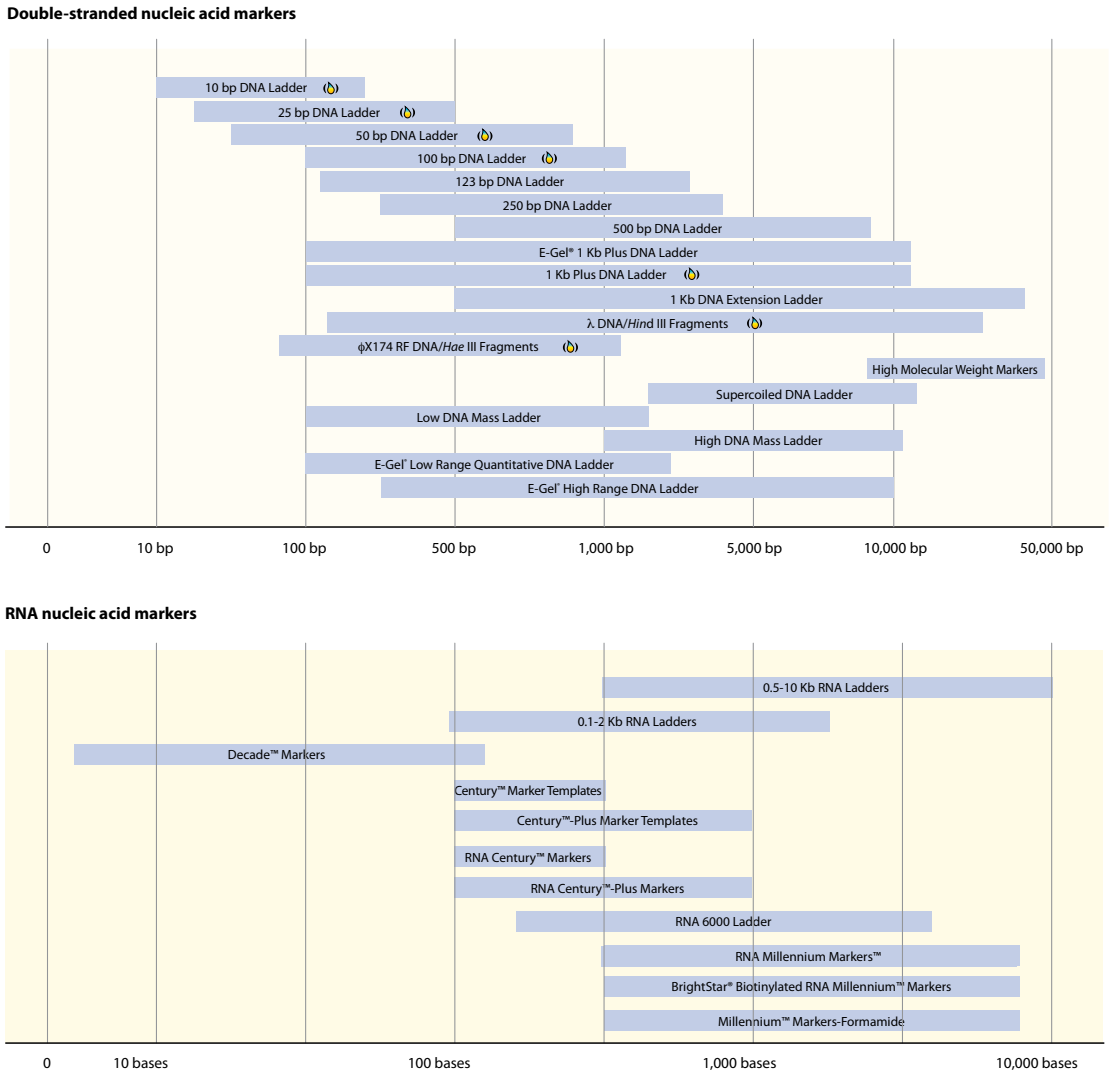
For a complete listing of DNA ladders, please see pages 111–113 or go to www.invitrogen.com/nagels and select "Nucleic Acid Markers" from the menu on the left side of the page.

Overview of nucleic acid markers

Life Technologies supplies a wide range of products for accurate size and mass estimations (quantitation) of nucleic acid fragments. Nucleic acid markers are available for sizing double-stranded, single-stranded, or supercoiled DNA as well as single-stranded RNA fragments. A variety of these markers are also available in the ready-to-load TrackIt™ format. There is no need to heat, mix, or dilute these markers prior to loading them on your gel. TrackIt™ markers offer two tracking dyes, which indicate when maximum resolution of the DNA fragments has been achieved.

Nucleic acid marker selection guide

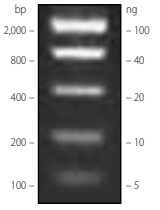
Life Technologies offers unique nucleic acid markers for a wide variety of size ranges, applications, and formats. The key to accurate band analysis is to use the correct marker or standard for your particular application. Use the following figure to help you choose the one you need.



Nucleic acid marker selection guide.

Nucleic acid ladders

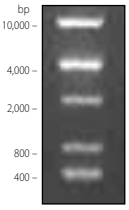
Clear band resolution with ready-to-load ladders from Life Technologies



E-Gel® Low Range Quantitative DNA Ladder

10 µL run on a E-Gel® 2% double comb gel.

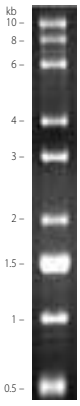
Cat. No. 12373031



E-Gel® High Range DNA Ladder

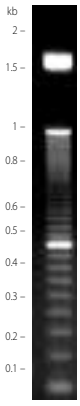
10 µL run on a E-Gel® 0.8% double comb gel.

Cat. No. 12352019



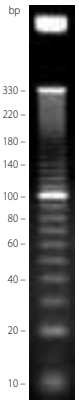
0.5–10 Kb RNA Ladder

Cat. No. 15623200



0.1–2 Kb RNA Ladder

Cat. No. 15623100



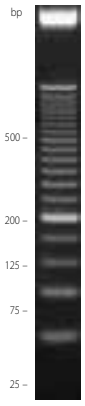
10 bp DNA Ladder

2 µg/lane; 4% LMP agarose gel stained with ethidium bromide.

Cat. No. 10821015

TrackIt™ 10 bp DNA Ladder

Cat. No. 10488019



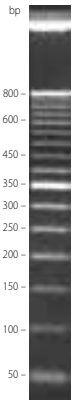
25 bp DNA Ladder

0.5 µg/lane; 2% Agarose 1000 in 1X TBE stained with ethidium bromide.

Cat. No. 10597011

TrackIt™ 25 bp DNA Ladder

Cat. No. 10488022



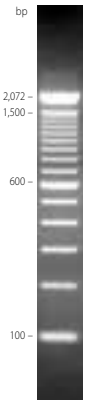
50 bp DNA Ladder

0.5 µg/lane; 2% agarose gel stained with ethidium bromide.

Cat. No. 10416014

TrackIt™ 50 bp DNA Ladder

Cat. No. 10488043



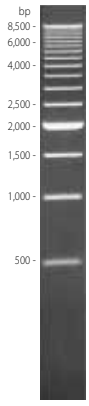
100 bp DNA Ladder

3 µg/lane; 1.5% agarose gel stained with ethidium bromide.

Cat. No. 15628019

TrackIt™ 100 bp DNA Ladder

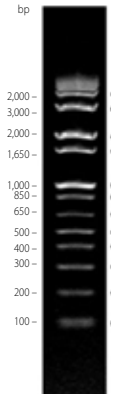
Cat. No. 10488058



500 bp DNA Ladder

0.75 µg/lane; 1% agarose gel stained with ethidium bromide.

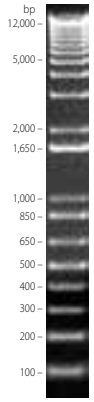
Cat. No. 10594018



E-Gel® 1 Kb Plus DNA Ladder

175 ng/lane; 1% E-Gel® gel

Cat. No. 10488-090



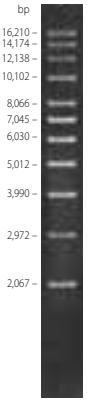
1 Kb Plus DNA Ladder

0.9 µg/lane; 0.9% agarose gel stained with ethidium bromide.

Cat. No. 10787018

TrackIt™ 1 Kb Plus DNA Ladder

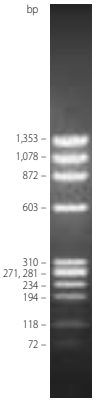
Cat. No. 10488085



Supercoiled DNA Ladder

0.2 µL/lane: 0.9% agarose gel containing 2 µg/mL ethidium bromide.

Cat. No. 15622012



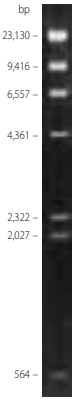
ϕX174 RF DNA/ *Hae* III Fragments

2 µg/lane; 1.5% agarose gel stained with ethidium bromide.

Cat. No. 15611015

TrackIt™ ϕX174 RF DNA/ Hae III Fragments

Cat. No. 10488037



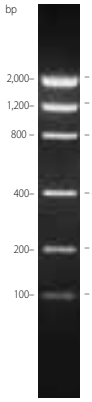
λ DNA/*Hin*d III Fragments

1 µg/lane; 1.5% agarose gel stained with ethidium bromide.

Cat. No. 15612013

TrackIt™ λ DNA/Hind III Fragments

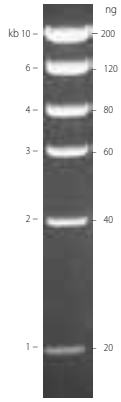
Cat. No. 10488064



Low DNA Mass Ladder

4 µL/lane; 2% agarose gel stained with ethidium bromide.

Cat. No. 10068013



High DNA Mass Ladder

4 µL/lane; 1% agarose gel stained with ethidium bromide.

Cat. No. 10496016

Nucleic acid marker selection guide.

Summary of ladders

Selected nucleic acid markers from Life Technologies.

Product	Quantity	Cat. No.
TrackIt™ Ladders—ready-to-run formats for ultimate convenience and clear visualization		
TrackIt™ 10 bp DNA Ladder*	0.5 mg/mL (20 apps)	10488019
TrackIt™ 25 bp DNA Ladder*	0.5 mg/mL (20 apps)	10488022
TrackIt™ 50 bp DNA Ladder*	0.1 mg/mL (100 apps)	10488043
TrackIt™ 100 bp DNA Ladder*	0.1 mg/mL (100 apps)	10488058
TrackIt™ 1 Kb Plus DNA Ladder†	0.1 mg/mL (100 apps)	10488085
TrackIt™ φX174 RF DNA/Hae III Fragments	0.1 mg/mL (100 apps)	10488037
TrackIt™ λDNA/Hind III Fragments	0.1 mg/mL (100 apps)	10488064
DNA ladders—traditional DNA ladders for sizing linear, double-stranded or single-stranded DNA fragments		
10 bp DNA Ladder*	50 µg at 1 mg/mL	10821015
25 bp DNA Ladder*	50 µg at 1 mg/mL	10597011
50 bp DNA Ladder*	50 µg at 1 mg/mL	10416014
100 bp DNA Ladder*	50 µg at 1 mg/mL	15628019
	250 µg at 1 mg/mL	15628050
123 bp DNA Ladder†	100 µg at 1 mg/mL	15613011
	250 µg at 1 mg/mL	15613029
250 bp DNA Ladder*	75 µg at 1 mg/mL	10596013
500 bp DNA Ladder*	100 µg at 1 mg/mL	10594018
1 Kb Plus DNA Ladder†	250 µg at 1 mg/mL	10787018
1 µg at 1 mg/mL	1 µg at 1 mg/mL	10787026
1 Kb DNA Extension Ladder	100 µg at 1 mg/mL	10511012
Supercoiled DNA Ladder§	25 µg at 0.25 mg/mL	15622012
High Molecular Weight DNA Makers	50 µg at 40 mg/mL	15618010
φX174 RF DNA/Hae III Fragments	40 µg at 0.5 mg/mL	15611015
λDNA/Hind III Fragments	500 µg at 0.5 mg/mL	15612013
Quantitation ladders—equimolar mixtures of DNA band fragments for estimating the mass of DNA samples		
E-Gel® Low Range Quantitative DNA Ladder‡	100 apps	12373031
E-Gel® High Range Quantitative DNA Ladder‡	100 apps	12352019
Low DNA Mass Ladder‡	50 apps	10068013
High DNA Mass Ladder‡	50 apps	10496016
RNA ladders—consist of RNA fragments for sizing single-stranded RNA in glyoxal or formaldehyde agarose gels		
0.1-2 Kb RNA Ladder	75 µg, 1 µg /µL	15623100
0.5-10 Kb RNA Ladder	75 µg, 1 µg /µL	15623200

* Can be radiolabeled using T4 DNA polymerase or T4 polynucleotide kinase

† Can be radiolabeled using T4 DNA polymerase, T4 polynucleotide kinase, DNA polymerase I, or the Klenow fragment

‡ Suitable for estimating mass (quantity) of unknown DNA samples by ethidium bromide staining

§ Can be probed with pBR322 ß-lactamase (ampicillin-resistance) gene sequence

For more information and a complete list of nucleic acid markers and ladders, go to www.invitrogen.com/nagels and select "Nucleic Acid Markers" from the menu on the left side of the page.

Nucleic acid gel sample loading buffers

Life Technologies offers a variety of gel loading buffers for easy loading and tracking of DNA samples. Here, we highlight the loading buffers that are primarily for use with agarose gels.

TrackIt™ loading buffers

- TrackIt™ Cyan/Orange Loading Buffer—for DNA fragments between 10 bp and 1 Kb.
- TrackIt™ Cyan/Yellow Loading Buffer—for DNA fragments between 100 bp and 10 Kb.

The TrackIt™ loading buffers are a 6X solution used for easy loading and tracking of DNA samples in agarose gels, including E-Gel® precast agarose gels. These loading buffers contain two tracking dyes, one that runs behind the sample and one that runs ahead of the sample. The tracking dyes serve as visual markers for following migration during electrophoresis and indicate when maximum resolution of the DNA fragments has been achieved. Additionally, visualization of DNA bands will not be obscured by the tracking dyes because they run outside the limits of most DNA samples. The TrackIt™ Cyan/Orange Loading Buffer contains Xylene Cyanol FF and Orange G dyes, and the TrackIt™ Cyan/Yellow Loading Buffer contains xylene cyanol FF and tartrazine dyes.

BlueJuice™ Gel Loading Buffer

- All-purpose loading buffer for DNA
- Compatible with agarose and native polyacrylamide gels

BlueJuice™ Gel Loading Buffer is designed for easy loading and tracking of DNA samples in agarose, including E-Gel® precast agarose gels, or native polyacrylamide gels. The recommended concentration of this buffer for use with all DNA samples run on agarose gels is 2X (one part buffer plus four parts sample). Any concentration may be used in agarose gels without affecting band appearance (except for bands that may be obscured by the bromophenol blue tracking dye). For acrylamide gels, the recommended concentration is 1X (one part buffer plus nine parts sample). Concentrations higher than 1X applied to native polyacrylamide gels may cause the bands to "smile" slightly.

E-Gel® Sample Loading Buffer

- Proprietary formulation optimized for all types of E-Gel® gels

E-Gel® Sample Buffer is supplied as a ready-to-use 1X solution, and is formulated specifically for maximum performance on E-Gel® EX gels as well as other E-Gel® precast gels. This buffer contains xylene cyanol FF and tartrazine dyes.

Product	Quantity	Cat. No.
TrackIt™ Cyan/Orange Loading Buffer (agarose gels)*	3 x 0.5 mL	10482028
TrackIt™ Cyan/Yellow Loading Buffer (agarose gels)†	3 x 0.5 mL	10482035
BlueJuice™ Gel Loading Buffer (agarose or native polyacrylamide gels)‡	3 x 1 mL	10816015
E-Gel® Sample Loading Buffer (E-gel® precast agarose gels)§	4 x 1.25 mL	10482-055

* TrackIt™ Cyan/Orange Loading Buffer is a 6X solution composed of 30% (v/v) glycerol, 60 mM Tris-HCl (pH 7.5), 60 mM EDTA, 0.36% (w/v) XCFF, and 2.4% (w/v) Orange G.

† TrackIt™ Cyan/Yellow Loading Buffer is a 6X solution composed of 30% (v/v) glycerol, 60 mM Tris-HCl (pH 7.5), 60 mM EDTA, 0.36% (w/v) XCFF, and 3.6% (w/v) tartrazine.

‡ BlueJuice™ Gel Loading Buffer is a 6X solution composed of 65% (w/v) sucrose, 10 mM Tris-HCl (pH 7.5), 10 mM EDTA, and 0.3% (w/v) bromophenol blue.

§ E-Gel® Sample Loading Buffer is a 1X solution composed of 0.0094% XCFF and 0.06% tartrazine.

For a complete listing of nucleic acid gel loading buffers, go to www.invitrogen.com.

Overview of UltraPure™ reagents

Life Technologies offers a wide range of UltraPure™ molecular biology reagents, including water, salts, urea, cesium chloride, guanidine, glycerol, formamide, and sheared sperm/thymus DNA, as well as a variety of agarose, buffer, and phenol formulations. All UltraPure™ reagents are made from the purest biochemicals for maximum reliability and superior performance.

Selected UltraPure™ products from Life Technologies.

Products	Quantity	Cat. No.
UltraPure™ Agarose	500 g	16500500
UltraPure™ Agarose 1000	100 g	16550100
UltraPure™ Low Melting Point Agarose	100 g	16520100
UltraPure™ 10X TBE Buffer	1 L	15581044
UltraPure™ 10 mg/mL ethidium bromide	10 mL	15585011
UltraPure™ DNase/RNase-free distilled water	10 x 500 mL	10977049
UltraPure™ Salmon sperm DNA solution	5 x 1 mL	15632011
UltraPure™ 20X SSC	4 L	15557036
UltraPure™ DEPC-treated water	1 L	750023
UltraPure™ 0.5 M EDTA, pH 8.0	500 g	15576028
UltraPure™ 20X SSPE	10 L	15591027
UltraPure™ 10% SDS solution	1 L	24730020
UltraPure™ Formamide	500 g	15515026
UltraPure™ DNase/RNase-free distilled water	1 x 500 mL	10977035
UltraPure™ Herring sperm DNA solution	5 x 1 mL	15634017
UltraPure™ Tris HCl	500 g	15506017
UltraPure™ 5 M NaCl	10 L	24740011
50X Denhardt's solution	100 mL	750018

For a complete list of UltraPure™ reagents and sizes, go to www.invitrogen.com/nagels and select "UltraPure™ Reagents" from the menu on the left side of the page.

UltraPure™ agarose products

- Separate nucleic acid fragments into sharp, distinct bands
- Environmentally friendlier packaging—pouch with 75% less plastic than the original bottle
- Easy-pour spout for reduced contamination

Agarose gel electrophoresis remains the most widely used technique for separating nucleic acid fragments due to its ease of use, nontoxicity, and broad separation range. By varying the agarose concentration, gel pore diameter can be varied to separate nucleic acid molecules with a wide range of sizes. The migration of nucleic acids in agarose gels is affected by the choice of buffer and applied voltage. Life Technologies offers a range of DNase- and RNase-free UltraPure™ agarose products to meet all your nucleic acid electrophoresis needs.

Agarose selection guide for nucleic acid electrophoresis.

Application	>1 kb fragments	<1 kb fragments
DNA/RNA analytical separation	UltraPure™ Agarose	UltraPure™ Agarose-1000
Southern and northern blotting	UltraPure™ Agarose	UltraPure™ Agarose-1000
DNA/RNA recovery	UltraPure™ Low Melting Point Agarose	UltraPure™ Agarose-1000
In-gel manipulations	UltraPure™ Low Melting Point Agarose	—
High resolution of PCR fragments	—	UltraPure™ Agarose-1000
Tissue/cell culture	UltraPure™ Low Melting Point Agarose	—
Viral titer assays	UltraPure™ Low Melting Point Agarose	—
Ouchterlony	UltraPure™ Agarose	—
Radial immunodiffusion	UltraPure™ Agarose	—

Additional featured UltraPure™ reagents

UltraPure™ water

UltraPure™ DNase/RNase-Free Distilled Water is designed for use in all molecular biology applications. It is 0.1 µm membrane-filtered and tested for DNase and RNase activity.

UltraPure™ DEPC-treated Water is suitable for use with RNA. It is prepared by incubating with 0.1% diethylpyrocarbonate (DEPC) and is then autoclaved to remove the DEPC, sterile filtered, and tested for DNase and RNase activity.

UltraPure™ 10X TAE Buffer

UltraPure™ 10X TAE Buffer is a sterile-filtered solution of 0.4 M Tris-acetate and 0.01 M EDTA. TAE buffer is commonly used for agarose DNA electrophoresis. It is supplied in a 1-L plastic bottle or in a 4-L or 10-L stackable Cubitainer® Box.

UltraPure™ 10X TBE Buffer

UltraPure™ 10X TBE Buffer is a sterile-filtered solution of 1 M Tris, 0.9 M boric acid, and 0.01 M EDTA used to prepare 1X buffer for polyacrylamide and agarose gel electrophoresis. This product is available in a 1-L plastic bottle or in a 10-L Cubitainer® Box.

UltraPure™ Tris

UltraPure™ Tris [tris(hydroxymethyl)aminomethane] is widely used in buffers because of its buffering range (pH 7.0–9.0) and compatibility with many enzymes, including restriction endonucleases and DNA-modifying enzymes.

UltraPure™ DNA solutions

UltraPure™ DNA solutions from Calf Thymus, Salmon Sperm, or Herring Sperm are ready-to-use, sheared DNA solutions that are used directly in the preparation of prehybridization and hybridization solutions. These prepared solutions are used to block the non-specific attachment of probe to the surface of a membrane. The DNA solutions are prepared from highly pure, phenol/chloroform-extracted DNA and DNase-free and RNase-free (DEPC-treated) distilled, deionized water and are sheared to an average size of ≤2,000 bp. The concentration is adjusted to 10 mg/mL.

UltraPure™ phenol solutions

UltraPure™ Phenol is used in the purification of nucleic acids. In a mixture of phenol and buffered aqueous solution, proteins are denatured and collect at the interphase while most nucleic acids remain in the aqueous phase.

UltraPure™ Phenol:Chloroform:Isoamyl Alcohol (25:24:1, v/v) is used in the purification of nucleic acids. This reagent consists of highly pure chloroform, isoamyl alcohol, and UltraPure™ Phenol saturated with Tris-HCl. When mixtures are extracted with Phenol:Chloroform:Isoamyl Alcohol, proteins are denatured and collected in the organic phase or at the interphase, while nucleic acids remain in the aqueous phase.

UltraPure™ Phenol:Water (3.75:1, v/v) is used in the purification of nucleic acids. This liquid reagent consists of UltraPure™ Phenol and highly pure deionized water. The reagent can be used to prepare different types of buffer-saturated phenol. When mixtures are extracted with buffer-saturated phenol, proteins are denatured and collect in the organic phase or at the interphase, while nucleic acids remain in the aqueous phase.

All UltraPure™ phenol solutions contain no preservatives and are packaged under an inert gas in shatter-resistant, plastic-coated amber bottles.

Product	Quantity	Cat. No.
UltraPure™ DNase/RNase-Free Distilled Water	10 x 500 mL	10977049
UltraPure™ DEPC-Treated Water	1 L	750023
UltraPure™ 10X TAE Buffer	4 L 1 L	15558026 15581044
UltraPure™ 10X TBE Buffer	1 L	15581044
UltraPure™ Tris	1 kg	15504020
UltraPure™ Tris HCl	500 g	15506017
UltraPure™ Calf Thymus DNA Solution	5 x 1 mL	15633019
UltraPure™ Salmon Sperm DNA Solution	5 x 1 mL	15632011
UltraPure™ Herring Sperm DNA Solution	5 x 1 mL	15634017
UltraPure™ Phenol	500 g	15509037
UltraPure™ Phenol:Chloroform:Isoamyl Alcohol (25:24:1, v/v)	100 mL 400 mL	15593031 15593049
UltraPure™ Phenol:Water (3.75:1, v/v)	400 mL	15594047

For a complete list of UltraPure™ reagents and sizes, go to www.invitrogen.com/nagels and select "UltraPure™ Reagents" from the menu on the left side of the page.

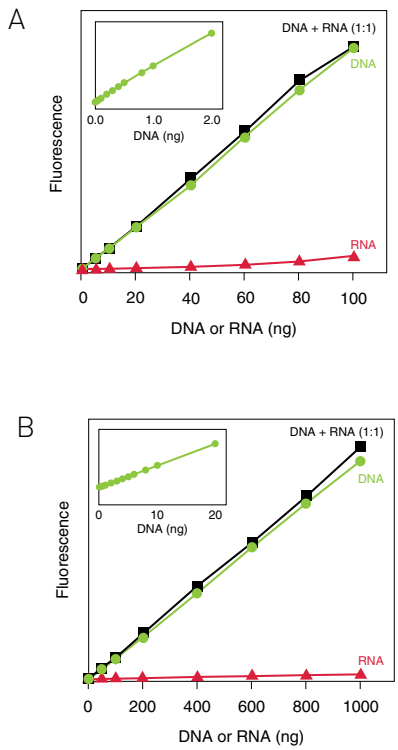
Quant-iT™ technology

Accurate, sensitive, and specific quantification of DNA and RNA

- Selectivity and accuracy unsurpassed by absorbance assays
- Effectively quantitate dilute and low-abundance samples
- Available for DNA and RNA samples

Quant-iT™ technology employs extreme selectivity not possible with absorbance measurements, resulting in accuracy high enough to quantitate even the most dilute or low-abundance samples, while still leaving enough sample for downstream applications. Upon binding to nucleic acid, the fluorescence of the Quant-iT™ dyes increases several hundred-fold, giving a very high signal-to-background ratio for exceedingly high sensitivity—up to 1,000 times more sensitive than absorbance readings.

Product	Quantity	Cat. No.
Quant-iT™ PicoGreen® dsDNA Assay Kit	1 kit, 1 mL	P7589
Quant-iT™ OliGreen® ssDNA Assay Kit	1 kit	O11492
Quant-iT™ RiboGreen® RNA Assay Kit	1 kit	R11490
Quant-iT™ RNA Assay Kit	1 kit	Q33140
Quant-iT™ Broad-Range DNA Assay Kit	1 kit	Q33130
Quant-iT™ High-Sensitivity DNA Assay Kit	1 kit	Q33120



Quant-iT™ High-Sensitivity and Broad-Range DNA Assay Kits. The Quant-iT™ High-Sensitivity DNA Assay Kit (A) and the Quant-iT™ Broad-Range DNA Assay Kit (B) have a linear detection range of 0.2–100 ng and 2–1,000 ng, respectively. Each kit is selective for dsDNA, even in the presence of an equal mass of RNA. The x-axis gives the mass of nucleic acid at a given point when DNA or RNA is assayed alone. In the 1:1 mixture, the total mass of nucleic acid at a given point is double what is stated on the x-axis.

Qubit® 2.0 Fluorometer

Accurate, precise benchtop quantitation of DNA and RNA

- Selective—each Qubit® assay kit is highly sensitive for a single analyte (RNA, DNA, or protein)
- Sensitive—samples with concentrations as low as 10 pg/µL of DNA and 12.5 µg/mL of protein may be accurately and reliably quantitated
- Simple and intuitive—the Qubit® 2.0 Fluorometer provides the same high accuracy you've come to expect but now is even faster and requires less effort to use.

The Qubit® 2.0 Fluorometer utilizes specifically designed fluorometric technology using Molecular Probes® dyes that only fluoresce when bound to DNA, RNA, or protein. These fluorescent dyes emit signals ONLY when bound to specific target molecules, even in the presence of free nucleotides or degraded nucleic acids. This specificity allows you to get very accurate results because Qubit® technology only reports the concentration of the molecule of interest, not contaminants. Qubit® fluorometric quantitation provides exceptionally specific and sensitive DNA and RNA quantitation, even at low concentrations.

Qubit® 2.0 Fluorometer

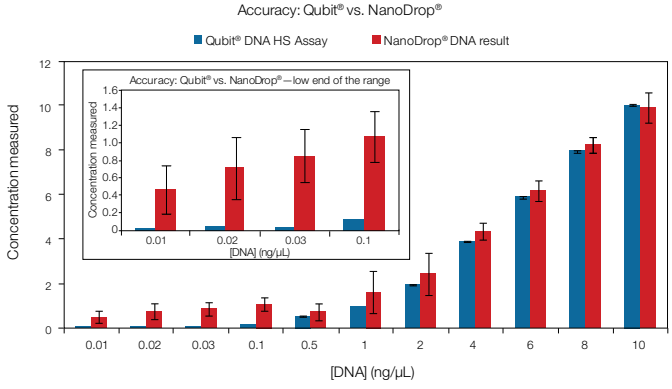
The new features include:

- Large LCD color touch screen
- Automatic data logging and USB port for data management
- Easy workflow navigation
- Standard curve display after calibration completion

The NanoDrop® and other UV spectrophotometers use UV absorbance, which cannot distinguish between DNA, RNA, degraded nucleic acids, free nucleotides, and other contaminants. The Qubit® Quantitation Platform, in contrast, uses fluorescent dyes to measure the concentration of the specific molecules of interest.

Although UV absorbance is one of the most common methods used to quantitate DNA or RNA, it can be unreliable and inaccurate [1–4]. UV absorbance readings indiscriminately measure anything that absorbs at 260 nm, including DNA, RNA, protein, degraded nucleic acids, and free nucleotides. While measurements are typically lower than A_{260} measurements, quantitation by the Qubit® 2.0 Fluorometer is more accurate since it detects only the molecule of interest.

In addition, the sensitivity of spectrophotometry is often inadequate, prohibiting quantitation of DNA and RNA at low concentrations. In contrast, the Qubit® 2.0 Fluorometer generates more accurate and precise results across a lower concentration range than those obtained by UV absorbance measurements on the NanoDrop® spectrophotometer (see figure). Due to this accuracy and precision, fluorescent quantitation of nucleic acids is recommended in the MIQE (Minimal Information for Publication of Quantitative Real-Time PCR Experiments) Guidelines [5].



Accuracy and precision of the Qubit® 2.0 fluorometer. Ten replicates of lambda DNA at concentrations from 0.01 to 10 ng/µL were assayed using the Qubit® DNA HS Assay on the Qubit® 2.0 Fluorometer according to the standard kit protocol. The same concentrations of DNA were measured in ten replicates using a NanoDrop® ND-1000 Spectrophotometer, and results were compared for both accuracy and precision. Each bar represents the average of 10 replicates. Error bars represent the standard deviations of the 10 replicates. The concentrations indicated are the concentrations of DNA in the starting samples, before dilution in the Qubit® assay tubes.

Product	Quantity	Cat. No.
Qubit® 2.0 Fluorometer*	1 instrument	Q32866
Qubit® 2.0 Quantitation Starter Kit†	1 kit	Q32871
Qubit® 2.0 dsDNA HS Assay Kit (0.2–100 ng) for Qubit® 2.0 Fluorometer‡	100 assays	Q32851
Qubit® 2.0 dsDNA BR Assay Kit (2–1,000 ng) for Qubit® 2.0 Fluorometer‡	100 assays	Q32850
Qubit® 2.0 ssDNA Assay Kit (1–200 ng) for Qubit® 2.0 Fluorometer‡	100 assays	Q10212
Qubit® 2.0 RNA Assay Kit (5–100 ng) for Qubit® 2.0 Fluorometer‡	100 assays	Q32852
Qubit® 2.0 RNA BR Assay Kit (20–1,000 ng) for Qubit® 2.0 Fluorometer‡	100 assays	Q10210

* The Qubit® 2.0 Fluorometer is supplied with a 9 V universal power supply and four plug adapters.

† The Qubit® 2.0 Quantitation Starter Kit includes 1 Qubit® 2.0 Fluorometer, 1 Qubit® dsDNA HS Assay Kit (100 assays), 1 Qubit® dsDNA BR Assay Kit (100 assays), 1 Qubit® RNA Assay Kit (100 assays), 1 Qubit® Protein Assay Kit (100 assays), and 500 Qubit® assay tubes.

References

1. Glasel JA (1995) *Biotechniques* 18:62–63.
2. Huberman JA (1995) *Biotechniques* 18:636.
3. Manchester KL (1995) *Biotechniques* 19:208–210.
4. Manchester KL (1996) *Biotechniques* 20:968–970.
5. Bustin SA (2010) *Clinical Chemistry* 55:611–622.

Qubit® assay kits for the Qubit® 2.0 Fluorometer kits contain reagent, premade calibration standards, and premade buffer and are also available in quantities of 500 assays. Please go to www.invitrogen.com/qubit for a full product list.

Notes

end-point PCR

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Overview of PCR

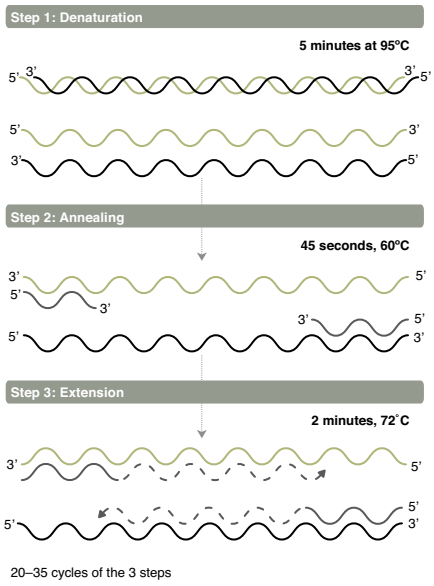
PCR (polymerase chain reaction) is a technique that is central to molecular biology research. Developed by Kary Mullis in 1983, this technique revolutionized genetic research, opening many doors to new applications in medicine and biotechnology. PCR is currently used in a variety of applications ranging from cloning, gene expression analysis, genotyping, sequencing, resequencing, methylation, and mutagenesis. PCR is also used in research for infectious diseases, cancer, and in forensic investigation. It is also a critical tool in agricultural biotechnology at numerous steps from discovery research, all the way to applications such as plant pathogen detection or GMO testing for labeling and QC purposes.

The PCR process includes several steps: denaturation where DNA strands are melted and separated, annealing where the primers anneal to template DNA, and extension where the enzyme works to elongate the DNA strands. These steps are carried out 20–35 times in a thermal cycler to produce many replicates of the DNA template. PCR is highly efficient and specific, generating over 10 million copies of target DNA from just a few molecules. Because the PCR process is so sensitive and specific, it is important to choose high-quality PCR products to produce optimal results.

PCR application guide

Cloning	Fast PCR	Genotyping	Sequencing and resequencing	Methylation
Application				
PCR is the preferred method to obtain a greater quantity of target DNA to clone. Downstream applications using high-purity cloned genes include: •cDNA library construction •Gene family characterization •Large-scale genome mapping using YACs/BACs	Fast PCR not only saves you time, it also enables you to do more research with your resources. Ideal applications include: •Standard PCR •High-throughput screening •Colony PCR library screening	Use PCR to study differences in DNA sequences. Related applications include: •Allele-specific PCR •Fragment length polymorphism analysis •Haplotyping •Linking emulsion PCR •SNP analysis •Microsatellite studies	PCR is used upstream of direct sequencing applications. Use direct sequencing for: •Mutation detection •Candidate gene analysis •Genetic linkage studies •Evolutionary studies •Genome (gap filling and shotgun) sequencing •Primer walking	Study epigenetic effects by amplification of bisulfite-treated DNA: •Specific amplification of long CpG islands •Difficult targets
Recommended enzymes				
•AmpliTaq Gold® 360 DNA Polymerase (or Master Mix) •Platinum® *Taq* DNA Polymerase High Fidelity •Platinum® *Taq* DNA Polymerase •AccuPrime™ *Taq* DNA Polymerase High Fidelity	•AmpliTaq Gold® Fast PCR Master Mix	•AmpliTaq Gold® Fast PCR Master Mix •AmpliTaq® and AmpliTaq Gold® 360 DNA Polymerases •Platinum® Multiplex PCR Master Mix •Platinum® GenoType *Tsp* DNA Polymerase	•AmpliTaq Gold® Fast PCR Master Mix •AmpliTaq® and AmpliTaq Gold® 360 DNA Polymerases •Platinum® *Taq* DNA Polymerase High Fidelity	•AmpliTaq Gold® 360 Master Mix and DNA Polymerase •AmpliTaq® 360 DNA Polymerase •Platinum® *Taq* DNA Polymerase
Compatible thermal cyclers				
•Veriti® Thermal Cyclers •GeneAmp® PCR System 9700 •2720 Thermal Cycler	•Veriti® 96-Well Fast Thermal Cycler (0.1 mL) •Veriti® 96-Well Thermal Cycler (0.2 mL)	•Veriti® Thermal Cyclers •GeneAmp® PCR System 9700 •2720 Thermal Cycler	•Veriti® Thermal Cyclers •GeneAmp® PCR System 9700 •2720 Thermal Cycler	•Veriti® Thermal Cyclers •GeneAmp® PCR System 9700 •2720 Thermal Cycler

At Life Technologies we continue to develop and support Applied Biosystems® PCR instruments and Invitrogen™ and Applied Biosystems® reagents to advance molecular biology research. Today we offer a variety of Applied Biosystems® thermal cyclers with features that suit a range of research needs and PCR applications. Optimized reagents and thermal cyclers enable researchers to speed up the PCR process, allowing results in just a third of the time. From leading-edge thermal cyclers to best-in-class enzymes to plates and tubes, you'll find everything you need to streamline your research.



The steps in a PCR cycle.

Bacterial gene amplification	Viral gene amplification	Long PCR (5–20 kb)	Multiplex PCR
Use PCR to analyze bacterial genomes—vital for antibiotic and vaccine development. Also useful for: •Pathogen detection •Amplified fragment length polymorphism (AFLP) analysis used in comparative bacterial genomics	PCR assays are a highly sensitive, specific, and fast means of detecting viruses for: •Nucleic acid sequence–based amplification (NASBA) •Pathogen detection	An enzyme blend efficiently amplifies larger targets for: •Sequence mapping •Mitochondrial genome PCR •Gene cluster studies •cDNA cloning of long transcripts	Use multiplex PCR to amplify many targets in a single tube. This method provides target-specific amplification with primers specific for each target. Used for: •Human identification studies •Variable number of tandem repeats (VNTR) screening
•AmpliTaq Gold® Fast PCR Master Mix •AmpliTaq Gold® 360 Master Mix and DNA Polymerase •AmpliTaq Gold® 360 Master Mix •Platinum® *Taq* DNA Polymerase	•AmpliTaq® and AmpliTaq Gold® 360 DNA Polymerases •Platinum® *Taq* DNA Polymerase	•Platinum® *Taq* DNA Polymerase High Fidelity •AccuPrime™ *Taq* DNA Polymerase High Fidelity •Elongase® Enzyme Mix	•Platinum® *Taq* DNA Polymerase High Fidelity •AmpliTaq Gold® 360 Master Mix and DNA Polymerase •Platinum® Multiplex PCR Master Mix
•Veriti® Thermal Cyclers •GeneAmp® PCR System 9700 •2720 Thermal Cycler	•Veriti® Thermal Cyclers •GeneAmp® PCR System 9700 •2720 Thermal Cycler	•Veriti® Thermal Cyclers •GeneAmp® PCR System 9700 •2720 Thermal Cycler	•Veriti® Thermal Cyclers •GeneAmp® PCR System 9700 •2720 Thermal Cycler

PCR instrument selection guide

Instrument model	Veriti® 96-Well System	Veriti® 384-Well System	Veriti® 60-Well System
Sample block	0.1 mL or 0.2 mL alloy blocks (non-interchangeable)	0.02 mL aluminum	0.5 mL aluminum
Function	PCR optimization, fast PCR, standard PCR	High-throughput 384-well isothermal sample block	60-well (0.5 mL) isothermal sample block for larger post-PCR sample volumes
Features	VeriFlex™ Blocks: six Peltier blocks for PCR optimization or to run up to six annealing temperatures in the same run	VeritiLink™ networking capability provides control of 12 instruments from a single instrument	Supports 0.5-mL thin-walled tubes, which enable better heat transfer and more efficient cycling
Temperature accuracy	±0.25°C (35°C–99.9°C)		
Temperature range	4°C to 99°C		
Dimensions	Height: 24.5 cm (9.6 in) Width: 23.7 cm (9.3 in) Depth: 48.5 cm (19.1 in)		
Cat. No.	4375305 (0.1 mL), 4375786 (0.2 mL)	4388444	4384638

Applied Biosystems® Veriti® Thermal Cycler

Designed to meet your current and future PCR needs

- Maximum flexibility in PCR optimization from six independently controlled temperature blocks*
- Reliability you expect
- Intuitive color touch screen for exceptionally easy use
- Get peace of mind through networking and email notifications with optional remote management software

VeriFlex™ blocks: better than gradient

On the 96 well Veriti® models, VeriFlex™ blocks allow you maximum flexibility in programming your PCR conditions. Six independently controlled blocks allow you to set unique temperatures for precise PCR optimization. In addition, the VeriFlex™ blocks can be used to run multiple PCR experiments that use different annealing temperatures and run under the same reaction conditions, allowing you to save time and maximize the use of your Veriti® Thermal Cycler.

Applied Biosystems® Veriti® Thermal Cycler.

* Available on Veriti® 96-Well Thermal Cyclers

	96-Well 9700	Dual 96-Well 9700	Dual 384-Well 9700	60-Well 9700	2720
	0.2 mL aluminum, silver, or gold-plated silver	0.2 mL aluminum (2 x 96-well)	0.02 mL aluminum (2 x 384-well)	0.5 mL aluminum	0.2 mL aluminum
	Most flexible research format	Medium-/high-throughput dual 96-well sample blocks enable 192 samples per run	High-throughput, small sample volume	Larger post-PCR sample volumes	96-well personalized cycler
	Standard 0.2 mL format and sample block options enable enhanced performance and durability	High-throughput dual 96-well sample blocks enable 192 samples per run	High-throughput dual 384-well sample blocks enable 768 samples per run, optional auto-lid	Supports 0.5-mL thin-walled tubes, which enable better heat transfer and more efficient cycling	Exceptionally small footprint designed with vents in the rear, allowing several cyclers to be placed side by side to conserve valuable bench space
				→	± 0.5°C (35°C–100°C)
				→	
	Height: 26 cm (10 in) Width: 30 cm (12 in) Depth: 40.6 cm (16 in)	Height: 26 cm (10 in) Width: 30 cm (12 in) Depth: 52 cm (20.5 in)	→	Height: 26 cm (10 in) Width: 30 cm (12 in) Depth: 40.6 cm (16 in)	Height: 22 cm (8.7 in) Width: 21 cm (8.3 in) Depth: 36 cm (14.2 in)
	4314879, N8050001, 4314878	4343176	N8050002, 4314487	4310899	4359659

Proven reliability

The Veriti® Thermal Cycler delivers the proven reliability you expect from an Applied Biosystems® thermal cycler.

Control at your fingertips

The 6.5-inch (16.51-cm), VGA color touch screen provides a powerful and intuitive, yet simple-to-operate, user interface on the Veriti® system. The large screen and navigation buttons allow easy programming of your temperature profiles. Setup and navigation of the Veriti® Thermal Cycler does not require the use of a stylus or mouse.

Fast setup, fast results

The Veriti® system has two different options for method navigation. For quick setup, you may select one of the pre-programmed methods, which provides advice based on the type of PCR or PCR reagent that you are using. The Veriti® Thermal Cycler also allows you to program your own methods and save them on the instrument or a USB drive.

Product	Quantity	Cat. No.
Applied Biosystems® Veriti® 96-Well Fast Thermal Cycler, 0.1 mL	1 instrument	4375305
Applied Biosystems® Veriti® 96-Well Thermal Cycler, 0.2 mL	1 instrument	4375786
Applied Biosystems® Veriti® 384-Well Thermal Cycler, 0.02 mL	1 instrument	4388444
Applied Biosystems® Veriti® 60-Well Thermal Cycler, 0.5 mL	1 instrument	4384638

Applied Biosystems® 2720 Thermal Cycler

Economical benchtop instrument

- Proven Applied Biosystems reliability and performance
- Compact design maximizes bench space
- Precise and uniform heating and cooling
- Convenient graphical interface
- Cost-effective

The personal-sized Applied Biosystems® 2720 Thermal Cycler is ideal for both basic PCR and cycle sequencing applications. It incorporates many features of the GeneAmp® PCR System 9700 to enable similar performance and reliability in a more compact package and at a lower price. The 2720 Thermal Cycler's exceptionally small footprint enables you to fit it almost anywhere. And because its cooling vents are in the back, you can safely place it in tight spaces to save valuable bench space.

Product	Quantity	Cat. No.
Applied Biosystems® 2720 Thermal Cycler	1 instrument	4359659

Applied Biosystems® 2720 Thermal Cycler.

Applied Biosystems® GeneAmp® PCR System 9700

High performance with built-in flexibility for wide-ranging needs

- Convenient graphical interface
- Interchangeable blocks available: 60-well, 0.5 mL; 96-well, 0.2 mL; and dual 96- or 384-well formats
- Optional robotics-compatible auto-lid available with dual 384-well block

The Applied Biosystems® GeneAmp® PCR System 9700 is a high-performance thermal cycler with built-in flexibility. The GeneAmp® PCR System 9700 consists of a base module and one of many interchangeable sample block modules that let you quickly change throughput and well volumes to match your applications. An intuitive graphical user interface with comprehensive programming features and real-time display makes your protocol setup fast and easy.

Applied Biosystems® GeneAmp® PCR System 9700.

GeneAmp® PCR System 9700 Configurations

96-Well GeneAmp® PCR System 9700

The 96-Well GeneAmp® PCR System 9700 is designed for use with 0.2-mL reaction tubes or 96-well reaction plates for all of your routine PCR applications. The 96-Well GeneAmp® PCR System 9700 can be used with gold-plated silver and aluminum sample blocks modules. The aluminum block has been designed for routine use of PCRs and cycle sequencing in a conventional 8 x 12-well format. The gold-plated silver sample block has been engineered for maximum performance and durability, utilizing a rapid heat transfer design of electroformed silver to maximize heating/cooling rates and gold-plating for maximum durability.

Dual 96-well GeneAmp® PCR System 9700

The Dual 96-well GeneAmp® PCR System 9700 is designed for use with 0.2-mL reaction tubes or 96-well reaction plates for high-throughput PCR and cycle sequencing applications. The Dual 96-Well GeneAmp® PCR System 9700 contains two sample well blocks; each holds a maximum of 96 samples. Fully loaded, the instrument accommodates up to 192 samples per run. The system can also be used with a single 96-well block for smaller runs.

Dual 384-Well GeneAmp® PCR System 9700 and Auto-Lid Dual 384-well GeneAmp® PCR System 9700

The Dual 384-Well GeneAmp® PCR System 9700 and Auto-Lid Dual 384-well GeneAmp® PCR System 9700 are designed for use with 384-well reaction plates for high-throughput PCR and cycle sequencing applications. The two 384-well aluminum sample blocks can run 768 reactions simultaneously. The Auto-Lid (automated heated lid) system opens and closes automatically and features a unique plate-ejection mechanism.

0.5-mL GeneAmp® PCR System 9700

The 0.5-mL GeneAmp® PCR System 9700 is designed specifically for high–sample volume PCR applications (both DNA and RNA). The 0.5-mL GeneAmp® PCR System 9700 is designed for use with 0.5-mL reaction tubes for all of your large-volume PCR applications.

Product	Quantity	Cat. No.
Gold 96-Well GeneAmp® PCR System 9700	Base module + sample block	4314878
Silver 96-Well GeneAmp® PCR System 9700	Base module + sample block	N8050001
Aluminum 96-Well GeneAmp® PCR System 9700	Base module + sample block	4314879
Dual 96-Well GeneAmp® PCR System 9700	Base module + sample block	4343176
Dual 384-Well GeneAmp® PCR System 9700	Base module + sample block	N8050002
Auto-Lid Dual 384-Well GeneAmp® PCR System 9700	Base module + sample block	4314487
0.5 mL GeneAmp® PCR System 9700	Base module + sample block	4310899

PCR enzyme selection chart

Product	Target size	Speed	Fidelity (vs. *Taq*)	Specificity	Yield	Amplify wide target range	Convenience
AmpliTaq Gold® 360 Master Mix	<3 kb	●●●	1x	●●	●●●●	●●●●●	●●●●●
AmpliTaq Gold® 360 DNA Polymerase	<3 kb	●●	1x	●●	●●●	●●●●	●●●●
Platinum® *Taq* DNA Polymerase	<5 kb	●●	1x	●●	●●●	●●●●	●●●●
Platinum® *Taq* DNA Polymerase, High Fidelity	<20 kb	●●	6x	●●	●●●●	●●●	●●●●
Platinum® Multiplex PCR Master Mix	50 bp–2.5 kb	●●	1x	●●●	●●●	●●	●●●●
AccuPrime™ *Taq* High Fidelity	<20 kb	●●	9x	●●●	●●●●	●●●	●●●●
AmpliTaq Gold® Fast PCR Master Mix, UP	<1.5 kb	●●●●●	1x	●●	●●●	●●●●	●●●●●
GeneAmp® Fast PCR Master Mix	<2 kb	●●●●●	1x	●●	●●●	●●●	●●●●●
AmpliTaq® 360 DNA Polymerase	<5 kb	●	1x	●	●	●●●●	●

Standard native and recombinant *Taq* Polymerase are also available. Please go www.invitrogen.com/pcr to get current pricing for these and all our PCR enzymes.

AmpliTaq Gold® Fast PCR Master Mix, UP

Fast PCR optimized for use in sequencing applications

- Fast results using existing primers to amplify standard 600-bp targets from genomic DNA in about 40 minutes
- Sensitive detection of up to 10 copies of a single gene in 10 ng of genomic DNA
- Easy to use and includes 2X master mix for quick experiment setup
- Better sequencing quality resulting in specific, high-yield amplicons suitable for sequencing applications that yield low peak-under-peak, longer continous read length, and higher QV-30 count

AmpliTaq Gold® Fast PCR Master Mix, UP helps reduce your time to results while delivering specific, high-yield amplicons, enabling easy production of high-quality sequencing data. When combined with modified cycle sequencing conditions for Fast cycle sequencing, you can go from sample to basecalling in approximately 4 hours. This premixed, hot-start master mix works with your existing primers to provide sensitive, specific, and reproducible results.

Product	Quantity	Cat. No.
AmpliTaq Gold® Fast PCR Master Mix, UP	25 reactions	4390937
	250 reactions	4390939
	2,500 reactions	4390941

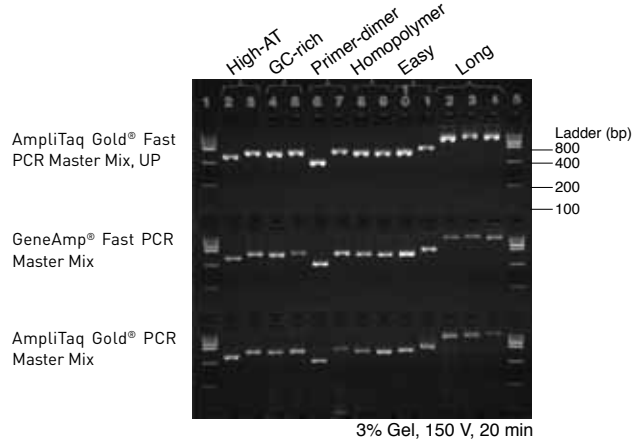
GeneAmp® Fast PCR Master Mix

Fast and reliable amplification typically in under 25 minutes

- Amplifies standard 500-bp targets from genomic DNA typically in under 25 minutes
- Detects single copy genes in 10 ng of human genomic DNA
- Premixed hot-start formulation enables you to set up experiments quickly

The Applied Biosystems® GeneAmp® Fast PCR Master Mix is specifically designed to amplify DNA quickly and robustly. The premixed GeneAmp® Fast PCR Master Mix's sensitive hot-start chemistries reduce PCR time to as little as 25 minutes.

Product	Quantity	Cat. No.
GeneAmp® Fast PCR Master Mix (2X) with protocol	250 reactions	4362070
GeneAmp® Fast PCR Master Mix (2X) without protocol	250 reactions	4359187



Robust results with Applied Biosystems® Fast PCR Master Mixes. Various target types, including high-AT, GC-rich, primer-dimer, homopolymer, easy, and long, were amplified on the Veriti® 96-Well Fast Thermal Cycler using a three-step protocol. Each amplicon was amplified from 10 ng of human genomic DNA. Amplification times for a 600-bp fragment varied as follows: 40 min for AmpliTaq Gold® Fast PCR Master Mix, UP, 25 min for GeneAmp® Fast PCR Master Mix, and 2 hr for the standard AmpliTaq Gold® PCR Master Mix.

AmpliTaq Gold® 360 DNA Polymerase and Master Mix

Hot-start amplification of a broad range of targets

- Increased specificity with trusted hot-start technology
- Amplify the broadest range of targets (<3 kb) that contain high GC, high AT, homopolymer runs, and more
- Increased yield compared with AmpliTaq Gold® DNA Polymerase
- Achieve excellent sequencing data from all amplicons
- Available in a master mix format for increased convenience and even better results

AmpliTaq Gold® 360 Polymerase was designed for 360° coverage of a full range of targets. It has been optimized to amplify a wide range of targets while delivering the high specificity and reliability you have come to expect from traditional AmpliTaq Gold® DNA Polymerase. AmpliTaq Gold® 360 DNA Polymerase provides the same hot-start specificity as AmpliTaq Gold® DNA Polymerase, is optimized for both easy and challenging targets, and provides high sensitivity and yield for a broad range of targets.

AmpliTaq Gold® 360 PCR Master Mix

AmpliTaq Gold® 360 PCR Master Mix contains everything required for successful PCR amplification in one convenient package, with all components premixed and premeasured. Supplied at 2X the recommended usage concentration for easy dilution when adding template and primers, the convenient AmpliTaq Gold® 360 PCR Master Mix scales to various reaction volumes for greater application and format flexibility. The main ingredient of the master mix is AmpliTaq Gold® 360 DNA Polymerase, but it also contains the world-class 360 GC Enhancer, which can be added optionally for high GC content templates. With wide template coverage, optimization of PCR conditions is virtually eliminated with the use of AmpliTaq Gold® 360 Master Mix.

Product	Quantity	Cat. No.
AmpliTaq Gold® 360 DNA Polymerase	100 units	4398813
	250 units	4398823
	1,000 units	4398833
	1,500 units	4398892
	2 x 1,500 units	4398894
	5 x 1,000 units	4398896
	25 x 1,000 units	4398898
AmpliTaq Gold® 360 Buffer, 25mM $MgCl_2$, and 360 GC Enhancer	1.5 mL	4398853
AmpliTaq Gold® 360 PCR Master Mix	1 mL	4398876
	5 mL	4398881
	10 x 5 mL	4398901
	1 x 50 mL	4398886

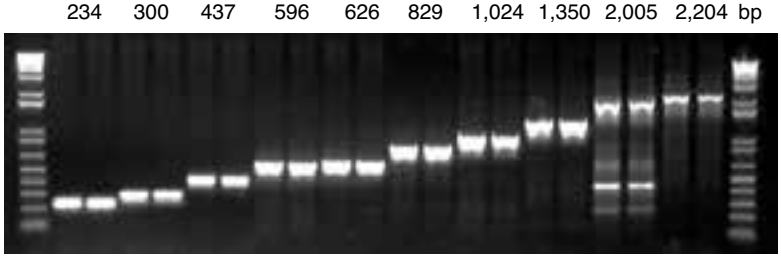
Standard AmpliTaq® 360 DNA Polymerase is also available. For more information about our standard (non-hot-start) *Taq* Polymerase products go to www.invitrogen.com/pcr.

Platinum® *Taq* DNA Polymerase and SuperMix

Hot-start amplification of longer targets

- Helps decrease time spent optimizing PCR reactions (e.g., [Mg^{2+}], annealing temperature, primer concentration)
- Increases yield of PCR product
- Allows for assembly of reactions at room temperature
- Less background and nonspecific PCR products, including reduced primer-dimer products
- Enables increased sensitivity so less target is required
- Available in convenient SuperMix format or with a blue loading dye for subsequent gel use (Platinum® Blue PCR SuperMix)

Platinum® *Taq* DNA Polymerase uses antibody-mediated hot start to enable increased specificity, yield, and sensitivity over standard *Taq* products. Platinum® *Taq* DNA Polymerase is derived from recombinant *Taq* DNA polymerase by binding of a thermolabile inhibitor containing monoclonal antibodies to *Taq* DNA polymerase. During the initial denaturation step of PCR, the inhibitor is denatured and active *Taq* DNA polymerase is released into the reaction. The result is improved specificity over *Taq* DNA polymerase.



Platinum® *Taq* PCR SuperMix provides high yield and specificity in PCR. Data shows effective amplification of increasing fragment lengths ranging from 234 to 2,204 bp.

Product	Quantity	Cat. No.
Platinum® *Taq* DNA Polymerase, 5 U/µL	100 reactions	10966018
	250 reactions	10966026
	500 reactions	10966034
	5,000 reactions	10966083
Platinum® PCR SuperMix, 1.1X concentration	100 reactions	11306016
Platinum® Blue PCR SuperMix, 1.1X concentration	100 reactions	12580015

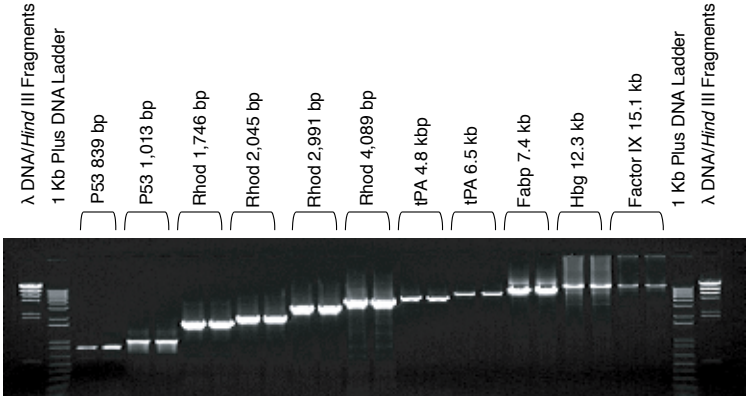
Platinum® *Taq* is supplied with 10X PCR buffer and $MgCl_2$. SuperMixes contain Platinum® *Taq*, dNTPs, magnesium, and buffer. Platinum® Blue SuperMix also contains blue tracking dye

Platinum® *Taq* DNA Polymerase High Fidelity

Robust PCR amplification with high yields

- Greater than six times higher fidelity than *Taq* DNA polymerase
- Amplification of fragments up to 20 kb
- Room temperature reaction assembly

Platinum® *Taq* DNA Polymerase High Fidelity and SuperMix are ideal for high-yield robust amplification of DNA from complex genomic, viral, and plasmid templates, and RT-PCR. High fidelity is provided by Platinum® *Taq* DNA Polymerase and the proofreading (3´–5´ exonuclease activity) enzyme *Pyrococcus* species *GB-D* polymerase. PCR specificity is improved with the incorporation of Platinum® automatic hot-start technology and is especially effective for high-fidelity applications such as cloning or mutagenesis.



Platinum® *Taq* DNA Polymerase High Fidelity easily amplifies a wide range of targets lengths from 839 bp to 15.1 kb.

Product	Quantity	Cat. No.
Platinum® *Taq* DNA Polymerase High Fidelity	100 reactions	11304011
	500 reactions	11304029
	5,000 reactions	11304102
Platinum® PCR SuperMix High Fidelity	100 reactions	12532016

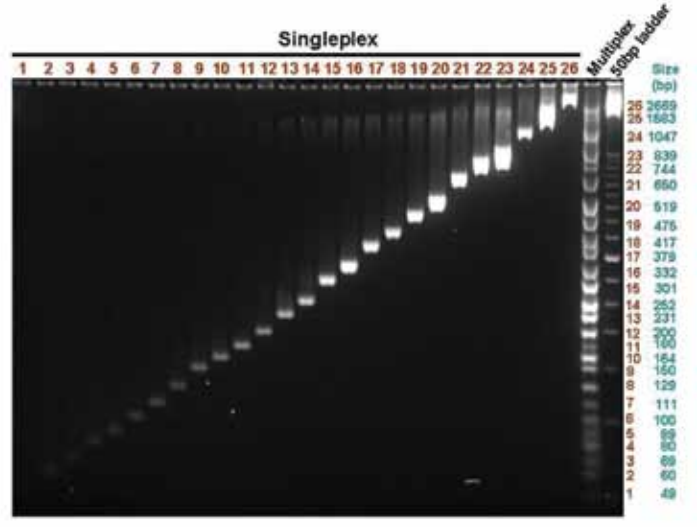
Platinum® *Taq* DNA Polymerase High Fidelity is supplied with 10X Buffer and $MgSO_4$. Platinum® PCR SuperMix High Fidelity provides premixed enzyme, dNTPs, salts, and $MgSO_4$.

Platinum® Multiplex PCR Master Mix

Optimization-free multiplex PCR master mix, designed specifically for end-point, multiplex PCR

- Convenient, optimization-free master mix
- Superior multiplex capability for amplification of up to 20 amplicons in a single reaction
- Wide amplicon size range enables amplification of products from 50 bp to 2.5 kb in length

The Platinum® Multiplex PCR Master Mix is an optimization-free multiplex PCR master mix with better specificity than currently available products. Designed specifically for end-point, multiplex PCR, it allows users to easily perform multiplexing with minimal optimization. Its proven performance for a wide range of amplicon sizes enables users to amplify products from 50 bp to 2.5 kb, greatly enhancing workflow flexibility. With its unparalleled 20-plex capabilities, Platinum® Multiplex PCR Master Mix not only provides a high-throughput solution, but also boasts better specificity through less non-specific primer binding events, thereby reducing reaction and primer waste.



Platinum® Multiplex PCR Master Mix specifically amplifies 26 targets in both a singleplex and multiplex fashion using the same universal PCR conditions. The products were resolved on a 4% agarose gel. Each lane was loaded with 10 µL of product from a single 50-µL PCR containing 100 ng of Jurkat genomic DNA template, 1X Platinum® Multiplex PCR Master Mix, and 100 nM each forward and reverse primers. Universal cycling conditions were used: enzyme activation at 95°C for 2 min, followed by 35 cycles of 95°C, 30 sec; 60°C, 1.5 min; and 72°C, 3 min.

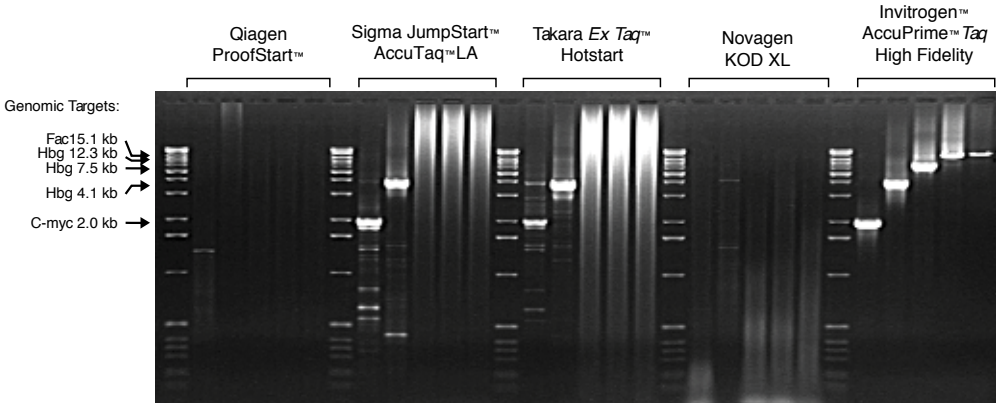
Product	Quantity	Cat. No.
Platinum® Multiplex PCR Master Mix	50 reactions	4464268
	250 reactions	4464269
	2,000 reactions	4464270

AccuPrime™ *Taq* DNA Polymerase High Fidelity

High-fidelity PCR with improved specificity and yield

- High specificity and yield for robust PCR amplification
- Up to nine times higher fidelity than *Taq* DNA polymerase alone
- Mimimal optimization steps, even with non-optimized primer sets

AccuPrime™ *Taq* DNA Polymerase High Fidelity amplifies nucleic acid templates up to 20 kb in length using an antibody-mediated hot start, a blend of *Taq* DNA Polymerase and proofreading enzyme, and AccuPrime™ accessory proteins for improved PCR fidelity, yield, and specificity over other hot-start DNA polymerases. High fidelity is achieved by a combination of Platinum® anti-*Taq* DNA polymerase antibodies that inhibit polymerase activity at room temperature, providing an automatic hot-start, and the proofreading (3´–5´ exonuclease activity) enzyme *Pyrococcus* species *GB-D*. The thermostable AccuPrime™ accessory proteins enhance specific primer-template hybridization during every cycle of PCR, preventing mispriming and enhancing PCR specificity and yield.



AccuPrime™ *Taq* DNA Polymerase High Fidelity outperforms other enzyme blends. Various targets were amplified from genomic DNA, as recommended by each manufacturer. Ten percent of each 50-µL PCR was loaded onto a 1% agarose gel.

Product	Quantity	Cat. No.
AccuPrime™ *Taq* DNA Polymerase High Fidelity	200 reactions	12346086
	1,000 reactions	12346094

AccuPrime™ *Taq* DNA Polymerase High Fidelity is supplied with $MgSO_4$ and two 10X reaction buffers containing dNTPs specialized for template type.

Platinum® *Pfx* DNA Polymerase

Very high-fidelity thermostable PCR enzyme

- Up to 26 times higher fidelity than *Taq* DNA polymerase
- Amplification of fragments up to 12 kb in length
- Room temperature reaction assembly

Platinum® *Pfx* DNA Polymerase is ideal for amplification of DNA fragments for high-fidelity PCR applications. High fidelity is provided by a proprietary enzyme preparation containing recombinant DNA Polymerase from *Thermococcus* species *KOD* with proofreading (3´–5´ exonuclease) activity. Platinum® antibody technology provides a simple, automatic hot-start method that improves PCR specificity. PCR_X Enhancer Solution is included for higher primer specificity, broader magnesium concentration, broader annealing temperature, and improved thermostability. The PCR_X Enhancer Solution also helps optimize PCR of problematic and/or GC-rich templates.

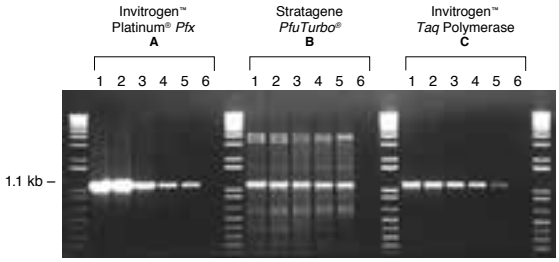


Figure 1. Platinum® *Pfx* DNA Polymerase provides higher yield and specificity than other enzymes. Genomic DNA (100, 50, 10, 5, 1, 0 ng, lanes 1–6, respectively) was amplified for 35 cycles with primers for human thrombospondin. (A) Reactions were set up at room temperature with 1 unit of Platinum® *Pfx* DNA Polymerase. (B) Reactions were set up on ice with 2.5 units of *PfuTurbo*® DNA Polymerase (Stratagene). (C) Reactions were set up at room temperature with 1 unit of *Taq* DNA Polymerase.

Product	Quantity	Cat. No.
Platinum® *Pfx* DNA Polymerase	100 reactions	11708013
	250 reactions	11708021
	500 reactions	11708039

Platinum® *Pfx* DNA Polymerase is supplied with 10X PCR_x Enhancer Solution, 10X buffer, and $MgSO_4$.

AccuPrime™ *Pfx* DNA Polymerase

Very high PCR fidelity with improved yield and specificity

- Up to 26 times higher fidelity than *Taq* DNA polymerase
- Minimal PCR optimization
- Available as a SuperMix

AccuPrime™ *Pfx* DNA Polymerase is ideal for high-fidelity amplification of DNA fragments up to 12 kb in length for downstream applications such as cloning and mutagenesis. High fidelity is provided by a proprietary enzyme preparation containing recombinant DNA polymerase from *Thermococcus* species *KOD* with proofreading (3´–5´ exonuclease) activity. Platinum® anti-*Pfx* DNA polymerase antibodies inhibit polymerase activity at room temperature, providing hot-start capabilities for improved PCR specificity. Thermostable AccuPrime™ accessory proteins enhance specific primer-template hybridization during every cycle of PCR, increasing specificity, yield, and robustness over Platinum® *Pfx* alone.

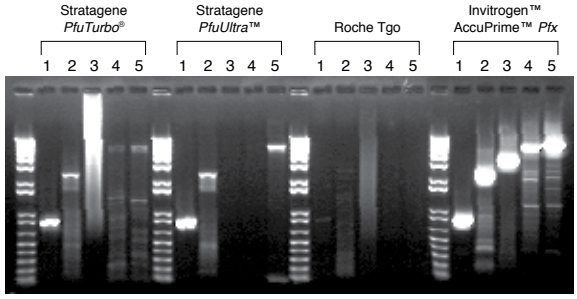


Figure 2. AccuPrime™ *Pfx* DNA Polymerase delivers higher yield and specificity than other proofreading enzymes. Each 50-µL reaction was set up according to the enzyme manufacturers' recommendations. Twenty percent of each reaction was analyzed on a 0.8% agarose gel.

Product	Quantity	Cat. No.
AccuPrime™ *Pfx* DNA Polymerase	200 reactions	12344024
	1,000 reactions	12344032
AccuPrime™ *Pfx* SuperMix	200 reactions	12344040

AccuPrime™ *Pfx* DNA Polymerase is provided with 10X buffer that contains dNTPs and a separate vial of $MgSO_4$. AccuPrime™ *Pfx* SuperMix is supplied in a 1.1X concentration and contains polymerase, salts, magnesium, and dNTPs.

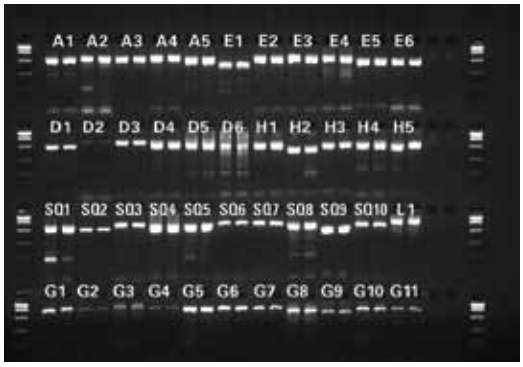
AmpliTaq® 360 DNA Polymerase

For reliable amplification of a broad range of targets

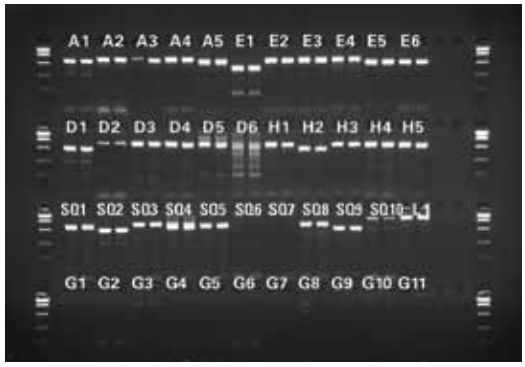
- Reliably amplify a broad range of targets that contain high GC, high AT, homopolymer runs, and more
- Optional 360 GC Enhancer allows you to tackle the most challenging GC-rich fragments with ease
- Superior yield allows you to do more with your PCR amplicons
- Enables fewer false positives and reduced bacterial background
- Obtain reproducible results from the most trusted source for PCR
- Enables superior sequencing results even with difficult targets

AmpliTaq® 360 DNA Polymerase has been optimized to amplify a wide range of targets while delivering the reliability you have come to expect from traditional AmpliTaq® DNA Polymerase. When used with the new enhanced AmpliTaq® 360 Buffer and the optional 360 GC Enhancer, AmpliTaq® 360 DNA Polymerase amplifies a vast range of DNA sequence contexts. Compared to the original AmpliTaq® DNA Polymerase, AmpliTaq® 360 Polymerase is purified by an additional proprietary separation process to minimize contaminating bacterial DNA sequences from the enzyme preparation. This ultra-pure enzyme helps reduce false positive results, amplifies low-level target sequences, and when combined with the proprietary AmpliTaq® 360 Buffer Kit, promotes the amplification of a variety of templates, including those from bacterial and human genomes.

Panel A. AmpliTaq® 360 DNA Polymerase



Panel B. Promega GoTaq® Green DNA Polymerase



AmpliTaq® 360 DNA Polymerase amplifies a broader range of targets than Promega GoTaq® Green DNA Polymerase. The same targets were amplified with (A) AmpliTaq® 360 DNA Polymerase and (B) Promega GoTaq® DNA Polymerase. PCR was performed using 1 ng of template DNA and 1.25 units of enzyme in each 50-µL reaction. Annealing was uniform across the selected targets. Amplicons ranged from 300 to 1,400 bp in length, with an average length of 553 bp. Each reaction was performed in duplicate. Amplicons are labeled as follows: E = easy amplification; A = high-AT; G = high-GC; D = primer-dimer; H = homopolymer; SQ = sequencing challenge. The high-GC target (G) reactions included 2–10 µL of 360 GC Enhancer, depending upon the target used.

Product	Quantity	Cat. No.
AmpliTaq® 360 DNA Polymerase	100 units	4398808
	250 units	4398818
	1,000 units	4398828
	1,500 units	4398891
	3,000 units	4398893
	5,000 units	4398895
	25,000 units	4398897

Plastic consumables compatibility chart for Applied Biosystems® instruments.

		End-point PCR systems							
Category/part description	Cat. No.	Veriti® 96-Well 0.1 mL	Veriti® 96-Well 0.2 mL	Veriti® 384-Well	60-Well Veriti®/9700 (60-well)	2720	6100	9700 (96-well)	9700 (384-well)
96-well plates									
MicroAmp® Fast Optical 96-Well Reaction Plate (0.1 mL)—10 plates	4346907	●							
MicroAmp® Fast Optical 96-Well Reaction Plate with Barcode (0.1 mL)—20 plates	4346906	●							
MicroAmp® Fast Optical 96-Well Reaction Plate with Barcode (0.1 mL)—200 plates	4366932	●							
MicroAmp® Optical 96-Well Reaction Plate—10 plates	N8010560		●			●	●	●	
MicroAmp® Optical 96-Well Reaction Plate—500 plates	4316813		●			●	●	●	
MicroAmp® Optical 96-Well Reaction Plate with Barcode—20 plates	4306737		●			●	●	●	
MicroAmp® Optical 96-Well Reaction Plate with Barcode—500 plates	4326659		●			●	●	●	
MicroAmp® Optical 96-Well Reaction Plate with Barcode and Optical Caps—20 plates	403012		●			●		●	
MicroAmp® Optical 96-Well Reaction Plate with Barcode and Optical Adhesive Films—100 plates	4314320		●					●	
384-well plates									
MicroAmp® Optical 384-Well Reaction Plate with Barcode—50 plates	4309849			●					●
MicroAmp® Optical 384-Well Reaction Plate with Barcode—500 plates	4326270			●					●
MicroAmp® Optical 384-Well Reaction Plate with Barcode—1,000 plates	4343814			●					●
MicroAmp® Optical 384-Well Reaction Plate—1,000 plates	4343370			●					●
48-well plates									
MicroAmp® Fast Optical 48-Well Reaction Plate—20 plates	4375816	●							
Single tubes									
MicroAmp® Fast Reaction Tube with Cap (0.1mL)—1,000 tubes	4358297	●							
MicroAmp® Reaction Tube with Cap (0.2 mL)—1,000 tubes	N8010540		●			●		●	
MicroAmp® Reaction Tube with Cap (0.2 mL)—10,000 tubes	N8011540		●			●		●	
MicroAmp® Reaction Tube with Cap, Assorted Colors (0.2 mL)—1,000 tubes	N8010840		●			●		●	
MicroAmp® Reaction Tube with Cap, Autoclaved (0.2 mL)—1,000 tubes	N8010612		●			●		●	
MicroAmp® Reaction Tube without Cap (0.2 mL)—2,000 tubes	N8010533		●			●		●	
MicroAmp® Reaction Tube without Cap (0.2 mL)—10,000 tubes	N8011533		●			●		●	
MicroAmp® Reaction Tube without Cap, Assorted Colors (0.2 mL)—1,000 tubes	N8010833		●			●		●	
MicroAmp® Optical Tube without Cap (0.2mL)—2,000 tubes	N8010933		●			●		●	
GeneAmp® Thin-Walled Reaction Tube with Flat Cap (0.5 mL)—1,000 tubes	N8010737				●				
GeneAmp® Thin-Walled Reaction Tube with Domed Cap (0.5 mL)—2,000 tubes	N8010537				●				
GeneAmp® Thin-Walled Reaction Tube with Domed Cap, Autoclaved (0.5 mL)—1,000 tubes	N8010611				●				

	Real-Time PCR systems								Genetic analyzers	
	StepOne™	StepOne-Plus™	7000	7300/ 7500	7500 Fast	7900HT (96-well)	7900HT (384-well)	7900HT Fast	310*	3100/3130 3700/3730
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Continued on next page.

Plastic consumables compatibility chart for Applied Biosystems® instruments, cont.

Category/part description	Cat. No.	End-point PCR systems							
		Veriti® 96-Well 0.1 mL	Veriti® 96-Well 0.2 mL	Veriti® 384-Well	60-Well Veriti®/9700 (60-well)	2720	6100	9700 (96-well)	9700 (384-well)
MicroAmp® 96-Well Reaction Tube/Tray/ Retainer Set (0.2 mL)—20 assemblies	403083					●		●	
MicroAmp® 96-Well Reaction Tube/Tray/ Retainer Set (0.2 mL)—100 assemblies	403086					●		●	
Strip-tubes and caps									
MicroAmp® 12-Cap Strip—200 strips	N8010534	●	●			●		●	
MicroAmp® 12-Cap Strip—1,000 strips	N8011534	●	●			●		●	
MicroAmp® 12-Cap Strip, Assorted Colors—200 strips	N8010834	●	●			●		●	
MicroAmp® Fast 8-Tube Strip (0.1 mL)—125 strips	4358293	●							
MicroAmp® 8-Tube Strip (0.2 mL)—125 strips	N8010580		●			●		●	
MicroAmp® 8-Tube Strip, Assorted Colors (0.2 mL)—120 strips	N8010838		●			●		●	
MicroAmp® Optical 8-Tube Strip (0.2 mL)—125 strips	4316567		●			●		●	
MicroAmp® 8-Cap Strip—300 strips	N8010535	●	●			●		●	
MicroAmp® 8-Cap Strip—1,500 strips	N8011535	●	●			●		●	
MicroAmp® 8-Cap Strip, Assorted Colors—300 strips	N8010835	●	●			●		●	
MicroAmp® Optical 8-Cap Strip—300 strips	4323032	●	●			●		●	
Seals and covers									
MicroAmp® Clear Adhesive Film—100 films	4306311	●	●	●				●	●
MicroAmp® Optical Adhesive Film—25 films	4360954	●	●	●				●	●
MicroAmp® Optical Adhesive Film—100 films	4311971	●	●	●				●	●
MicroAmp® 48-Well Optical Adhesive Film—100 films	4375323	●							
MicroAmp® 48-Well Optical Adhesive Film—25 films	4375928	●							
MicroAmp® Optical Adhesive Film Kit—20 kits	4313663							●	●
MicroAmp® 96-Well Full Plate Cover—5 covers	N8010550							●	
Reaction trays									
MicroAmp® 96-Well Tray/Retainer Set—10 assemblies	403081					●		●	
MicroAmp® 96-Well Tray—10 trays	N8010541					●		●	
MicroAmp® 96-Well Tray for VeriFlex™ Blocks—10 trays	4379983	●	●						
MicroAmp® 96-Well Tray/Retainer Set for Veriti® Systems—10 assemblies	4381850		●						
MicroAmp® Fast 48-Well Tray—10 trays	4375282								
Compression pads									
MicroAmp® Optical Film Compression Pad—5 pads	4312639								
MicroAmp® Snap-On Optical Film Compression Pad—9 pads	4333292								

	Real-time PCR systems								Genetic analyzers	
	StepOne™	StepOne-Plus™	7000	7300/ 7500	7500 Fast	7900HT (96-well)	7900HT (384-well)	7900HT Fast	310*	3100/3130 3700/3730
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96-well reaction plates

- Polypropylene, autoclavable, and nonsterile
- Frosted plastic minimizes interfering fluorescence from cycling block
- Optimized for well-to-well temperature uniformity
- Optimized for use with Applied Biosystems® end-point and real-time PCR systems

MicroAmp® 96-Well Reaction Plates are designed for automated or high-throughput PCR applications. All optical reaction plates are quality controlled to minimize fluorescent background. For fluorescent applications, the 96-well plates require MicroAmp® Optical Caps or Optical Adhesive Films. For nonfluorescent applications, MicroAmp® Caps, Full Plate Covers, or Clear Adhesive Films can be used.

Plate capacity

- MicroAmp® Fast 96-Well Plate capacity—maximum reaction volume is 30 µL, and maximum fill volume is 100 µL
- MicroAmp® 96-Well Plate capacity—maximum reaction volume is 100 µL, and maximum fill volume is 200 µL
- Also available in individual tubes formatted into a 96-well retainer/tray assembly

Product	Quantity	Cat. No.
MicroAmp® Fast 96-Well Reaction Plate (0.1 mL)	10 plates	4346907
MicroAmp® Fast Optical 96-Well Reaction Plate with Barcode (0.1 mL)	20 plates	4346906
	200 plates	4366932
MicroAmp® Optical 96-Well Reaction Plate	10 plates	N8010560
	500 plates	4316813
MicroAmp® Optical 96-Well Reaction Plate with Barcode	20 plates	4306737
	500 plates	4326659
MicroAmp® Optical 96-Well Reaction Plate with Barcode and Optical Caps	20 plates, 300 strips (8 caps/strip)	403012
MicroAmp® Optical 96-Well Reaction Plate with Barcode and Optical Adhesive Films	100 plates, 100 films	4314320
MicroAmp® 96-Well Reaction Tube/Tray/Retainer Set (0.2 mL)	20 sets (96 tubes/set)	403083
	100 sets (96 tubes/set)	403086
MicroAmp® Fast Optical 48-Well Reaction Plate	20 plates	4375816

Figure 1. MicroAmp® Fast 96-Well Reaction Plate.

Figure 2. MicroAmp® Fast Optical 96-Well Reaction Plate with Barcode.

Figure 3. MicroAmp® Optical 96-Well Reaction Plate with Barcode.

Figure 4. MicroAmp® 96-Well Reaction Tube/Tray/Retainer Set.

384-well reaction plates

- Polypropylene, autoclavable, and nonsterile
- Best plate for real-time PCR
- Best plate for high-throughput PCR

MicroAmp® 384-Well Clear Optical Reaction Plates are engineered for use with the Dual 384-Well GeneAmp® PCR System 9700, the Veriti® 384-Well Thermal Cycler, the 7900HT Real-Time PCR System, and the 3130/3730 Genetic Analyzer. For fluorescent applications, the 384-well plates require MicroAmp® Optical Adhesive Films. For nonfluorescent applications, MicroAmp® Clear Adhesive Films can be used.

Plate capacity

- MicroAmp® Optical 384-Well Plate capacity—maximum reaction volume is 20 µL

Product	Quantity	Cat. No.
MicroAmp® Optical 384-Well Reaction Plate with Barcode	50 plates	4309849
	500 plates	4326270
	1,000 plates	4343814
MicroAmp® Optical 384-Well Reaction Plate	1,000 plates	4343370

MicroAmp® Optical 384-Well Reaction Plate with Barcode.

Reaction tubes and strips

- Available in regular or optical caps for real-time PCR
- Available in 0.1-mL fast tubes and strips
- Captive lid with positive-click closure for proper seating
- Polished surface and conical bottom for maximum sample recovery
- Option of autoclavable tubes allows clean, controlled start

Reaction tubes

MicroAmp® and GeneAmp® Reaction Tubes are essential for high performance PCR. They are optimized to provide accurate and uniform sample temperatures for fast, oil-free PCR amplification using our GeneAmp® PCR System 9700s, 2700, 2720, Veriti®, Veriti® Fast, StepOne™, and StepOnePlus™ Real-Time PCR systems. For the 0.1-mL MicroAmp® Fast PCR Reaction Tubes, the recommended reactions volumes are 10–30 µL. For the 0.2-mL MicroAmp® Reaction Tubes, the recommended maximum volume for PCR amplification is 100 µL. For larger PCR volumes, try our 0.5-mL reaction tubes. Tubes are available with or without caps.

GeneAmp® Reaction Tubes are made of specially engineered polypropylene and optimized for use in the 0.5-mL GeneAmp® PCR System 9700 and on the Veriti® 60-Well Thermal Cycler. The thin-walled tubes facilitate rapid heat transfer to and from samples to achieve faster cycle times and increased specificity. All GeneAmp® Reaction Tubes undergo stringent quality testing to enable reproducible results.

MicroAmp® 96-Well Tube/Tray/Retainer Assemblies

For maximum convenience, the assemblies contain MicroAmp® Reaction Tubes without Caps loaded in MicroAmp® Tray/Retainer Sets. Designed for the GeneAmp® PCR System 9700 and 2700/2720, the sets require MicroAmp® Caps or Full Plate Covers. These assemblies are nonsterile and autoclavable.

Reaction tube strips

These specially engineered one-piece strips consist of eight 0.2-mL MicroAmp® Reaction Tubes joined near the tube opening. The strips precisely fit the standard well spacing of the 96-well sample blocks and require either MicroAmp® Caps or Full Plate Covers, sold separately. The strips are also available in clear and colored sets of red, orange, blue, and green and are packaged in separate bags of 30 strips each. Optical reaction strips should be used for real-time PCR applications. The Fast 8-Tube Strip is available for the Veriti® 96-Well Fast Thermal Cycler, 9800 Fast Thermal Cycler, StepOne™ Real-Time PCR System, and StepOnePlus™ Real-Time PCR System.

Cap strips

MicroAmp® Caps can be placed on MicroAmp® Reaction Tubes. They are also compatible with MicroAmp® 96-Well Reaction Plates and are available in 8-cap and 12-cap strips in clear and colored sets of red, orange, blue, and green. MicroAmp® Optical Caps should be used for real-time PCR applications.

Product	Quantity	Cat. No.
Reaction tubes		
MicroAmp® Fast Reaction Tube with Cap (0.1 mL) Requires, but does not include, Fast Retainer Tray Cat. No. 4358305.	1,000 tubes	4358297
MicroAmp® Reaction Tube with Cap (0.2 mL)	1,000 tubes	N8010540
	10,000 tubes	N8011540
MicroAmp® Reaction Tube with Cap, Assorted Colors (0.2 mL)	1,000 tubes	N8010840
MicroAmp® Reaction Tube with Cap, Autoclaved (0.2 mL)	1,000 tubes	N8010612
GeneAmp® Thin-Walled Reaction Tube with Flat Cap (0.5 mL) For use on the 0.5-mL GeneAmp® PCR System 9700 and Veriti® 60-Well Thermal Cycler.	1,000 tubes	N8010737
GeneAmp® Thin-Walled Reaction Tube with Domed Cap, Autoclaved (0.5 mL)	1,000 tubes	N8010611
GeneAmp® Thin-Walled Reaction Tube with Domed Cap (0.5 mL)	2,000 tubes	N8010537
MicroAmp® Reaction Tube without Cap (0.2 mL)	2,000 tubes	N8010533
	10,000 tubes	N8011533
MicroAmp® Reaction Tube without Cap, Assorted Colors (0.2 mL)	1,000 tubes	N8010833
MicroAmp® Optical Tube without Cap (0.2 mL) Requires, but does not include, the proper MicroAmp® Tray/Retainer Sets.	2,000 tubes	N8010933
MicroAmp® 96-Well Reaction Tube/Tray/Retainer Set (0.2 mL) Requires, but does not include, MicroAmp® Caps or Full Plate Cover.	20 assemblies	403083
	100 assemblies	403086
Reaction tube strips		
MicroAmp® 8-Tube Strip (0.2 mL)	125 strips	N8010580
MicroAmp® 8-Tube Strip, Assorted Colors (0.2 mL)	120 strips	N8010838
MicroAmp® Optical 8-Tube Strip (0.2 mL)	125 strips	4316567
MicroAmp® Fast 8-Tube Strip (0.1 mL)	125 strips	4358293
Reaction cap strips		
MicroAmp® 8-Cap Strip	300 strips	N8010535
	1,500 strips	N8011535
MicroAmp® 8-Cap Strip, Assorted Colors	300 strips	N8010835
MicroAmp® 12-Cap Strip	200 strips	8010534
	1,000 strips	8011534
MicroAmp® 12-Cap Strip, Assorted Colors	200 strips	N8010834
MicroAmp® Optical 8-Cap Strip	300 strips	323032

Overview of reverse transcriptases—uncover what you've been missing

SuperScript® III gene expression tools provide higher yields of full-length cDNA for more complete gene representation compared to standard reverse transcriptases. Robust cDNA synthesis is a critical component of gene expression research. Suboptimal reverse transcription (RT) reactions can lead to low yields of cDNA that compromise sensitivity, or truncated cDNA that can produce false negative results. Most reverse transcription enzymes fail to consistently produce cDNA with full gene representation due to being performed at suboptimal temperatures (less than 50°C). The SuperScript® family of reverse transcriptases is the most widely used brand of RT enzymes, due to its proven ability to deliver reliable and consistent first-strand synthesis. To meet the need for increased sensitivity, SuperScript® III Reverse Transcriptase was developed to provide improved performance in a variety of gene expression applications. SuperScript® III Reverse Transcriptase has been engineered for higher thermostability and a longer half-life at 50°C. This increases its ability to process RNA with secondary structure. In addition, like the SuperScript® II enzyme, SuperScript® III Reverse Transcriptase also exhibits reduced RNase H activity, allowing a greater production of full-length cDNA for more complete gene representation. These attributes also result in increased sensitivity for qRT-PCR and microarray experiments.

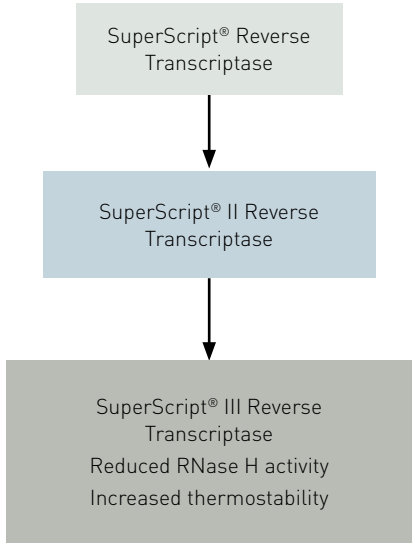


Figure 1. Performance-based evolution of SuperScript® III Reverse Transcriptase. To meet the growing need for sensitivity, we developed SuperScript® III Reverse Transcriptase, the latest-generation RT enzyme that provides improved performance in a variety of gene expression applications.

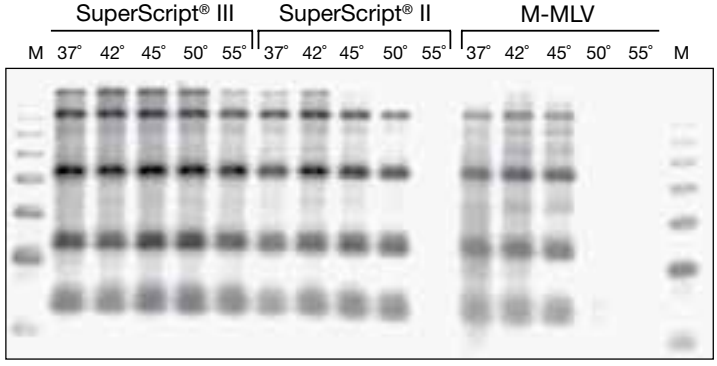


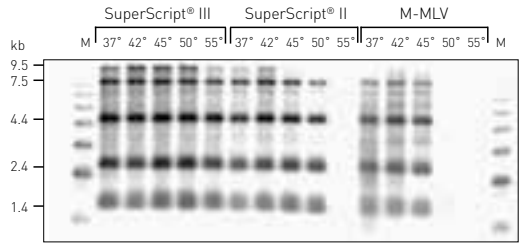
Figure 2. SuperScript® III Reverse Transcriptase generates the highest yields of full-length cDNA. The autoradiograph shows ^{32}P-labeled cDNA synthesized from 0.25 µg of mixed RNA, consisting of 1.35, 2.4, 4.4, 7.5, and 9.5 kb fragments, with 200 units of each enzyme at various temperatures (°C). Lane M: ^{32}P-labeled 1 Kb DNA Ladder.

SuperScript® III Reverse Transcriptase

Engineered for higher thermostability, increased half-life, and reduced RNase H activity

- Half-life of 220 minutes at 50°C for high cDNA yields
- Reduced RNase H activity for more full-length cDNA
- Full activity at 50°C for increased specificity with gene specific primers (GSPs)
- Ability to increase RT units without inhibiting subsequent PCR

SuperScript® III Reverse Transcriptase (RT) is a proprietary mutant of SuperScript® II RT that is active at 50°C and has a half-life of 220 minutes, providing increased specificity with gene specific primers and high cDNA yield. Like SuperScript® II RT, it synthesizes a complementary DNA strand from single-stranded RNA, DNA, or an RNA:DNA hybrid. SuperScript® III RT can be used for RT-PCR of a specific gene, or for generating cDNA from total or poly (A)+ RNA samples. It is ideal for synthesis of first-strand cDNA, array labeling, cDNA libraries, RT-PCR, primer extension, and 3´ and 5´ RACE.



Comparison of SuperScript® III Reverse Transcriptase to SuperScript® II RT and M-MLV RT. An autoradiograph is shown of ^{32}P-labeled cDNA synthesized from a mixture of 0.25 µg of RNA each of 1.35 kb, 2.4 kb, 4.4 kb, 7.5 kb, and 9.5 kb with 200 units of each RT at various temperatures. Lane M is ^{32}P-labeled 1 kb DNA ladder.

Product	Quantity	Cat. No.
SuperScript® III Reverse Transcriptase	10 reactions	18080-093
	50 reactions	18080-044
	200 reactions	18080-085

SuperScript® III First-Strand Synthesis System

High cDNA yields in a convenient kit format

- Contains SuperScript® III Reverse Transcriptase for higher cDNA yields
- Detects template molecules from as little as 1.0 pg total RNA
- Includes all components needed for first-strand cDNA synthesis
- Consists of optimized reagents for more robust performance

The SuperScript® III First-Strand Synthesis System for RT-PCR provides all the necessary components for synthesis of high-quality, first-strand cDNA from total or poly(A)+ RNA. Used with PCR, this functionally tested system enables you to detect rare messages and generate enough material for cloning.

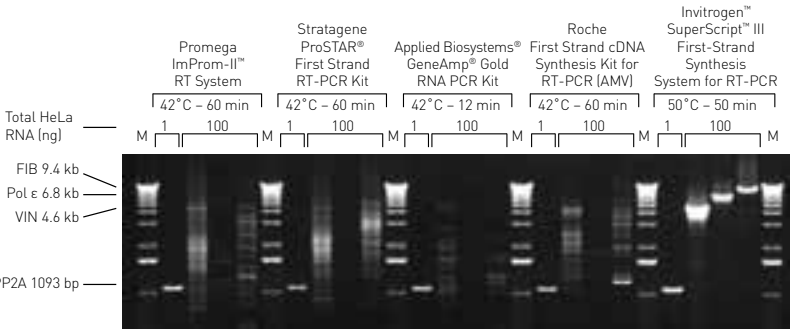


Figure 2. The SuperScript™ III First-Strand Synthesis System outperforms the competition. RT reactions containing 1 and 100 ng of total HeLa RNA were performed with each kit using reagents and conditions specified in each manufacturer's protocol. Ten percent (2-5 µL) of the resulting cDNA was added to PCR reactions containing 1 unit of Platinum® *Taq* DNA Polymerase High Fidelity for 35 PCR cycles, 1 min/kb. PCR products were separated on a 1% agarose gel containing 0.4 µg/mL ethidium bromide.

1_2.tiff

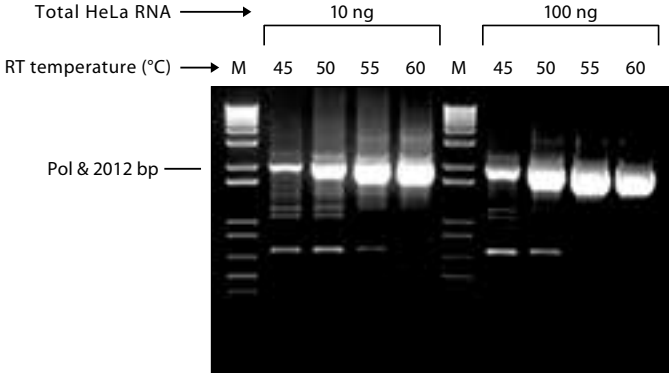
Product	Quantity	Cat. No.
SuperScript® III First-Strand Synthesis System	50 reactions	18080-051

SuperScript® III One-Step RT-PCR System with Platinum® *Taq* DNA Polymerase

High sensitivity one-step RT-PCR for end point detection

- Contains SuperScript® III Reverse Transcriptase (RT) for higher cDNA yields and increased thermostability
- Enables routine detection down to 0.01 pg total RNA
- Detects targets up to 4.5 kb in length for greater flexibility
- One-step format for speed, convenience, and less variability from reaction to reaction

The SuperScript® III One-Step RT-PCR System with Platinum® *Taq* DNA Polymerase combines the high thermostability of SuperScript® III RT with the specificity of Platinum® *Taq* DNA Polymerase to increase priming specificity, generate higher product yields, and identify a larger range of targets. The one-step format enables easy analysis of gene expression or detection of rare or viral RNA.



SuperScript® III One-Step RT-PCR System with Platinum® *Taq* provides increased specificity with gene specific primers. One-Step RT-PCR reactions were performed with 10 and 100 ng of total HeLa RNA at the temperatures indicated using the SuperScript® III One-Step RT-PCR System with Platinum® *Taq* DNA Polymerase. Reactions were amplified with 40 cycles of PCR, 1 min/kb.

Platinum® *Taq* provides increased specificity with gene specific primers. One-step RT-PCR reactions were performed with 10 and 100 ng of total HeLa RNA at the temperatures indicated using the SuperScript® III One-Step RT-PCR System with Platinum® *Taq* DNA Polymerase. Reactions were amplified with 40 cycles of PCR, 1 min/kb.

Product	Quantity	Cat. No.
SuperScript® III One-Step RT-PCR System with Platinum® *Taq* DNA Polymerase	25 reactions	12574-018
	100 reactions	12574-026
SuperScript® III One-Step RT-PCR System with Platinum® *Taq* High Fidelity	25 reactions	12574-030
	100 reactions	12574-035

The SuperScript® One-Step RT-PCR System with Platinum® *Taq* DNA Polymerase kits include SuperScript® RT/Platinum® *Taq* Mix, reaction mix (includes dNTPs), and $MgSO_4$.

For a complete listing of RT-PCR products, go to www.invitrogen.com/pcr.

SuperScript® VILO™ cDNA Synthesis Kit

- Greater accuracy—reduce bias generated from low- or high-abundance genes
- Consistent results—obtain reliable analysis of more genes
- Reduced qPCR inhibition—use greater amounts of cDNA in qPCR to increase sensitivity up to 4-fold without fear of inhibiting the reaction
- Increased yield—archive cDNA for future studies

The SuperScript® VILO™ cDNA Synthesis Kit is designed to increase cDNA yields and improve the dynamic range of your qRT-PCR assays. This means that you obtain the same relative representation in your cDNA, irrespective of the abundance level of your gene of interest.

Reduce bias in reverse transcriptase reactions

Using the SuperScript® VILO™ cDNA Synthesis Kit enables you to obtain consistent and accurate results with unprecedented linearity across the broadest range of input material (see figure). Regardless of the abundance of your gene of interest in the starting material, you can expect the same relative representation in the cDNA template that is generated. Because the bias toward abundant genes is reduced, data derived from comparing the gene of interest and the reference genes are more accurate.

Reliable analysis of more genes

The SuperScript® VILO™ cDNA Synthesis Kit, containing SuperScript® III Reverse Transcriptase and a proprietary helper protein, minimizes carryover of PCR inhibitors, which allows you to add greater amounts of cDNA to the qRT-PCR experiments without inhibiting downstream steps. This enables the analysis of both low- and high-abundance genes, regardless of their expression level or the amount of RNA used in the reaction.

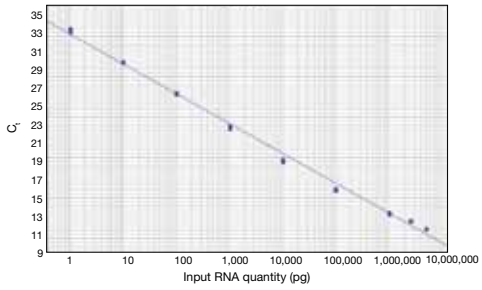
Detection flexibility

Get the most sensitive qPCR performance with the SuperScript® VILO™ cDNA Synthesis Kit regardless of detection method. Using either TaqMan®- or SYBR®- based real-time PCR detection, you can expect up to 8-fold sensitivity increases compared to other available kits. SuperScript® VILO™ cDNA Synthesis Kits are designed to work with Applied Biosystems® fast-cycling, real-time PCR instruments.

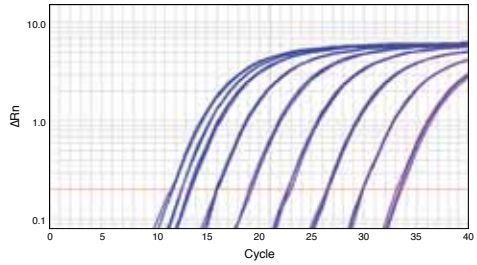
Archive cDNA for future studies

Including SuperScript® III Reverse Transcriptase, RNaseOUT™ Recombinant Ribonuclease Inhibitor, and a proprietary helper protein, the SuperScript® VILO™ cDNA Synthesis Kit generates far greater yields of cDNA. The extra cDNA product can be archived for subsequent applications, and you also have the option of increasing the amount of cDNA in your experiment for improved sensitivity (up to 4-fold) without fear of inhibiting the reaction.

A. Standard curve.



B. Amplification plot.



SuperScript® VILO™ cDNA Synthesis Kit provides reliable performance across an extended linear range of RNA input. A serial dilution of total RNA from HeLa cells was reverse transcribed using the SuperScript® VILO™ cDNA Synthesis Kit followed by qPCR reactions using human ß-actin Applied Biosystems® TaqMan® Assays with Invitrogen™ EXPRESS qPCR Supermix Universal on an ABI PRISM® 7700. The standard curve (A) shows that even outside the recommended input of 1 pg to 2.5 µg, the kit exhibits a coefficient of correlation of 0.996 for up to 5 µg of input RNA. The amplification plot (B) illustrates that the triplicates are aligned across all dilutions, demonstrating the robustness of the SuperScript® VILO™ cDNA Synthesis Kit over a broad linear range of RNA input. Normalize your lower-abundance genes to your reference genes without worrying about potential variation of RT efficiency at different RNA input levels.

Product	Quantity	Cat. No.
SuperScript® VILO™ cDNA Synthesis Kit	50 reactions	11754-050
	250 reactions	11754-250

SuperScript® VILO™ Mastermix

The SuperScript® VILO™ Mastermix provides the same superior performance of the SuperScript® VILO™ cDNA Synthesis Kit, but in a convenient single-tube format.

- Convenient—single-tube format
- Greater accuracy—reduce bias generated from low- or high-abundance genes
- Consistent results—obtain reliable analysis of more genes
- Reduced qPCR inhibition—use greater amounts of cDNA in qPCR to increase sensitivity up to 4-fold without fear of inhibiting the reaction

Product	Quantity	Cat. No.
SuperScript® VILO™ Mastermix	50 reactions	11755-050
	250 reactions	11755-250
	500 reactions	11755-500

High Capacity RNA-to-cDNA™ Kit

High-quality reverse transcription kit for gene expression research

- Easy workflow with few pipetting steps (2 tubes)
- Short reaction time (0.5–1 hr)
- Reliable reverse transcription (RT) of abundant and limited targets
- Optimized 2-step protocol enabling multiple PCRs from a single reverse transcription reaction

The High Capacity RNA-to-cDNA™ Kit is a streamlined reverse transcription kit designed for optimum performance with the TaqMan® Gene Expression Master Mix, *Power* SYBR® Green PCR Master Mix, and others. The components of the two-tube kit work together to provide sensitive and specific RT across a broad range of template quantities. The High Capacity RNA-to-cDNA™ Kit formulation reduces RT time by at least half and requires fewer pipetting steps compared with the classic High Capacity cDNA Reverse Transcription Kit, while maintaining similar high performance.

The High Capacity RNA-to-cDNA™ Kit includes:

- 20X Enzyme mix containing MuLV and RNase inhibitor protein
- 2X RT Buffer mix containing dNTPs

Streamlined workflow

The High Capacity RNA-to-cDNA™ Kit consolidates the components of the RT reaction into two tubes; 2X RT Buffer and 20X Enzyme mix, including RNase inhibitor. This enzyme-buffer system has been optimized to provide superior cDNA archiving for real-time gene expression analysis. The kit minimizes the number of reagent additions required, thereby decreasing the likelihood of pipetting errors.

Optimized RT kits for TaqMan® Gene Expression Master Mix or *Power* SYBR® Green PCR Master Mix

The High Capacity RNA-to-cDNA™ Kit also works well with the *Power* SYBR® Green PCR Master Mix in an integrated, 2-step, RT workflow with maximum sensitivity and dynamic range.

Product	Quantity	Cat. No.
High Capacity RNA-to-cDNA™ Kit	50 reactions	4387406

High Capacity cDNA Reverse Transcription Kit

Proven single-stranded cDNA synthesis for quantitative amplification of total RNA

- High yield, linearity, and precision
- Linear amplification of all targets for real-time PCR
- Excellent value

The High Capacity cDNA Reverse Transcription Kit contains the components necessary for the quantitative conversion of up to 2 µg of total RNA in a single 20-µL reaction to single-stranded cDNA.

cDNA from total RNA

The High Capacity cDNA Reverse Transcription Kit (formerly the High Capacity cDNA Archive Kit) delivers extremely high-quality, single-stranded cDNA from total RNA. While designed for short- or long-term archiving of cDNA, the kit yields very quantitative amplification of total RNA from 0.02 to 2 µg of total RNA. Downstream applications include real-time PCR, standard PCR, and microarrays.

Extensively tested with a variety of templates

Quantitative, first-strand synthesis of all RNA species is achieved with the use of random primers. The kit has been extensively tested with a variety of RNA templates including "difficult targets" (e.g., GC- and AU-rich RNAs). Our scientists have also demonstrated consistent amplification of RNA transcripts from a wide range of expression levels. cRNA can also be efficiently generated from *in vitro* transcription of cDNA.

Product	Quantity	Cat. No.
High Capacity cDNA Reverse Transcription Kit	200 reactions	4368814
	1,000 reactions	4368813
High Capacity cDNA Reverse Transcription Kit with RNase Inhibitor	200 reactions	4374966
	1,000 reactions	4374967

Custom DNA primers

Overview of custom primers

Invitrogen custom DNA primers are synthetic oligonucleotides made from your specified sequence for use in a variety of applications, from PCR and sequencing, to probes for gene detection. Custom primers are offered with standard deoxynucleotides, modified bases, and 5´- and 3´-modified nucleotides. Available modifications include fluorescent dyes, enzyme conjugates, and S-oligos for antisense studies. Invitrogen offers five synthesis scales and four purity levels.

The Invitrogen custom primers service offers:

- High capacity allows for prompt, reliable delivery
- Rigorous quality control and validation of suppliers and raw materials that includes 100% in-process trityl monitoring in real time, and post-synthesis QC by capillary electrophoresis or mass spectroscopy to ensure quality
- A Certificate of Analysis that includes the sequence and length, average coupling efficiency, melting temperature, GC content, quantity in OD units, µg and nmol amounts, molecular weight, and extinction coefficient
- A variety of formats that include tubes (2 mL), 96-well (6 choices), and 384-well (2 choices) plates, lyophilized or normalized to your desired volume and/or concentration

Oligo ordering process

Orders can be submitted easily via our online tool. Web orders require a simple upload of your sequences and easy-to-obtain technical information about your oligos.

To view complete product descriptions, access online ordering links, or fax or email order forms, go to www.invitrogen.com/oligos.

For information regarding the shipping of oligos, go to the "Delivery Schedules" in our oligo portal at www.invitrogen.com/oligos.

ASR/GMP oligonucleotides

ASR/GMP oligonucleotides are oligos manufactured in compliance with the US Food and Drug Administration (FDA), guidance documents 21 CFR Part 820 and 21 CFR 864.4020 and designed for in-house laboratory testing applications. Analyte Specific Reagent (ASR*) designates a reagent that is produced for *in vitro* diagnostic manufacturers, for Clinical Laboratory Improved Amendments (CLIA)-regulated clinical labs, or for organizations that use the reagents to provide analytical results. Invitrogen is registered with the FDA as a Class I ASR manufacturer. For details on scales, purifications, and prices, go to www.invitrogen.com/oligos.

For information regarding minimum yield guarantees, purity specifications, modifications, and recommendations based on your application, go to www.invitrogen.com/oligos.

* Analyte Specific Reagent. Analytical and performance characteristics are not established.

Oligo purity selection guide

Different applications demand different DNA oligo purity or scale to be successful. Below are guidelines for purity and synthesis scale selection for different applications.

Understanding why oligos may require purification

Following DNA synthesis, the completed DNA chain is released from the solid support by incubation in basic solutions such as ammonium hydroxide. This solution contains the required full-length oligo, but also contains all of the DNA chains that were aborted during synthesis (failure sequences). If a 20-mer was synthesized, the solution would also contain 19-mer failures, 18-mer failures, 17-mer failures, etc. The amount of failure sequences present is influenced by the coupling efficiency. These failure sequences can compete with the full-length product in some applications such as PCR and may therefore need removing before the oligo can be used successfully.

Purification options offered

Purification method	Description	Benefit
Desalted	Oligos are processed through a normal phase chromatography column that removes salts but not failure sequences	A salt-free DNA solution, ready to use; suitable for many PCR and sequencing applications without further purification
Cartridge	Based on reverse-phase chromatography; removes failure sequences from the completed synthesis	Provides full-length sequences needed in some applications
HPLC	Reverse-phase high-performance liquid chromatography (HPLC) removes failure sequences or unincorporated label the same way as cartridge purification	Guarantees highly purified primer required in some applications (≥85% full-length)
PAGE	Method used to differentiate full-length product from failure sequences based on size and conformation	Provides the highest percentage of full-length oligos (≥85%) required for certain demanding applications, such as mutagenesis or adapter production

Application purity guide

Application	Suggested purity
AFLP® analysis	Desalted oligos have been used successfully for Amplified Fragment Length Polymorphism
Antisense	HPLC-purified oligos are cited most frequently in references for antisense studies
First-strand cDNA synthesis for generation of libraries	Generally oligos for first-strand cDNA synthesis for library construction have some sequence at the end that codes for 5´ restriction endonuclease cloning sites. Therefore, it is best to use full-length, cartridge-, HPLC-, or PAGE-purified oligos
Fluorescent sequencing	All four purity grades have worked successfully for Life Technologies scientists
Gel shift assays	Cartridge-, HPLC-, and PAGE-purified oligos are recommended for gel shift assays, so as to have a homogeneous population of DNA fragments
Isothermal sequencing	Desalted oligos are sufficient for this application, along with cartridge-, HPLC-, and PAGE-purified oligos
Microarrays	Standard, desalted oligos are sufficient for printing onto arrays
PCR	Desalted oligos work well for standard PCR. Higher purity options will also work
PCR using oligos with critical 5´ sequences (e.g., restriction endonuclease sites, RNA polymerase promoters)	Cartridge-, HPLC-, and PAGE-purified oligos are best for the greatest efficiency. Because oligos are synthesized 3´ to 5´, incomplete oligos (n-x oligos) will be missing the 5´ sequence. It is important to use full-length oligos that have the 5´ sequence present; otherwise, there will be a population of PCR products missing the sequence intended to be installed before PCR
Production of cloning adapters	Full-length oligos work best for efficient cloning. Utilize cartridge-, HPLC-, or PAGE-purified oligos for full length
Site-directed mutagenesis	Full-length (e.g., cartridge-, HPLC-, and PAGE-purified) oligos tend to give the highest percentage of mutagenized clones (especially if the intended mutation is close to the 5´ end of the oligo). Desired mutations have been obtained using desalted oligos. However, some wild-type parental vector clones tend to carry over

Custom primers specialized to meet your needs

Use the following table to choose your Invitrogen custom primers.

Purification, lengths, and scale	Starting scale				
	25 nmol	50 nmol	200 nmol	1 µmol	10 µmol
Desalted	10–100-mers	5–100-mers	5–100-mers	5–100-mers	5–100-mers
Cartridge	NA	7–60-mers	7–60-mers	7–60-mers	7–60-mers
HPLC	NA	7–55-mers	7–55-mers	7–55-mers	7–55-mers
PAGE	NA	NA	7–100-mers	7–100-mers	7–100-mers
Internal modifications and scale	**Starting scale**				
	25 nmol	50 nmol	200 nmol	1 µmol	10 µmol
Mixed bases	●	●	●	●	●
Deoxyuracil	●	●	●	●	●
Deoxyinosine	●	●	●	●	●
S-oligos	NA	●	●	●	●
3´ modifications and scale	**Starting scale**				
	25 nmol	50 nmol	200 nmol	1 µmol	10 µmol
Phosphate**	NA	●	●	●	Inquire
Biotin**	NA	NA	●	●	Inquire
5´ modifications and scale	**Starting scale**				
	25 nmol	50 nmol	200 nmol	1 µmol	10 µmol
Aldehyde**	NA	●	●	Inquire	Inquire
Amine	●	●	●	●	●
Biotin†	●	●	●	●	●
General modifications	**Starting scale**				
	25 nmol	50 nmol	200 nmol	1 µmol	10 µmol
Phosphate**,†	●	●	●	●	●
Thiol	NA	●	●	●	Inquire
Fluorescent dye modifications	**Starting scale**				
	25 nmol	50 nmol	200 nmol	1 µmol	10 µmol
FAM™ dye	●	●	●	●	●
Fluorescein	●	●	●	●	●
HEX™ dye	●	●	●	●	●
ROX™ dye	●	●	●	●	●
TET™ dye	●	●	●	●	●
TAMRA™ dye	NA	NA	●	●	●

* Note: not available in all regions.

● Indicates an item is available in that scale. NA indicates that the item is not available at that scale. Modifications in blue require HPLC purification. ** Cartridge purification not available. † HPLC not available in 50 nmol scale.

For "Inquire" or further questions about Invitrogen™ Custom Primers, contact Technical Services at 800.955.6288 (in the USA) or email techsupport@invitrogen.com.

Custom primers specialized to meet your needs, cont.

Molecular Probes® dyes	Starting scale				
	25 nmol	50 nmol	200 nmol	1 µmol	10 µmol
Alexa Fluor® 350; 405; 430; 488; 514; 532; 546; 555; 568;594; 633; 647; 660; 680; 700; 750 dyes	NA	●	●	Inquire	Inquire
BODIPY® 530/550; 493/503; 558/569; 564/570; 576/589; 581/591; 630/650-X; 650/665 dyes	NA	NA	●	●	●
BODIPY® FL; FL-X; TR-X; TMR; R6G; R6G-X dyes	NA	NA	●	●	●
Cascade Blue® dye	NA	NA	●	●	●
Marina Blue® dye	NA	●	●	●	●
Oregon Green® 500; 514; 488; 488-X dyes	NA	NA	●	●	●
Pacific Blue™ dye	NA	NA	●	●	●
Rhodamine Green™ dye	NA	NA	●	●	●
Rhodamine Green™-X dye					
Rhodol Green™ dye	NA	NA	●	●	●
Rhodamine Red™-X dye	NA	NA	●	●	●
Texas Red®; Texas Red®-X dyes	NA	NA	●	●	●
Other modifications	**Starting scale**				
	25 nmol	50 nmol	200 nmol	1 µmol	10 µmol
Gateway® FW	NA	●	●	●	Inquire
Gateway® RV	NA	●	●	●	Inquire
M13 tails	NA	NA	●	●	NA

• Indicates an item is available in that scale. NA indicates that the item is not available at that scale. Modifications in blue require HPLC purification. ** Cartridge purification not available. † HPLC not available in 50 nmol scale.

For "Inquire" or further questions about Invitrogen™ Custom Primers, contact Technical Services at 800.955.6288 (in the USA) or email techsupport@invitrogen.com.

For more information on ordering custom primers, yield guarantees, designing tools, technical resources, protocols, and FAQs, go to www.invitrogen.com/oligos.

cloning

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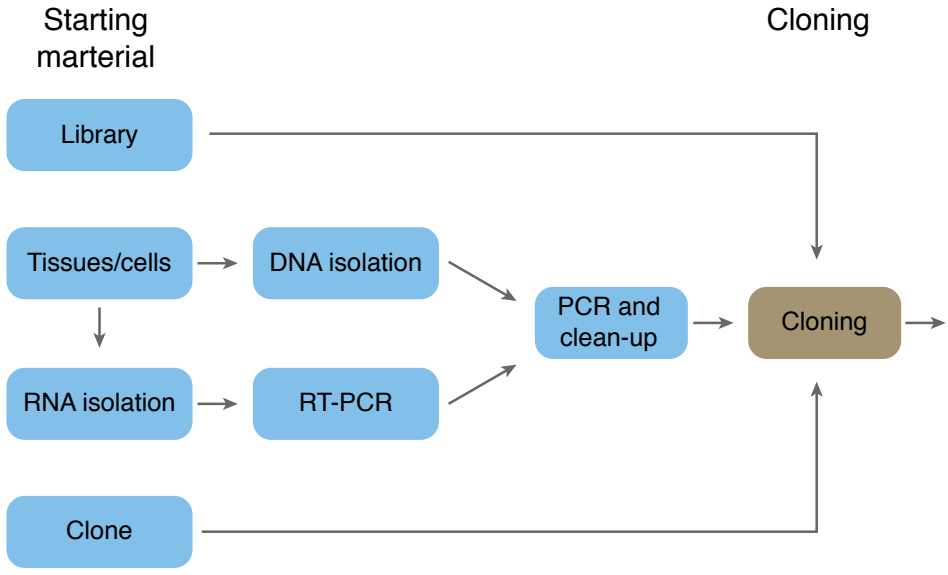
Overview of cloning

We offer a number of proven technologies, kits, and reagents to help you achieve your cloning goals. To determine the best path for your particular cloning research, you first need to determine the source of the genomic material you will be cloning. There are a variety of ways to prepare or obtain ready-to-clone DNA, including:

- Making or buying a library
- Purchasing a premade clone
- Reverse-transcribing from RNA
- Purifying from tissues or cells
- Starting with cDNA

If you obtain your starting material from a source other than a cDNA library or premade clone, you will need to amplify it via PCR to generate a sufficient amount of material for the actual cloning reaction. We offer a number of amplification enzymes, each optimized for use with different DNA challenges such as fidelity, sensitivity, yield, length, and GC-rich content.



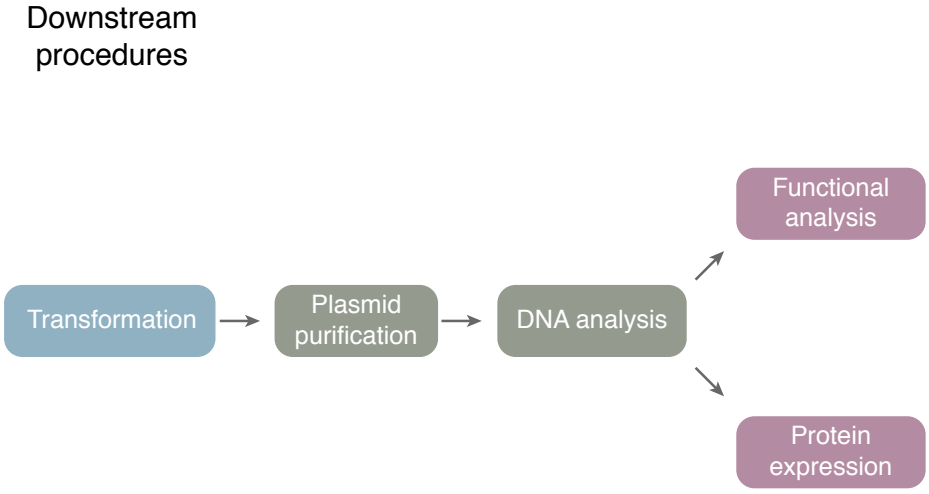
Select the cloning method that fits your requirements.

	Restriction enzymes	TA vector	TOPO® (TA, blunt, directional)
Fragments cloned simultaneously	1	1	1
Max fragment(s) size	Variable	1–3 kb	<5 kb (<10 kb for XL-TOPO®)
Gene shuttling between vectors without PCR or restriction enzymes	No	No	No
Seamless (no extra sequences)	No	No	No
Use your own vector without modification	Yes	No	No
Use preexisting fragments without modifications	No	No	No
Time to clone multiple fragments	Days to weeks	Not possible	Not possible
4-fragment cloning efficiency	NA	NA	NA
Web-based vector design tool	No	No	No

Choosing a cloning method

A number of different methods, technologies, and protocols are available for performing the actual cloning reaction. To choose the one that best meets your needs, you will need to analyze the current parameters of your experiment and plan ahead for any downstream procedures. This section discusses several highly efficient cloning technologies:

- Restriction digestion, phosphatase treatment, and ligase cloning is an industry standard for molecular biologists
- TOPO® cloning provides simple, convenient reactions typically in less than 5 minutes
- Gateway® technology offers the greatest flexibility in vector choices and downstream applications
- Seamless Cloning and Genetic Assembly offer tools optimized to clone up to 4 DNA fragments simultaneously into virtually any linearized *E. coli* vector in a 30-minute room temperature reaction (up to 13 kb total size)
- Clone up to 10 fragments simultaneously and seamlessly in yeast with the GeneArt® High-Order Genetic Assembly Kit
- Gene synthesis offers 100% accuracy and optimization of genes to maximize expression



	Gateway®	GeneArt® Seamless Cloning	GeneArt® High-Order Genetic Assembly
	Up to 4	Up to 4	Up to 10
	Variable	Up to 10 kb, max total size of 13 kb	Up to 100 kb, max total size of 100 kb
	Yes	No	No
	No	Yes	Yes
	Yes	Yes	Yes
	No	Yes (1 fragment)	Yes (3 fragments)
	>4 days	1 day	3 days
	30–85%	75%	>90%
	No	Yes	Yes

Ultimate™ ORF expression-ready clones

Quality clones let you move directly to protein expression

- Vast selection—over 16,000 Ultimate™ Human ORF Clones and over 2,500 Ultimate™ Mouse ORF Clones
- Gateway® entry clone format—1-hour recombinatorial cloning into expression vectors gets you to expression and analysis faster
- 100% amino acid match guarantee—sequence verification against GenBank®, Ensembl®, and Swiss-Prot® databases

Ultimate™ ORF clones eliminate the initial cloning and verification steps of your gene discovery research. There is no need to perform tedious RNA isolation, cDNA synthesis, PCR amplification, cloning, sequencing, and validation procedures. You will save literally weeks of time by moving directly to the expression and protein analysis steps of your workflow.

All Ultimate™ ORFs are provided in the Gateway® entry vector pENTR™221, and they have the amber stop codon that makes them compatible with the Tag-On-Demand™ technology. You can rapidly and efficiently recombine the ORF of interest into any expression vector and go straight to gene analysis in your system of choice. To learn more about how Gateway® technology can save you time and effort please visit www.invitrogen.com/gateway.

Quality tested

The Ultimate™ ORF Clone Collection is updated quarterly with the most current information from the GenBank®, Ensembl®, and Swiss-Prot® databases and if any Ultimate™ ORF Clone becomes invalid, they are deactivated from the collection and moved to the ORFanage Collection. The Ultimate™ ORF Clones are designed to match GenBank® sequence information 100% at the amino acid level. To ensure that you receive the highest-quality and most scientifically relevant clones, Ultimate™ ORF clones have been full-insert sequenced. In addition, before shipping, the clone culture must pass a stringent QC test that includes end-sequencing to guarantee the identity of the clone. You can have peace of mind knowing the clone you ordered is the clone you will receive.

Volume discounts are available

Whether you are looking at purchasing one gene or a subset of druggable targets from our collection, we have options for you. Ultimate™ ORF Clones are available in tube format with price discounts based on volume. Also, for large orders we offer cost-effective formats such as the 96-well plates or the Ultimate™ ORF Clone LITE.

A budget version of our Ultimate™ ORF clones

- Vast selection of over 16,000 Ultimate™ Human ORF Clones
- Gateway® entry clone format—1-hour recombinatorial cloning into expression vectors gets you to expression and analysis faster

Ultimate™ ORF LITE clones are a budget-friendly version of our regular Ultimate™ ORF Collection. The clone stocks used to prepare the LITE clones are the same as for our premium Ultimate™ ORF offering. However, the quality control skips the end-sequencing prior to the shipment of each clone ID to pass the savings to you.

Quality control

The Ultimate™ ORF Clone LITE Collection is updated quarterly with the most current information from the GenBank®, Ensembl®, and Swiss-Prot® databases, and if any Ultimate ORF Clone becomes invalid, it is deactivated from the collection and moved to the ORFanage Collection. The Ultimate™ ORF LITE Clones are not guaranteed to match GenBank® sequence, and in some cases, screening two to four individual colonies might be needed to isolate the clone that matches 100% the amino acid sequence.

Comparison of the standard and LITE versions of the Ultimate™ ORF Clone Collections.

(Parent stock for both collections are fully sequenced, single PCR ORF with start and stop codons)

	Ultimate™ ORF Clone Collection	Ultimate™ ORF Clone LITE Collection
Gateway® vector–compatible	Yes	Yes
Tested for phage	Yes	Yes
Tested for growth and antibiotic resistance	Yes	Yes
Isolation of individual colonies for each clone ID	Yes	No
Validation of clone identity by sequencing on both ends of the ORF	Yes	No
Sequenced-guaranteed	Yes	No

CloneMiner™ II cDNA Library Construction Kit

Construct highly representative cDNA libraries

CloneMiner™ II cDNA Library Construction Kits enable rapid construction of highly representative cDNA libraries. Gateway® recombination cloning avoids restriction digestion and ligation reactions, making library construction faster and offering more complete cDNA representation. The high-efficiency SuperScript® III reverse transcriptase is included, enhancing the possibility that users may discover previously unobtainable, fully intact clones.

Product	Quantity	Cat. No.
CloneMiner™ II cDNA Library Construction Kit	1 kit	A11180

GeneRacer® kits

Advanced RACE method for amplification of full-length cDNA ends

- Generate cDNA from transcripts up to 10 kb in length
- Obtain the full-length 5´ end of rare transcripts at fewer than 30 copies per cell
- Clone the full-length 5´ and 3´ ends to construct a complete cDNA sequence

GeneRacer® kit is an advanced RACE (rapid amplification of cDNA ends) technique that improves efficiency and helps ensure that only transcripts containing full-length cDNA ends are amplified. The advanced protocol starts at the RNA level by specifically targeting only 5´ capped mRNA. In subsequent steps, the cap is removed and replaced with the GeneRacer® RNA Oligo. During reverse transcription, the GeneRacer® RNA Oligo sequence is incorporated into the cDNA. Only cDNA that is completely reverse transcribed will contain this known sequence. 5´ RACE PCR is then performed using the GeneRacer® 5´ Primer specific to the GeneRacer® RNA Oligo sequence and a gene-specific primer. The result is amplified DNA that contains the full-length 5´ cDNA sequence. The GeneRacer® Kit is available with SuperScript® III Reverse Transcriptase (RT) for improved amplification of the full-length 5´ end from long and complex mRNA.

Product	Quantity	Cat. No.
GeneRacer® Kits		
with SuperScript® III RT and TOPO® TA Cloning® Kit for Sequencing	1 kit*	L150201
with SuperScript® III RT and Zero Blunt® TOPO® PCR Cloning Kit for Sequencing	1 kit*	L150202
with Cloned AMV RT and TOPO® TA Cloning® Kit for Sequencing	1 kit*	L150001

Each GeneRacer® Kit contains sufficient reagents for five cDNA reactions plus one control reaction and primers for 50 PCRs. The kit also contains S.N.A.P.™ columns and a TOPO® Cloning Kit. * A thermostable DNA polymerase for PCR is available separately. This product contains components sold under a license from the Université Libre de Bruxelles.

SuperScript® III Reverse Transcriptase

Improved thermostability for better yields

- Longer half-life at 50°C for the highest cDNA yields
- Reduced RNase H activity for more full-length cDNA
- Ability to increase RT units without inhibiting subsequent PCR

SuperScript® III reverse transcriptase (RT) is a proprietary, genetically engineered M-MLV RT which synthesizes cDNAs from single-stranded RNA, DNA, or RNA:DNA hybrids. Like SuperScript® II, it provides high-yield and full-length cDNA due to reduced RNase H activity. The added benefit of SuperScript® III is that it also demonstrates increased thermal stability for enhanced specificity and sensitivity. Common applications for SuperScript® III RT include synthesis of first-strand cDNA, array labeling, cDNA libraries, RT-PCR, primer extension, and 3´ and 5´ RACE.

Selection guide for reverse transcriptase enzymes.

	SuperScript® III	SuperScript® II	M-MLV	AMV
Source	Moloney murine leukemia virus (M-MLV)	Moloney murine leukemia virus (M-MLV)	Moloney murine leukemia virus	Avian myeloblastosis virus
Modification	Clone with 7 point mutations to reduce the RNase H activity and increase thermal stability	Clone with 3 point mutations to reduce the RNase H activity	Clone	Clone
Template	ssRNA or DNA	ssRNA or DNA	ssRNA or DNA	ssRNA or DNA
Endonuclease activity	No	No	No	Yes
Inhibited by rRNA and tRNA	No	No	No	Yes
Optimum operating temperature	50–55°C	42°C	37°C	42–60°C
Target size	>12 kb	>12 kb	<7 kb	<9 kb
RNase H activity	•	•	•••••	•••••••
Yield	••••	•••	••	•
Sensitivity	••••	•••	•	•

Product	Concentration	Quantity	Cat. No.
SuperScript® III Reverse Transcriptase	200 units/µL	2,000 units	18080093
	200 units/µL	10,000 units	18080044
	200 units/µL	4 x 10,000 units	18080085
SuperScript® II Reverse Transcriptase	200 units/µL	2,000 units	18064022
	200 units/µL	10,000 units	18064014
	200 units/µL	4 x 10,000 units	18064071
M-MLV Reverse Transcriptase	200 units/µL	40,000 units	28025013
	200 units/µL	200,000 units	28025021
AMV Reverse Transcriptase, Cloned	15 units/µL	750 units	12328019
	15 units/µL	3,000 units	12328027

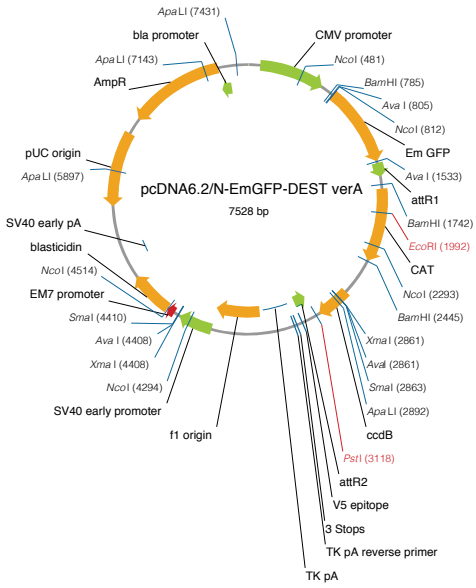
SuperScript® RTs and M-MLV are supplied with 5X first-strand buffer and DTT. Cloned AMV Reverse Transcriptase is supplied with 5X first-strand buffer, DTT, magnesium acetate, and sodium pyrophosphate. Other libraries are also available, including *Arabidopsis*, rat, and HeLa cell. For a complete list of SuperScript® premade libraries, visit www.invitrogen.com.

For more information, go to www.invitrogen.com/superscript.

Vector NTI Advance® v11.5 Sequence Analysis and Design Software

The leading sequence analysis and design software

Vector NTI Advance® software is a completely integrated suite of sequence analysis and design tools that allows you to manage, view, analyze, transform, share, and publicize diverse types of molecular biology data, all within a single, graphically rich analysis environment.



Vector NTI Advance® software empowers users to:

- Curate—store and manage collections, visualize maps, and search sequences
- Discover—analyze, compare, and contrast sequences
- Design—cloning, primers, and gel simulations of sequences
- Confirm—contig assembly, sequence validation, and literature validation

The software contains a comprehensive set of data analysis and management tools, implemented across five application modules:

Vector NTI® software

- Sequence analysis, annotation, and illustration
- Restriction mapping; recombinant molecule design, including Gateway® and TOPO® cloning; GeneArt® Seamless Cloning and High-Order Assembly; and *in silico* GeneArt® gene synthesis
- Synthetic biology workflows support and data management

AlignX® software

- Multiple sequence alignment of proteins and DNAs
- Alignment statistics, cladograms, alignment editing, annotation, and repeat identification

ContigExpress® software

- DNA sequence assembly and editing, contig building, SNP and mutation detection, and genotype analysis
- Chromatogram data analysis and editing; consensus creation using quality values (QV)
- Automatic sequence trimming and vector contamination trimming

GenomBench® software

- Visualization and analysis of megabase-sized genomic DNA fragments from numerous DAS servers
- Chromosomal views, cDNA-to-gene alignment, intron–exon boundary mapping, and annotation

BioAnnotator® software

- Protein motif mapping and annotation using Pfam, PROSITE, and BLOCKS motif databases
- Physiochemical analyses of DNA sequences

Vector NTI Advance® runs on Windows® 7, Windows® XP operating systems (including Japanese language editions), and Mac® OS X 10.6 with Parallels Desktop® 5 and above.

Catalogue numbers for our most popular academic and industrial licenses.

Academic licenses		Industrial licenses	
12605099	1-year academic license	A13784	1-year industrial license
12605103	3-year academic license	A13785	3-year industrial license
A13786	Static, non-expiring academic license	12605050	Static, non-expiring industrial license

To try Vector NTI Advance®, FREE for 30-days and for additional license configurations, go to www.invitrogen.com/vectornti.

GeneArt® gene synthesis and cloning tools

Gene synthesis has become the most cost-effective and time-saving method for obtaining nearly any desired DNA construct, outperforming conventional molecular biology techniques in many aspects—from time and cost savings to expression performance, stability, and quality. GeneArt® gene synthesis tools go beyond traditional synthesis and enable expression optimization and maximum performance.

Your benefits

- Free expression and mRNA stability optimization
- Unlimited flexibility in gene and vector design
- Empirically proven increases in expression
- Ready-to-use constructs for expression and transfection
- Subcloning in any vector

Quality

- All processes are ISO 9001:2008 quality certified
- Comprehensive quality documentation included (GMP conformity upgrades available)
- All production processes are highly automated

SuperSPEED gene synthesis service

- Up to 1,200 bp in 5 business days
- Up to 1,800 bp in 7 business days
- Emergency genes on request

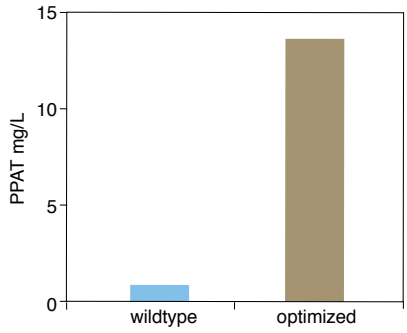


Figure 1. GeneOptimizer® expression *E. coli* optimization. Data reproduced from *Protein Expr Purif* 39:296 (2005).

Performance

- Project setup assistance and individual project support
- Maximum performance is guaranteed by the GeneOptimizer® sequence optimization tool—our proprietary and industry-preferred optimization algorithm (see Figure 1)
- Maximum speed of production and worldwide delivery; capacity and reliability is guaranteed by GeneAssembler® technology—the world's only industrial gene processing platform (Figure 2)

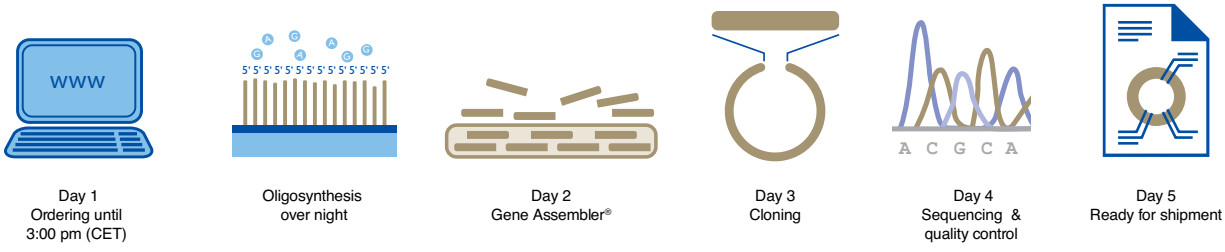


Figure 2. SuperSPEED production schedule.

To learn more and place your order, please go to www.invitrogen.com/genesynthesis.

GeneArt® Site-Directed Mutagenesis System

Efficiency in site-directed mutagenesis workflows

The GeneArt® Site-Directed Mutagenesis System is a simple and highly efficient method for *in vitro* site-directed mutagenesis (Figure 1). The system can generate base substitutions, deletions, or insertions in DNA plasmids from any source, with no specialized vectors, host strains, or restriction sites required.

- Control—insert, delete, or change up to 12 nucleotides in plasmids up to 14.5 kb
- Speed—entire protocol including transformation is complete typically in less than 3 hours (using a 3 kb plasmid)
- Precision and efficiency—mutagenesis efficiency over 90% (using a 3 kb plasmid)

The GeneArt® Site-Directed Mutagenesis System utilizes mutagenic oligonucleotide primers to generate mutations. The mutagenesis protocol is streamlined by combining DNA methylation and amplification steps into a single reaction and eliminating post-mutagenesis digestion and purification steps (Figure 2).

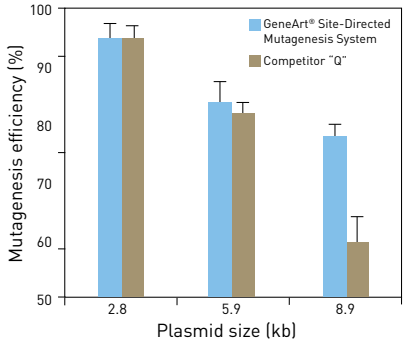


Figure 1. The GeneArt® Site-Directed Mutagenesis System delivers superior mutagenesis performance with a wide range of vector sizes. A comparative analysis against competitor "Q" reveals the advantage of using GeneArt® Site-Directed Mutagenesis for a single–base pair mutation.

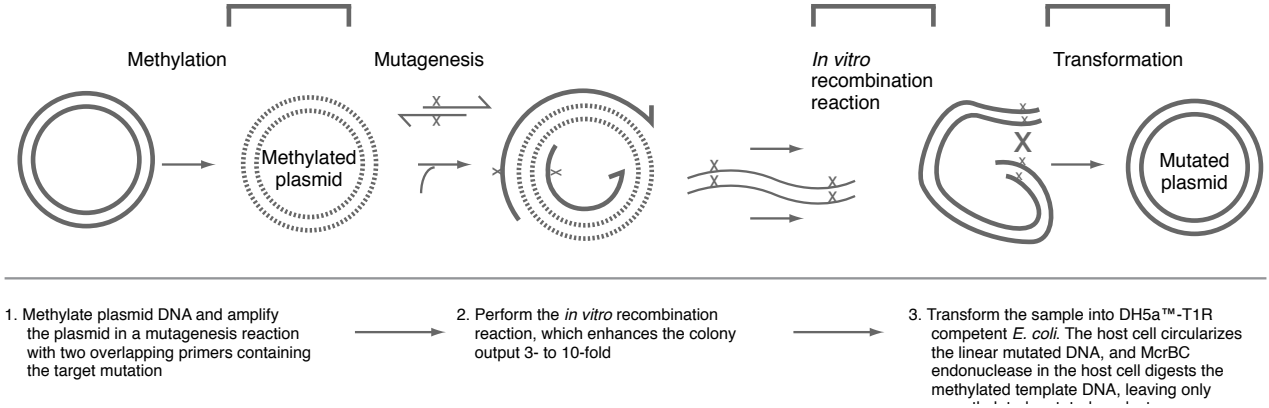


Figure 2. The GeneArt® Site-Directed Mutagenesis protocol streamlines and simplifies mutagenesis.

Product	Quantity	Cat. No.
GeneArt® Site-Directed Mutagenesis System	16 reactions	A13282

Kit contents: DNA methylase (4 units/µL), 20 µL; 200X SAM (S-adenosyl methionine), 10 µL; 10X Enhancer, 100 µL; 0.5 M EDTA, 500 µL; pUC19WHITE Control Plasmid (20 ng/µL), 100 ng; Control Primer Mix (10 µM), 25 µL; PCR water, 1.8 mL; 5X Reaction Buffer, 90 µL; 10X Enzyme Mix, 45 µL; One Shot® MAX Efficiency® DH5α™ T1^{R}, 1 box

To learn more about GeneArt® products, go to www.invitrogen.com/dnaassembly.

GeneArt® Seamless Cloning and Assembly System

Simultaneous, multiple-fragment cloning at rapid speed

The GeneArt® Seamless Cloning and Assembly System enables cloning of up to four DNA fragments simultaneously into virtually any linearized vector typically in 30 minutes, without extra DNA sequences, restriction endonucleases, or ligation (Figure 1).

- Speed—clone up to four DNA fragments simultaneously in a single tube, at room temperature, typically in 30 minutes
- Flexibility—use our linear vector or any vector of your choice
- Precision and efficiency—no extra sequences; just clone what you want where you want it (Figure 2)
- Simplicity—Web-based DNA Oligo Designer software designs oligos and assembles DNA molecules *in silico*

The GeneArt® Seamless Cloning and Assembly System uses a proprietary enzyme mix to recognize and precisely assemble DNA fragments sharing a 15–base pair end homology. End homology is created by PCR amplification using primers designed to generate overlap between adjacent DNA fragments to be assembled. The online DNA Oligo Designer (www.invitrogen.com/designdnaassembly) guides users through experimental design, including oligo design, and ordering.

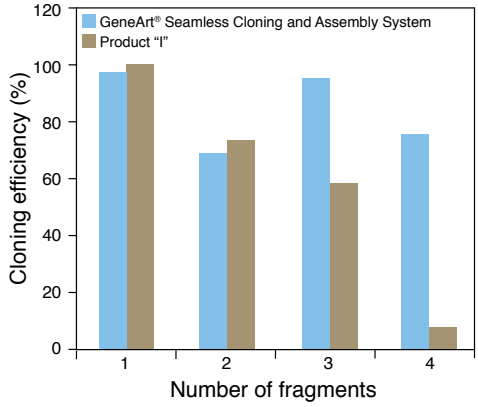


Figure 2. The GeneArt® Seamless Cloning and Assembly Kit delivers higher cloning efficiency than Product "I" when more than two inserts are cloned simultaneously into a linear vector. The two kits were compared for cloning efficiency using the same vector and inserts (1 kb fragments with 15 bp overlaps) in four cloning reactions using AccuPrime™ *Pfx* polymerase: 1: one insert and vector; 2: two inserts and vector; 3: three inserts and vector; 4: four inserts and vector.

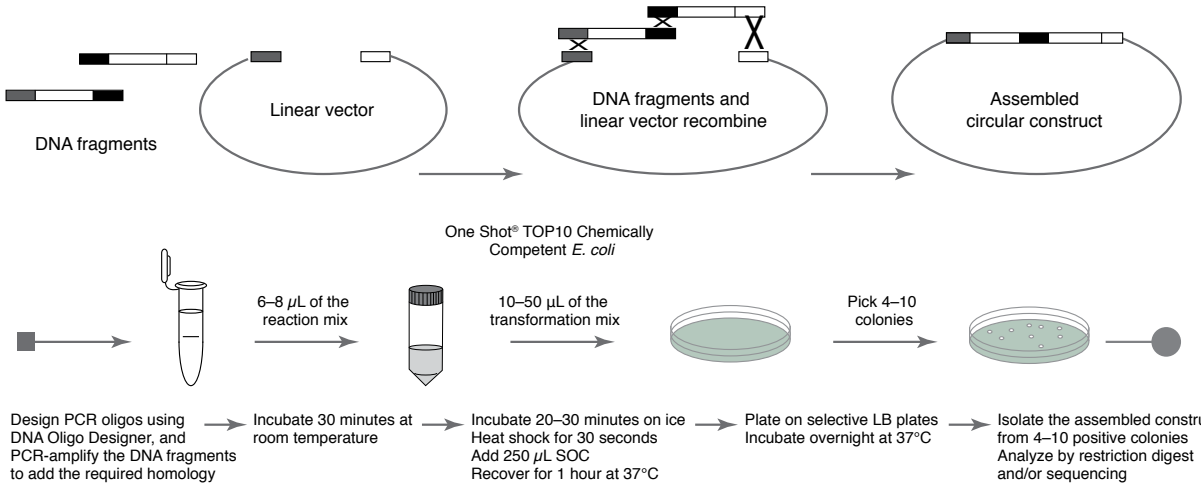


Figure 1. GeneArt® Seamless Cloning and Assembly workflow. The GeneArt® Seamless Cloning and Assembly protocol enables you to assemble up to four DNA fragments and a vector simultaneously and in precise order.

Product	Quantity	Cat. No.
GeneArt® Seamless Cloning and Assembly Kit	20 reactions	A13288

Kit contents: 10X Enzyme mix (45 µL/tube, 1 tube), 5X Enzyme buffer (90 µL/tube, 1 tube), linear pUC19L vector (50 ng/µL) (4 control reactions, 8 µL/tube, 1 tube), control insert (50 ng/µL) (5 µL/tube, 1 tube), One Shot® TOP10 Chemically Competent *E. coli* (50 µL/tube, 21 tubes), pUC19 control (10 pg/µL) (10 µL/tube, 1 tube), SOC medium (6 mL/bottle, 1 bottle)

GeneArt® Linear pUC19L Vector for Seamless Cloning	20 reactions	A13289

To learn more about GeneArt® products from, go to www.invitrogen.com/dnaassembly.

GeneArt® High-Order Genetic Assembly System

Ten-fragment assembly for up to 110 kb

The GeneArt® High-Order Genetic Assembly System is a highly efficient (Figure 1) vector-independent system for the simultaneous and seamless assembly of up to 10 DNA fragments (preexisting or synthetic) *in vivo*, totaling up to 110 kb in construct size.

- Speed—clone 10 or more DNA fragments simultaneously in a single vector (up to 110 kb); assemble existing DNA fragments without restriction digest or PCR amplification
- Flexibility—use our linear vector or a vector of your choice; oligonucleotide stitching feature allows end-editing and reuse of preexisting DNA fragments without the need for reamplification (Figure 2 , next page)
- Precision and efficiency—no extra sequences; just clone what you want, where you want it
- Simplicity—online DNA Oligo Designer walks you through your project, step-by-step; design your DNA oligos and assemble your DNA molecule *in silico* (www.invitrogen.com/designdnaassembly)

The GeneArt® High-Order Genetic Assembly System relies on yeast's ability to take up and recombine DNA fragments with high efficiency via transformation-associated recombination, greatly reducing *in vitro* handling of DNA and eliminating the need for restriction digestion and ligation.

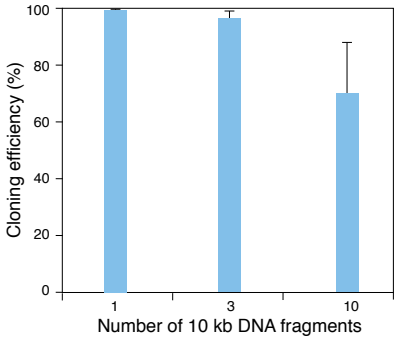
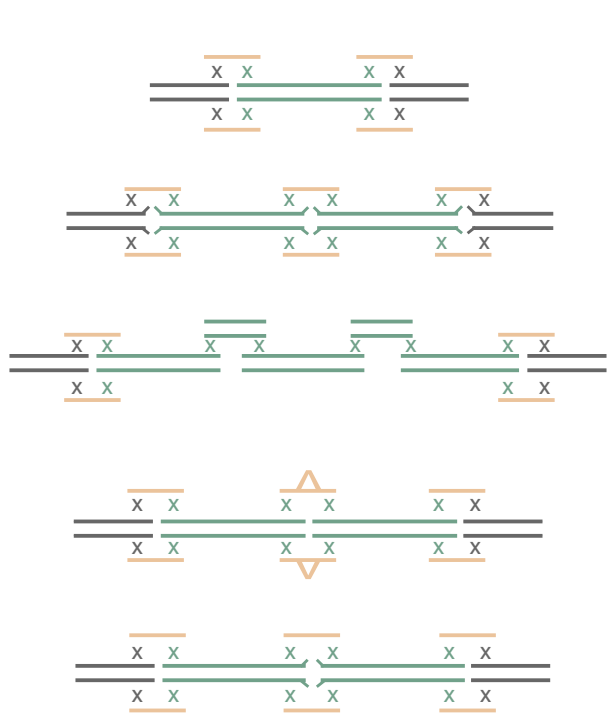


Figure 1. Cloning efficiency of the GeneArt® High-Order Genetic Assembly System with increasing numbers of 10 kb PCR fragments. Even in the case of a 100 kb assembly, the cloning efficiency remains over 65%, proving the system is a reliable solution for assembly of complex DNA molecules.

To learn more about GeneArt® products, go to www.invitrogen.com/dnaassembly.

GeneArt® High-Order Genetic Assembly System Contined



Fragment Size	Oligo Size	Cloning Efficiency
1 x 1 kb	60-mer	94%
2 x 5 kb	80-mer	75%
[3 x 5 kb] + [2 x 0.5] kb	60-mer	37%
[3 x 5 kb] + [2 x 0.5] kb	80-mer	75%
2 x 5 kb	80-mer 10 bp insertion	63%
2 x 5 kb	80-mer 20 bp insertion	50%
2 x 5 kb	80-mer 12 bp insertion	87%

Figure 2. In cases where DNA fragments do not share end-terminal homology (i.e., existing DNA fragments not created by PCR), fragments can be "stitched" together using recombinant linkers that provide sequence overlaps to promote recombinatorial joining of unrelated DNA fragments.

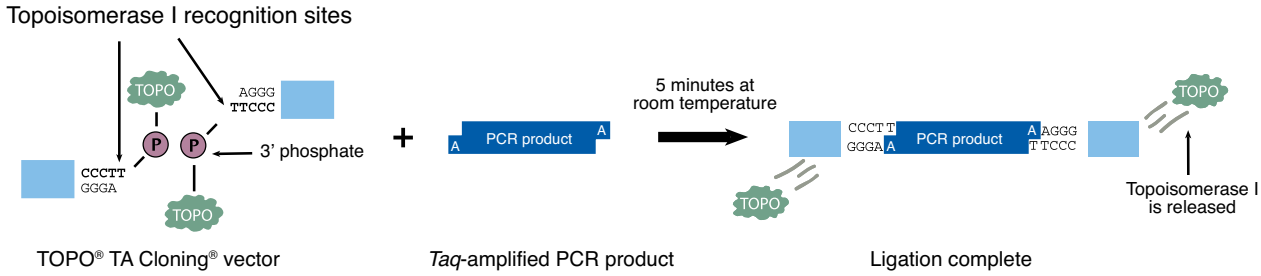
In addition, oligonucleotide stitching allows the editing of the fragment junctions to generate deletions and insertions. This feature also allows the reuse of existing DNA fragments without the need for re-amplifying them by PCR.

Product	Quantity	Cat. No.
GeneArt® High-Order Genetic Assembly System	10 reactions	A13285
GeneArt® High-Order Genetic Assembly System (with Yeast Growth Media)	10 reactions/2 L medium	A13286
GeneArt® High-Order Linear pYES1L Vector	10 reactions	A13287
CSM Media for Mav203 Yeast Cells	2 L	A13292
GeneArt® High-Order Vector Conversion Cassette	10 reactions	A13291

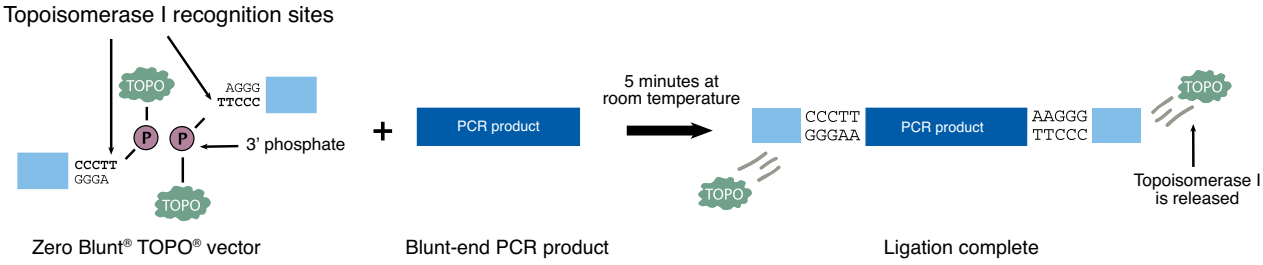
To learn more about GeneArt® products from, go to www.invitrogen.com/dnaassembly.

TOPO® cloning technology

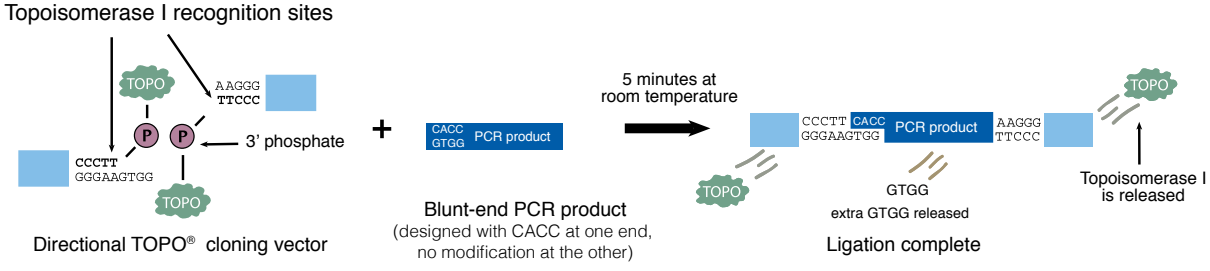
The key to TOPO® cloning is the enzyme DNA topoisomerase I, which functions both as a restriction enzyme and as a ligase. Its biological role is to cleave and rejoin DNA during replication. Vaccinia virus topoisomerase I specifically recognizes the pentameric sequence 5´-(C/T)CCTT-3´ and forms a covalent bond with the phosphate group attached to the 3´ thymidine. It cleaves one DNA strand, enabling the DNA to unwind. The enzyme then re-ligates the ends of the cleaved strand and releases itself from the DNA. To harness the re-ligating activity of topoisomerase, TOPO® vectors are provided linearized with topoisomerase I covalently bound to each 3´ phosphate. This enables the vectors to readily ligate DNA sequences with compatible ends (see figures). The ligation is complete in as little as 5 minutes at room temperature.



TOPO® TA Cloning® of *Taq*-amplified DNA.



Zero Blunt® TOPO® cloning of blunt-end DNA.



Directional TOPO® cloning of blunt-end DNA.

TOPO® TA Cloning® Kits for subcloning

Fast, effective cloning of *Taq*-amplified PCR products

- EcoRI sites flanking the PCR product insertion site for easy excision of inserts
- Kanamycin and ampicillin resistance genes for your choice of selection in *E. coli*
- Easy blue/white colony screening for selection of recombinants

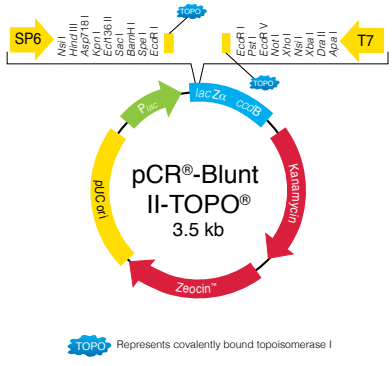
TOPO® TA Cloning® Kits are designed for cloning PCR products directly from a PCR reaction in as little as 5 minutes. They use a pCR™-TOPO® vector with covalently bound topoisomerase I for fast cloning and recombinants. pCR™-TOPO® vectors include 3´-T overhangs for direct ligation of *Taq*-amplified PCR products and a choice of T7 (pCR®2.1-TOPO® cloning) or T7 and SP6 (pCR®II-TOPO® cloning) promoters for *in vitro* RNA transcription and sequencing. Each vector also contains M13 forward and reverse primer sites for sequencing. TOPO® TA Cloning® Kits are available in combo kits combined with a PureLink® Quick Plasmid Miniprep Kit (50 preps) for fast plasmid purification of your TOPO®-cloned inserts for down-stream analysis.

Zero Blunt® TOPO® PCR Cloning Kit

Easy, high-efficiency cloning of blunt-end PCR products

- Up to 95% recombinants via rapid 5-minute benchtop ligation
- Selection of competent cells for high speed, electroporation, or routine cloning
- Minimizes high vector background

The Zero Blunt® TOPO® PCR Cloning Kit combines Zero Background™ and TOPO® technologies to allow easy, high-efficiency cloning of blunt-end PCR products. Zero Background™ technology uses the lethal *ccdB* (control of cell death) gene to cause degradation of the host chromosome and death of *E. coli*-containing empty vectors. When an insert is ligated into the vector, expression of the *ccdB* gene is disrupted, enabling only recombinant colonies to grow. By eliminating high vector background, the Zero Background™ technology yields up to 95% recombinants, saving you from countless hours of screening.

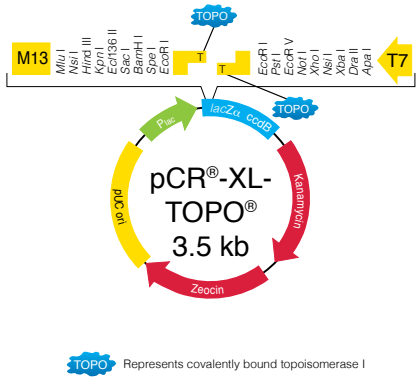


TOPO® XL PCR Cloning Kit

Efficient cloning of long PCR products

- High-efficiency cloning of long PCR products
- Crystal violet staining results for a greater percentage of recombinants
- Eliminates ethidium bromide and UV light exposure for safe gel purification

The TOPO® XL PCR Cloning Kit combines TOPO® Cloning, Zero Background™ technology, and a unique gel purification step for optimal cloning of long PCR products (3–10 kb). The pCR®-XL-TOPO® vector is provided linearized and topoisomerase I–activated for 5-minute bench-top ligation resulting in up to 95% recombinants. It contains 3´-T overhangs for cloning PCR products produced by most thermostable polymerase mixtures.

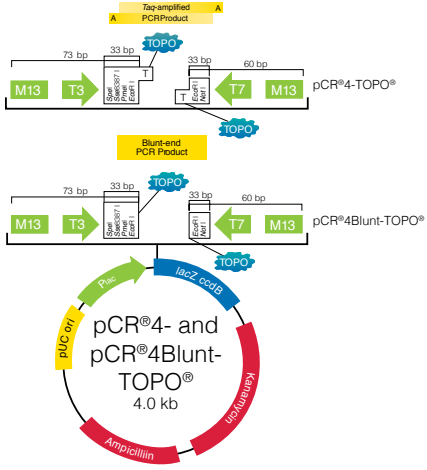


TOPO® Cloning Kits for Sequencing

Fast cloning and streamlined sequencing of PCR products

- T7 and T3 promoter/priming sites for sequencing and *in vitro* transcription/translation
- M13 forward (–20) and reverse priming sites for sequencing or PCR screening
- EcoRI sites flanking the PCR product insertion site for easy removal of inserts

These kits contain TOPO® cloning vectors with a minimized multiple cloning site that positions the T7 and T3 priming sites only 33 bp away from the PCR product insertion site. This means you'll sequence more of your insert and less of the vector. Choose the pCR®4-TOPO® vector for TA Cloning® or pCR®4Blunt-TOPO® for blunt-end cloning, depending on your preferred PCR enzyme. Both TOPO TA® Cloning and Zero Blunt® TOPO® Cloning Kits enable 5-minute benchtop ligations and yield up to 95% recombinants.



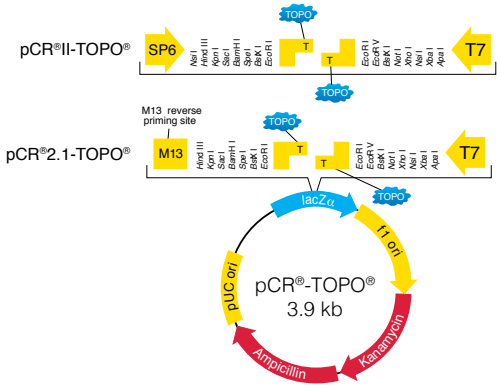
TA Cloning® Kits

Efficient cloning of *Taq*-amplified PCR products

- 3´-T overhangs for direct ligation of *Taq*-amplified PCR products
- Choice of kanamycin or ampicillin selection
- Easy blue/white colony screening

TA Cloning® Kits are designed for cloning *Taq*-amplified PCR products directly from a PCR reaction using an overnight ligation step. The kits use a pCR® vector and yield ≥80% recombinants. The TA Cloning® Kits are available with a choice of T7 (pCR®2.1) or T7 and Sp6 (pCR®II) promoters for *in vitro* RNA transcription and sequencing. All kits have a versatile polylinker with flanking EcoRI sites for easy excision of inserts, and M13 forward and reverse primer sites for sequencing.

Continued on next page.



Selecting the right TOPO® PCR cloning kit

Use the table below to choose the PCR cloning kit that best fits your needs. First, identify the enzyme you will be using for PCR. Next, choose the application for which you will use the cloned PCR product and the ligation method you prefer. Then, choose the kit that has been optimized for your particular application.

Application	Product	TA, blunt, or directional	Advantage
General subcloning	TOPO® TA Cloning® Kit	TA	• Up to 95% recombinants • M13 forward and priming sites for sequencing or PCR • Selection of competent cells for high-speed, electroporation, or routine cloning
	Zero Blunt® TOPO® PCR Cloning Kit	Blunt	• Up to 95% recombinants • Multiple primer sites (T7, T3, M13F, M13R) for sequence analysis or PCR • Selection of competent cells for high-speed, electroporation, or routine cloning • Unique technology to minimize background
Cloning long PCR fragments	TOPO® XL PCR Cloning Kit	TA	• High-efficiency cloning for fragments 3–10 kb • Unique technology to minimize background
In vitro transcription	TOPO® TA Cloning® Kit Dual Promoter	TA	• Up to 95% recombinants • Dual T7and SP6 priming sites for *in vitro* transcription
Sequencing	TOPO® TA Cloning® Kit for Sequencing	TA	• High-efficiency cloning for fragments 3–10 kb • Up to 95% recombinants • Dual T7and SP6 priming sites for *in vitro* transcription
	Zero Blunt® TOPO®	Blunt	• Up to 95% recombinants • Minimal multiple cloning site, so you sequence more insert, less vector
Expression in *E. coli*	Champion™ pET100 series Directional TOPO® Expression Kits	Directional	• Up to 90% of clones in the correct orientation • High-level protein expression in *E. coli* • Inducible expression using IPTG • 6xHis tag for convenient purification and detection • EK cleavage sequence
Expression in mammalian cells	pcDNA™ 3.2/GW/D-TOPO® Cloning Kit	Directional	• Greater than 90% of clones in correct orientation • CMV promoter delivers high level expression in mammalian cells • C-terminal V5 and 6xHis tags for convenient detection and purification • Neomycin selection and purification • Neomycin selection
Entry into Gateway® expression systems	pCR® 8/GW/TOPO® TA Cloning® Kit	TA	• Maximum convenience: includes PureLink® Quick Plasmid Miniprep Kit • Fast-growing Mach1™ *E. coli* shortens cloning time
		TA	• Fast-growing *E. coli* shortens cloning time
	pENTR™/D-TOPO® Cloning Kit	Directional	• Fast-growing *E. coli* shortens cloning time

* All Cat. Nos. are for 20-reaction kits. For ordering information for additional sizes, visit www.invitrogen.com/topo.

	Competent *E. coli*	Cat. No.*
	One Shot® Mach1™ T1R Cells	K451020
	One Shot® TOP10 Cells	K450001
	One Shot® TOP10 Cells	K456001
	Electrocomp™ Cells	K283020
	One Shot® Mach1™ T1R Cells	K280020
	One Shot® TOP10 Cells	K286020
	One Shot® TOP10	K703020
	Electrocomp™ Cells	K475010
	One Shot® Mach1™ T1R Cells	K470010
	One Shot® TOP10 Cells	K461020
	One Shot® TOP10	K460001
	Electrocomp™ Cells	K466001
	One Shot® Mach1™ T1R Cells	K453020
	One Shot® TOP10 Cells	K457501
	One Shot® TOP10 Electrocomp™ Cells	K458001
	One Shot® Mach1™ T1R Cells	K283520
	One Shot® TOP10 Cells	K287520
	One Shot® TOP10 Electrocomp™ Cells	K288020
	BL21 Star™ (DE3) One Shot® and One Shot® TOP10 Cells	K10001
		K10101
		K10201
		K15101
	One Shot® TOP10 Cells	K244020
	One Shot® Mach1™ T1R Cells	K252002
	One Shot® Mach1™ T1R Cells	K252020
	One Shot® TOP10 Chemically Competent *E. coli*	K240020

Gateway® recombination cloning technology

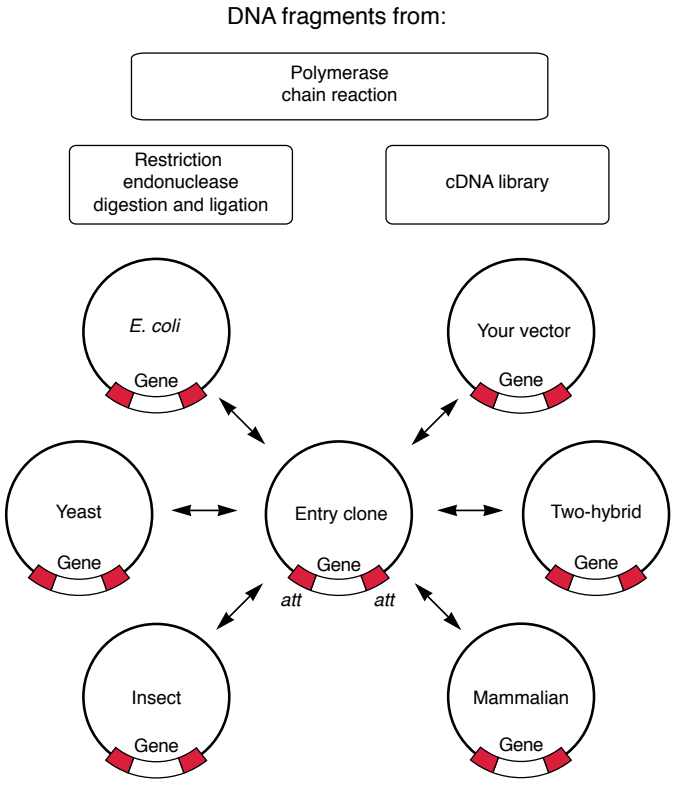
The typical cloning workflow involves many steps, particularly, traditional restriction enzyme cloning. This traditional method limits your cloning success. For example, certain restriction enzymes cannot be used because they might cut within your gene of interest, truncating the insert and making the gene useless for downstream expression. Additional clean-up steps are needed with this method, you experience low-efficiency recovery of recombinants from cloning large fragments, and you waste time screening colonies to find the clone you need—all of these steps take considerable time and effort and success is not guaranteed.

Amazingly versatile

In contrast, Gateway® recombination cloning technology circumvents these cloning limitations, enabling you to access virtually any expression system. Gateway® recombination cloning uses a one hour, 99%-efficient, reversible recombination reaction, without using restriction enzymes, ligase, subcloning steps, or screening of countless colonies, thereby helping you save time, money, and effort. Widely adopted in the research community with more than 1,500 references since its launch, Gateway® technology makes collaboration across research disciplines easy and convenient and enables access to a multitude of vectors from these research groups for truly multidisciplinary scientific studies. Finally, new advancements such as MultiSite Gateway® Technology makes our Gateway® cloning the ideal cloning method for protein expression and functional analysis.

Advantages of Gateway® technology

- Fast, one-hour, room-temperature cloning reactions with >99% efficiency deliver the clone you need
- Maintaining orientation and reading frame without using restriction enzymes or ligation makes expression-ready clones
- Eliminating resequencing helps ensure consistent results throughout your experiment using the same clone from target identification to validation
- Shuttling insert DNA from one expression vector to another affords flexibility while simplifying your cloning workflow



Rapidly move from one application to the next with Gateway® Technology.

How Gateway® technology works

Gateway® technology uses lambda phage–based site-specific recombination instead of restriction endonucleases and ligase to insert a gene of interest into an expression vector. Lambda recombination mediated by Gateway® Clonase® enzyme mixture is the foundation of Gateway® technology. Transferring a gene into a destination vector is accomplished in just two steps:

Step 1:

Clone your gene of interest into a Gateway® Entry vector. There are a number of ways to enter the Gateway® platform:

- TOPO® cloning vectors containing Gateway® *att* sites
- PCR-amplified fragments with primers containing *attB* sites
- Restriction-based cloning into Gateway® Entry vectors
- Purchase an Ultimate™ ORF clone already in a Gateway® vector

Step 2:

Mix the Entry clone containing the gene of interest *in vitro* with the appropriate Gateway® expression vector (Destination vector) and Gateway® LR Clonase® II enzyme mix.

Site-specific recombination between the *att* sites in each vector generates an expression clone with the gene of interest recombined into the Destination vector backbone, and a by-product. Following transformation and selection in *E. coli*, the expression clone is ready for expression in the appropriate host.

Ever-increasing expression options

A wide variety of Gateway® Destination vectors are available for protein expression and functional analysis experiments. From *in vitro* to bacterial to insect to mammalian to viral systems, there is a Gateway® vector for your application.

TOPO® TA Cloning® Kit for entry into Gateway® technology

Fast, effective cloning of *Taq*-amplified PCR products

- 3´-T overhangs for direct ligation of *Taq*-amplified PCR products
- Novel Gateway® primer sites for convenient sequencing
- EcoRI sites for easy excision of inserts

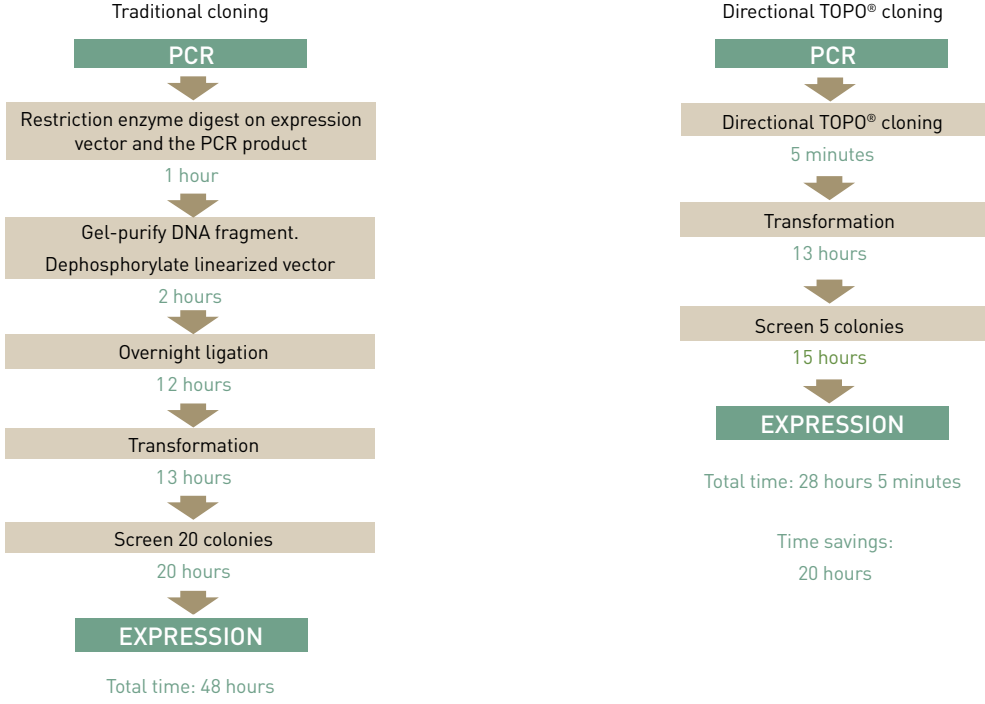
TOPO® TA Cloning® Kits are designed for cloning PCR products directly from a PCR reaction in as little as 5 minutes. The pCR®8/GW/TOPO® vector offers novel Gateway® primer sites within the *att* regions, located less than 55 base pairs from the PCR product insertion site, for convenient sequencing. This vector also incorporates streamlined sequence analysis so you sequence more of your insert than the vector, spectinomycin resistance gene for robust selection in *E. coli*, and EcoRI sites flanking the PCR product insertion site for easy excision of inserts.

Directional TOPO® cloning

The fastest method of entry into Gateway® technology

- Five-minute TOPO® cloning of PCR product
- >90% of recombinant clones in the correct orientation for expression
- Powerful promoter for high-level expression

Directional TOPO® cloning enables you to clone your blunt-end PCR products in 5´ to 3´ orientation into a proven expression vector using a 5-minute ligation reaction. The pENTR™/D-TOPO® Cloning Kits utilize a highly efficient, 5-minute cloning strategy ("TOPO® Cloning") to directionally clone a blunt-end PCR product into a vector for entry into the Gateway® System. Directional TOPO® cloning vectors contain a single-stranded GTGG overhang on the 5´ end and a blunt end on the 3´ end. The 4-nucleotide overhang invades the double-strand DNA of the PCR product and anneals to the CACC sequence that you place in your 5´ primer. Topoisomerase I then ligates the PCR product in the correct orientation. Once the PCR product is cloned into the Directional TOPO® Entry vector, the resulting entry clone can be recombined with any Gateway® Destination vector to create an expression clone.



Time comparison of cloning strategies.

Custom cloning services

Let the experts do the cloning for you

Save valuable time and enhance your research by letting us do your cloning work for you. We offer custom cloning services in the following areas: custom ORF cloning, TOPO® cloning adaptation of your vector, DNA fragment production, cloning and subcloning, cloning PCR products, Gateway® adaptation of your vector, Gateway® Entry clone cross into Destination vectors (L x R crosses), site-directed mutagenesis, custom cDNA library construction, and normalization and standardization.

ORF Cloning Services

Convenient access to unlimited downstream applications

- Full compatibility with Gateway® technology
- Adaptability to multiple Destination vectors
- Facilitated protein expression and functional analysis

The ORF Cloning Services offer a number of options. We can perform ORF cloning from a template that you provide or from an Invitrogen™ clone collection. PCR cloning is followed by cloning into a Gateway® Donor or TOPO® vector, and then full-length sequencing. You may also choose ORF cloning from a library. PCR cloning is performed from an Invitrogen™ cDNA library into a Gateway® Donor or TOPO® vector and then followed by end-read sequencing. The ORF Cloning Service can also transfer ORFs to a broad range of Gateway® expression vectors in bacterial, mammalian, yeast, lentiviral, adenoviral, baculoviral, and cell-free expression systems by an LR or BP recombination reaction.

Gateway® Vector Conversion Services

Powerful recombinational cloning

- Conversion
- Validation
- Delivery

The Gateway® Vector Conversion Service includes subcloning a Gateway® cassette into your vector of interest to create a Gateway® Destination vector. Validation of the vector is performed using a Gateway® LR reaction to show successful recombination, and sequencing to confirm the cassette is in frame with a tag, if one is present. The converted vector is delivered as a glycerol stock with vector maps and sequence reports.

Custom TOPO® Cloning Services

- Adaptation of any vector
- Prepared for 5-minute cloning with up to 95% recombinants
- No gel purification or post-PCR modifications needed

Our scientists will modify your favorite vector by covalently attaching the topoisomerase I enzyme and preparing the ends for either blunt or TA Cloning® methods. If you choose, we can also prepare the vector for directional cloning of PCR products. Whether you're cloning hundreds or thousands of PCR-amplified genes in your high-throughput (HTP) experiments, a TOPO® vector will save you a substantial amount of time. The Custom TOPO® Cloning Service includes functional testing of your vector in a TOPO® cloning reaction. Your modified vector is supplied in bulk for a minimum of 500 reactions, and high-efficiency competent cells are included, making the system perfect for HTP cloning.

For complete information on any of these services, go to www.invitrogen.com/customservices or email us at europeservices@invitrogen.com.

Restriction enzyme digestion and ligation

Enzymes for traditional cloning

- ExpressLink™ T4 DNA ligase for fast ligation
- Extensive selection of enzymes for cloning and subcloning
- Rigorous performance and quality-testing

We offer a number of enzymes needed to clone and subclone your gene of interest so you can achieve your goals in both simple and complex cloning projects. We recommend using SYBR® Safe DNA gel stain and the Safe Imager™ Blue-Light Transilluminator for safe DNA visualization that minimizes damage to your DNA samples.

Recommended universal buffer for double digestion.

Enzyme	AccI	BamHI	BglII	ClaI	EcoRI	EcoRV	HincII	HindIII	KpnI	NcoI
Supplied buffer	10X M	10X K	10X H	10X M	10X H	10X H	10X M	10X M	10X L	10X K +BSA
AccI	—	0.5X K	1X T	1X M	1X M	0.5X K	1X M	1X M	1X M	1X M +BSA
BamHI	0.5X K	—	1X K	1X K	1X K	1X K	0.5X K	1X K	0.5X K	1X K +BSA
BglII	1X T	1X K	—	1X H	1X H	1X H	2X K	1X K	1X T	1X K +BSA
ClaI	1X M	1X K	1X H	—	1X H	1X H	1X M	1X M	1X M	1X K +BSA
EcoRI	1X M	1X K	1X H	1X H	—	1X H	1X M	1X M	1X M	1X K +BSA
EcoRV	0.5X K	1X K	1X H	1X H	1X H	—	2X T	1X K	0.5X K	1X K +BSA
HincII	1X M	0.5X K	2X K	1X M	1X M	2X T	—	1X M	1X M	1X M +BSA
HindIII	1X M	1X K	1X K	1X M	1X M	1X K	1X M	—	1X M	1X K +BSA
KpnI	1X M	0.5X K	1X T	1X M	1X M	0.5X K	1X M	1X M	—	0.5X K +BSA
NcoI	1X M +BSA	1X K +BSA	1X K +BSA	1X K +BSA	1X K +BSA	1X K +BSA	1X M +BSA	1X K +BSA	0.5X K +BSA	—
NdeI	1X T	1X K	1X H	1X H	1X H	1X H	1X T	1X K	1X T	1X K +BSA
NotI	0.5X K +BSA	0.5X K +BSA	1X H +BSA	1X H +BSA	1X H +BSA	1X H +BSA	0.5X K +BSA	0.5X K +BSA	0.5X K +BSA	0.5X K +BSA
PstI	1X M	1X K	1X H	1X H	1X H	1X H	1X M	1X M	1X M	1X K +BSA
PvuI	0.5X K	1X K	1X K	1X K	1X K	1X K	0.5X K	1X K	0.5X K	1X K +BSA
SacI	1X M	0.5X K	0.5X K	1X M	1X M	0.5X K	1X M	1X M	1X L	0.5X K +BSA
SalI	1.5X T	1.5X T	1X H	1X H	1X H	1X H	1.5X K	1.5X K	1.5X T +BSA	1.5X T +BSA
SmaI	1X T +BSA	0.5X T +BSA	1X T +BSA	1X T +BSA	1X T +BSA	0.5X K +BSA	1X T +BSA	1X T +BSA	1X T +BSA	1X T +BSA
SpeI	1X M	1X K	1X H	1X M	1X H	1X H	1X M	1X M	1X M	1X K +BSA
SphI	0.5X K	1X K	1X H	1X H	1X H	1X H	2X T	1X K	0.5X K	1X K +BSA
XbaI	1X M	0.5X K	2X T	1X M	1X M	2X T	1X M	1X M	1X M	1X M +BSA
XhoI	1X M	1X K	1X H	1X H	1X H	1X H	1X M	1X M	1X M	1X K +BSA

Notes:

1. It is confirmed that 10 units of each enzyme completely digests 1 µg of DNA at 37°C in one hour in a 50-µL reaction mixture.
2. The concentration of glycerol should be less than 10% to minimize star activity.
3. DNA may not be digested completely when recognition sequences of two enzymes are close to one other or when DNA takes high-structure conformation.

Recommended universal buffer for double digestion

Double digestion (cutting a DNA with two restriction enzymes simultaneously) is widely used to save time. In this table, the suitable buffer type, dilution rate, and additive are shown to perform double digestion with the restriction enzymes shown below. The characters L, M, H, T, and K show the type of universal buffer recommended, which is supplied at 10X concentration. When "0.5X" is recommended, dilute 10X buffer 20-fold; when "1X" is recommended, dilute 10-fold; and when "2X" is recommended, dilute 5-fold. As BSA is also supplied at 10X concentration, dilute it 10-fold to a final concentration of 0.01% when used.

	NdeI	NotI	PstI	PvuI	SacI	SalI	SmaI	SpeI	SphI	XbaI	XhoI
	10X H	10XH +BSA +Triton	10X H	10X K +BSA	10X L	10X H	10X T +BSA	10X M	10X H	10X M +BSA	10X H
	1X T	0.5X K +BSA	1X M	0.5X K	1X M	1.5X T	1X T +BSA	1X M	0.5X K	1X M	1X M
	1X K	0.5X K +BSA	1X K	1X K	0.5X K	1.5X T	0.5X T +BSA	1X K	1X K	0.5X K	1X K
	1X H	1X H +BSA	1X H	1X K	0.5X K	1X H	1X T +BSA	1X H	1X H	2X T	1X H
	1X H	1X H +BSA	1X H	1X K	1X M	1X H	1X T +BSA	1X M	1X H	1X M	1X H
	1X H	1X H +BSA	1X H	1X K	1X M	1X H	1X T +BSA	1X H	1X H	1X M	1X H
	1X H	1X H +BSA	1X H	1X K	0.5X K	1X H	0.5X K +BSA	1X H	1X H	2X T	1X H
	1X T	0.5X K +BSA	1X M	0.5X K	1X M	1.5X K	1X T +BSA	1X M	2X T	1X M	1X M
	1X K	0.5X K +BSA	1X M	1X K	1X M	1.5X K	1X T +BSA	1X M	1X K	1X M	1X M
	1X T	0.5X K +BSA	1X M	0.5X K	1X L	1.5X T +BSA	1X T +BSA	1X M	0.5X K	1X M	1X M
	1X K +BSA	0.5X K +BSA	1X K +BSA	1X K +BSA	0.5X K +BSA	1.5X T +BSA	1X T +BSA	1X K +BSA	1X K +BSA	1X M +BSA	1X K +BSA
	—	1X H +BSA	1X H	1X K	1X T	1X H	1X T +BSA	1X H	1X H	1X T	1X H
	1X H +BSA	—	1X H +BSA	2X K +BSA	0.5X K +BSA	1X H +BSA	0.5X T +BSA	1X H +BSA	1X H +BSA	0.5X K +BSA	1X H +BSA
	1X H	1X H +BSA	—	1X K	1X M	1X H	0.5X T +BSA	1X H	1X H	1X M	1X H
	1X K	2X K +BSA	1X K	—	0.5X K	1.5X K +BSA	1X K +BSA	1X K	1X H	1X T	1X K
	1X T	0.5X K +BSA	1X M	0.5X K	—	1.5X T +BSA	1X T +BSA	1X M	0.5X K	1X M	1X M
	1X H	1X H +BSA	1X H	1.5X K +BSA	1.5X T +BSA	—	1.5X T +BSA	1X H	1X H	1.5X T	1X H
	1X T +BSA	0.5X T +BSA	0.5X T +BSA	1X K +BSA	1X T +BSA	1.5X T +BSA	—	1X T +BSA	0.5X T +BSA	1X T +BSA	1X T +BSA
	1X H	1X H +BSA	1X H	1X K	1X M	1X H	1X T +BSA	—	1X H	1X M	1X H
	1X H	1X H +BSA	1X H	1X K	0.5X K	1X H	0.5X T +BSA	1X H	—	2X T	1X H
	1X T	0.5X K +BSA	1X M	0.5X K	1X M	1.5X T	1X T +BSA	1X M	2X T	—	1X M
	1X H	1X H +BSA	1X H	1X K	1X M	1X H	1X T +BSA	1X H	1X H	1X M	—

Percent restriction enzyme activity in each reaction buffer

Restriction enzyme	Cut site	Buffer L	Buffer M	Buffer H	Buffer K	Buffer T (+ BSA)	Buffer
AccI	5′-GT↓(Py)(Pu) AC-3′ 3′-CA (Pu)(Py)↑TG-5′	20	100	<20	<20*	160	–
AfaI	5′-GT↓AC-3′ 3′-CA↑TG-5′	60	60	40	60	100	–
AluI	5′-AG↓CT-3′ 3′-TC↑AG-5′	100	120	<20	40	200	–
ApaI	5′-GGGCC↓C-3′ 3′-C↑CCGGG-5′	100	<20	<20	<20	<20	–
BalI	5′-TGG↓CCA-3′ 3′-ACC↑GGT-5′	20*	20*	<20*	<20*	40*	Ball buffer
BamHI	5′-G↓GATC C-3′ 3′-C CTAG↑G-5′	<20*	<20	40	100	<20*	–
BglII	5′-A↓GATC T-3′ 3′-T CTAG↑A-5′	<20	20	100	100*	60*	–
BmeT120 I (AvaI)	5′-C↓(Py)C G(Pu) G-3′ 3′-G (Pu)G C(Py)↑C-5′	<20	<20	20	100	<20	–
BshTI	5′-A↓AGCTT-3′ 3′-TT CGA↑A-5′	<20	20	80*	50	50	–
ClaI	5′-AT↓CGAT-3′ 3′-TAGC↑TA-5′	40	100	120	100	60	–
DdeI	5′-C↓TNA G-3′ 3′-G ANT↑C-5′	60	80	100	100	80	–
DpnI	5′-GmA↓TC-3′ 3′-C T↑mAG-5′	60*	100	<20	200	100*	–
DraI (AhaIII)	5′-TTT↓AAA-3′ 3′-AAA↑TTT-5′	100	100	60	100	80	–
EcoRI	5′-G↓AATT C-3′ 3′-C TTAA↑G-5′	20*	100*	100	120*	80*	–
EcoRV	5′-GAT↓ATC-3′ 3′-CTA↑TAG-5′	<20*	<20*	40*	100	120*	–
EcoT22 I (AvaII)	5′-ATGCA↓T-3′ 3′-T↑ACGTA-5′	<20	20	100	140*	20*	–
HaeIII	5′-GG↓CC-3′ 3′-CC↑GG-5′	60	100	100	60	100	–
HapII (HpaII)	5′-C↓CG G-3′ 3′-G GC↑G-5′	100	60	<20	<20	100	–
HhaI	5′-GCG↓C-3′ 3′-C↑GCG-5′	80	100	100	120	120	–
HincII (HindII)	5′-GT(Py)↓(Pu)AC-3′ 3′-CA(Pu)↑(Py)TG-5′	20	100	20	40	100	–
HindIII	5′-A↓AGCT T-3′ 3′-T TCGA↑A-5′	60*	100	<20	200	100*	–
HinfI	5′-G↓ANT C-3′ 3′-C T-↑G-5′	80	100	100	160	60	–
HpaI	5′-GTT↓AAC-3′ 3′-CAA↑TTG-5′	<20	40*	20	100	80*	–
KpnI	5′-GGTAC↓C-3′ 3′-C↑CATGG-5′	100	60	<20	<20	100*	–
MboI	5′-↓GATC-3′ 3′-CTAG↑-5′	20	40	60	100	40	–

Restriction enzyme	Cut site	Buffer L	Buffer M	Buffer H	Buffer K	Buffer T (+ BSA)	Buffer
MluI	5´-A↓CGCGT-3´ 3´-TGCGC↑A-5´	60	60	100	100*	60	–
NcoI	5´-C↓CATGG-3´ 3´-GGTAC↑C-5´	40*	60*	20	60*	60*	–
NdeI	5´-CA↓TATG-3´ 3´-GTAT↑AC-5´	<20	40	100	100	80	–
NheI	5´-G↓CTAGC-3´ 3´-CGATC↑G-5´	120*	100	<20	<20	160*	–
NotI	5´-GC↓GGCCGC-3´ 3´-CGCCGG↑CG-5´	<20*	<20*	20**	<20	<20*	–
NruI	5´-TCG↓CGA-3´ 3´-AGC↑GCT-5´	0*	<20*	20*	20*	<20*	NruI buffer
PstI	5´-C TGCA↓G-3´ 3´-G↑ACGT C-5´	<20*	60*	100	80	20*	–
PvuI	5´-CGAT↓CG-3´ 3´-GC↑TAGC-5´	<20*	20*	40*	80**	40*	–
PvuII	5´-CAG↓CTG-3´ 3´-GTC↑GAC-5´	80*	100	40	<20	40*	–
SacI	5´-GAGCT↓C-3´ 3´-C↑TCGAG-5´	100	60	<20	<20	80	–
SacII	5´-CCGC↓GG-3´ 3´-GG↑CGCC-5´	40	20	<20	<20	100	–
SalI	5´-G↓TCGA C-3´ 3´-C AGCT↑G-5´	<20	<20	100	20*	<20	–
ScaI	5´-AGT↓ACT-3´ 3´-TCA↑TGA-5´	<20*	<20*	100	60*	<20*	–
SmaI	5´-CCC↓GGG-3´ 3´-GGG↑CCC-5´	<20	<20	<20	<20	100	–
SpeI	5´-A↓CTAGT-3´ 3´-TGATC↑A-5´	80*	100	80	100	80*	–
SphI	5´-GCATG↓C-3´ 3´-C↑GTACG-5´	20*	40*	100	120	20*	–
SspI	5´-AAT↓ATT-3´ 3´-TTA↑TAA-5´	<20*	60*	40	100*	80*	SspI buffer
StuI	5´-AGG↓CCT-3´ 3´-TCC↑GGA-5´	60	100	60	80	140	–
TaqI (TthHB8 I)	5´-T↓CGA-3´ 3´-AGC↑T-5´	40	80	60	60	80	TaqI buffer
XbaI	5´-T↓CTAGA-3´ 3´-AGATC↑T-5´	<20	80*	20	<20	120	–
XhoI	5´-C↓TCGAG-3´ 3´-GAGCT↑C-5´	<20	60	100	160	60	–

* Weak star activity is detected
** 100% activity is obtained by addition of 0.01% BSA
For more information, visit www.invitrogen.com/restrictionenzymes.

Buffer compositions.

10X Buffer H	10X Buffer K	10X Buffer L	10X Buffer M	10X Buffer T (BSA-free)
• 500 mM Tris-HCl, pH 7.5 • 100 mM $MgCl_2$ • 10 mM dithiothreitol (DTT) • 1,000 mM NaCl	• 200 mM Tris-HCl, pH 8.5 • 100 mM $MgCl_2$ • 10 mM dithiothreitol (DTT) • 1,000 mM KCl	• 100 mM Tris-HCl, pH 7.5 • 100 mM $MgCl_2$ • 10 mM dithiothreitol (DTT)	• 100 mM Tris-HCl, pH 7.5 • 100 mM $MgCl_2$ • 10 mM dithiothreitol (DTT) • 500 mM NaCl	• 330 mM Tris acetate, pH 7.9 • 100 mM magnesium acetate • 5 mM dithiothreitol (DTT) • 660 mM potassium acetate

ExpressLink™ T4 DNA Ligase

ExpressLink™ T4 DNA Ligase is a T4 ligase that catalyzes the ligation of blunt or cohesive end DNA fragments in only 5 minutes at room temperature (25°C) and PCR fragments with "A" overhangs typically in 15 minutes, also at 25°C. T4 ligase catalyzes the formation of phosphodiester bonds between double-stranded DNA strands with 3´ hydroxyl and 5´ phosphate termini in the presence of ATP. ExpressLink™ T4 DNA Ligase formulation is optimized for fast ligase reaction times and a more convenient incubation temperature than our other formulations. Single-stranded nucleic acids are not substrates for this enzyme

- Fast, 5-minute ligation of blunt or cohesive end DNA fragments
- Fast ligation of PCR fragments with "A" overhangs typically in 15 minutes
- Convenient 25°C reaction temperature eliminates the need for water bath or temperature block.
- Ideal for cloning DNA into vectors, recircularization of linear DNA, library construction, TA cloning, and linker ligation
- Exonuclease free

See the difference the cleanest ligase makes

We apply the most stringent test available for exonuclease contamination to every lot of ExpressLink™ T4 DNA Ligase. The superior sensitivity of our assay will detect much lower active exonuclease concentrations than assays used by other manufacturers. As a result, we offer one of the best ligase products on the market (see figure). You can be more confident of your results with ExpressLink™ T4 DNA Ligase.

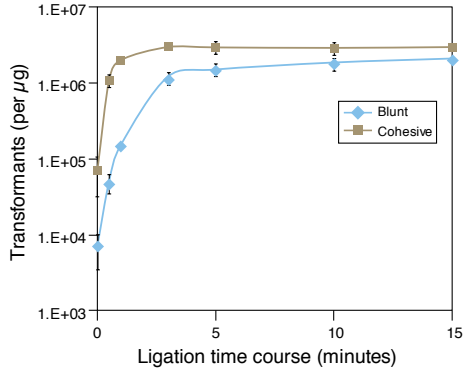


Figure 1. ExpressLink™ T4 DNA Ligase time course. LITMUS 28 vector was gel-purified after restriction digest with either EcoRV (blunt) or HindIII (cohesive) and treatment with calf intestinal alkaline phosphatase. Inserts from a HaeIII digest (blunt) of ϕX174 DNA and a HindIII digest (cohesive) of λ DNA were ligated into the respective vectors using ExpressLink™ T4 DNA Ligase. Ligation products were transformed into chemically competent DH10B cells and grown overnight on LB-Amp plates at 37°C.

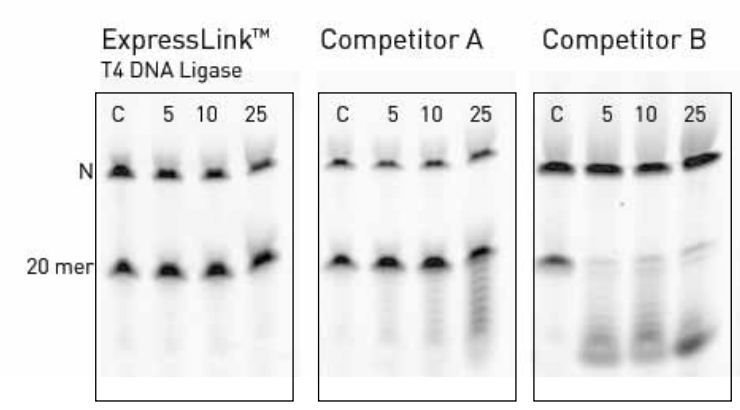


Figure 2. Exonuclease assay. A 5´-FAM-labeled 20-mer was mixed with 0, 5, 10, or 25 units of various ligase enzymes (ExpressLink™ T4 DNA Ligase, Competitor A, and Competitor B) and incubated at 37°C for 16 hours. The reaction was then mixed with a normalization marker (N), run on a TBE-urea gel, and imaged.

Product	Quantity	Cat. No.
ExpressLink™ T4 DNA Ligase	1 kit	A13726

BSA and modifying enzymes

Below is our selection of reagents for manipulating and modifying DNA for cloning, sequencing, expression, and other experimentation.

Product	Quantity	Cat. No.
Bovine Serum Albumin	150 mg	15561020
AcTEV™ Protease, 10 units/µL	1,000 units	12575015
	10,000 units	12575023
Bacterial Alkaline Phosphatase, 150 units/µL	2,500 units	18011015
Calf Intestinal Alkaline Phosphatase, 1 unit/µL	1,000 units	18009027
Calf Intestinal Alkaline Phosphatase, 20 units/µL	1,000 units	18009019
DNA Polymerase I, 10 units/µL	250 units	18010017
	1,000 units	18010025
DNA Polymerase I, Large (Klenow) Fragment, 3–9 units/µL	100 units	18012021
	500 units	18012039
DNA Polymerase I/DNase I	250 units	18162016
DNase I , 50–375 units/µL	20,000 units	18047019
DNase I, Amplification Grade, 1 unit/µL	100 units	18068015
E. coli DNA Ligase, 10 units/µL	100 units	18052019
EK-Away™ Resin	7.5 mL	R18001
EKMax™ Enterokinase	250 units	E18001
	1,000 units	E18002
λ Exonuclease, 1–10 units/µL	150 units	28023018
Proteinase K	100 mg	25530015
Proteinase K (Solution), 20 mg/mL	5 mL	25530049
Ribonuclease H, 2 units/µL	30 units	18021014
	4 x 30 units	18021071
Ribonuclease Inhibitor, Cloned, 10 units/µL	1,000 units	15518012
RNaseOUT™ Recombinant Ribonuclease Inhibitor, 40 units/µL	5,000 units	10777019
S1 Nuclease, 400–1,500 units/µL	20,000 units	18001016
SP6 RNA Polymerase, 15 units/µL	500 units	18018010
SUMO Protease (10X), 1 unit/µL	250 units	12588018
T4 DNA Ligase, 5 units/µL	250 units	15224041
T4 DNA Ligase, 1 unit/µL	100 units	15224017
	500 units	15224025
ExpressLink™ T4 DNA Ligase	1 kit	A13726
T4 DNA Polymerase, 5 units/µL	50 units	18005017
	250 units	18005025
T4 Polynucleotide Kinase, 10 units/µL	200 units	18004010
T7 RNA Polymerase, 50 units/µL	2,500 units	18033019
Terminal Deoxynucleotidyl Transferase, Recombinant, 15 units/µL	500 units	10533065
Topoisomerase I, 5–15 units/µL	500 units	38042024
Uracil DNA Glycosylase, 1 unit/µL	100 units	18054015

Overview of *E. coli* competent cells

We offer competent *E. coli* host strains with optimized genotypes for specialized cloning applications. A range of transformation efficiencies are available in convenient packaging formats. All competent cells are manufactured to strict quality control standards to help ensure you achieve the highest transformation efficiencies possible.

Optimized host strains

Several host strains offer distinct genotypic advantages for use in particular applications. For example, the DH10B™ and TOP10 strains include many desirable genetic markers, as well as enhanced genomic DNA cloning capabilities required in general cloning. The DH5α™ strain and its derivatives offer several useful genetic markers and high transformation efficiencies, making them a popular, versatile strain.

A range of efficiencies

The transformation efficiency required for success depends largely on the application to be performed. Therefore, Invitrogen™ competent cells are available in a wide range of efficiencies. Applications where the DNA is very limited or where the maximum number of clones is critical, such as cDNA library construction, require the highest-efficiency strains such as ElectroMAX™ and OmniMAX™ strains. These cell lines provide efficiencies in the range of 5 x 10^9 to 1 x 10^{10} cfu/µg. For routine cloning applications, efficiencies from 1 x 10^6 to 1 x 10^9 cfu/µg are available. Transformation efficiencies are defined as colony forming units/µg pUC19.

Transformation efficiencies of our competent cells.

Product	Description
ElectroMAX™ Competent Cells	Electrocompetent cells offering the highest transformation efficiency available; >1 x 10^{10} transformants/µg of control DNA in a 20 µL reaction are guaranteed. A high-voltage electroporation apparatus capable of generating field strengths of 16 kV/cm is required.
OmniMAX™ Competent Cells	OmniMAX™ cells are the highest-efficiency chemically competent cells available. These cells yield >5 x 10^9 transformants/µg control DNA in a 50 µL transformation reaction. Use when efficiency is critical but no electroporator is available.
MAX Efficiency® Competent Cells	High-efficiency chemically competent cells. For most strains, guaranteed >1 x 10^9 transformants/µg control DNA per 100 µL reaction.
Library Efficiency® Competent Cells	Ideal for difficult clone constructions. They yield >1 x 10^8 transformants/µg control DNA per 100 µL reaction.
Subcloning Efficiency™ Competent Cells	Subcloning Efficiency™ Competent Cells are ideal for routine subcloning procedures or any application where the starting DNA is not limiting. These economically priced cells yield >1 x 10^6 transformants/µg control DNA per 50 µL reaction.

Flexible packaging formats

To meet the unique requirements of different projects, we offer a variety of packaging formats.

Competent cells are available in a variety of formats.

Format	Chemically competent	Electro-competent	Single-use	High-throughput	Volume	Advantage
One Shot®	•	•	•		50 µL	Transformation and recovery in the same tube
MultiShot™ StripWell	•			•	50 µL per well	12 strips of 8 tubes; do as many or as few reactions as needed
MultiShot™	•			•	15 µL per well	96-well plates fit automated format
Standard	•	•			100 µL, 200 µL, or 500 µL	Economical option

Have it your way with a custom kit

Custom kits with any competent cell can be packaged in any of the above formats to meet your particular needs. Custom strains can also be made available. Contact your local Account Manager or email customcompcells@invitrogen.com to learn more.

For detailed information about all competent cells, go to www.invitrogen.com/compcells for detailed .

Choose the right host strain attributes for your application

We offer competent cells for everything from cloning to expression. The table below summarizes the strains currently available.

Competent cells are available for everything from cloning to protein expression.

High-efficiency cloning (chemically competent)	High-efficiency cloning (electro-competent)	High-throughput cloning	Fast growth	Routine cloning	cDNA and genomic library construction
One Shot® OmniMAX™ 2 T1R	MegaX DH10B™ T1R Electrocomp™	MultiShot™ StripWell Mach1™ T1R	One Shot® Mach1™ T1R	One Shot® TOP10	MegaX DH10B™ T1R Electrocomp™
One Shot® MAX Efficiency® DH10B™ T1R	ElectroMAX™ DH10B™ T1R	MultiShot™ TOP10		MAX Efficiency® DH5α™	One Shot® OmniMAX™ 2 T1R
One Shot® MAX Efficiency® DH5α™ T1R	ElectroMAX™ DH5α-E™			Library Efficiency® DH5α™	ElectroMAX™ DH10B™ T1R
One Shot® TOP10	ElectroMAX™ DH10B™			Subcloning Efficiency™ DH5α™	
One Shot® TOP10F´	One Shot® TOP10 Electrocomp™				
MAX Efficiency® DH10B™	TOP10F´ Electrocomp™				
MAX Efficiency® DH5α™	TOP10 Electrocomp™				

Cloning unstable DNA	ssDNA production	Preparing unmethylated DNA	Recombinant baculovirus production	Propagating vectors with *ccdB* gene	Protein expression
ElectroMAX™ Stbl4™	ElectroMAX™ DH12S™	One Shot® INV110	MAX Efficiency® DH10Bac™	One Shot® *ccdB* Survival™ 2 T1R	BL21 Star™ (DE3) One Shot®
One Shot® Stbl3™	One Shot® TOP10F´			Library Efficiency® DB3.1™	BL21-AI™ One Shot®
MAX Efficiency® Stbl2™	One Shot® OmniMAX™ 2 T1R				BL21 (DE3) One Shot®
	One Shot® INVαF´				BL21(DE3)pLysS One Shot®
	MAX Efficiency® DH5αF´ IQ™				BL21(DE3)pLysE One Shot®

For detailed information about all competent cells, go to www.invitrogen.com/compcells.

Robust cloning with chemically competent cells

Transformation efficiency. Genetic markers. Packaging formats that fit your workflow. These are the most important considerations when choosing a competent cell for your cloning experiment. Each of these factors will directly impact the time and effort required by your project, as well as the success. That's why we offer you a number of cell types and formats. The following pages discuss these factors and the attributes inherent to each cell line, helping you make the right choice so you can achieve your cloning goals.

Transformation efficiencies for a variety of applications

The transformation efficiency you need will be largely determined by your application. To meet these different requirements, competent cells are available in a wide range of transformation efficiencies—from >1 x 10^6 to >3 x 10^{10} cfu/µg.

OmniMAX™ 2 T1^R *E. coli*—high-efficiency chemically competent cells for cloning and subcloning

The OmniMAX™ 2 T1^R *E. coli* strain is an improved chemically competent cell line, perfect for use in all cloning applications, including Gateway® technology. It offers one of the highest transformation efficiencies of any chemically competent cell available, with >5 x 10^9 transformants/µg pUC19.

Mach1™ T1^R *E. coli* strain—the fastest-growing chemically competent strain

The Mach1™ T1^R *E. coli* strain is the fastest-growing chemically competent strain currently available. Clearly visible colonies typically within eight hours of plating the transformation mix (ampicillin selection only), and perform minipreps after as little as 4 hours of growth. Mach1™ T1^R cells also benefit from T1 phage resistance to protect your samples.

Other cells include:

- DH10B™ T1^R cells—the most frequently cited competent cell on the market
- DH5α™ cells—designed for general cloning procedures
- TOP10 cells—genetically similar to the reliable DH10B™ strain available in multiple formats and many cloning kits

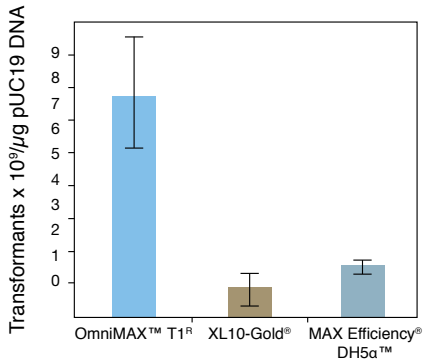


Figure 1. Highest-efficiency chemically competent cells available.

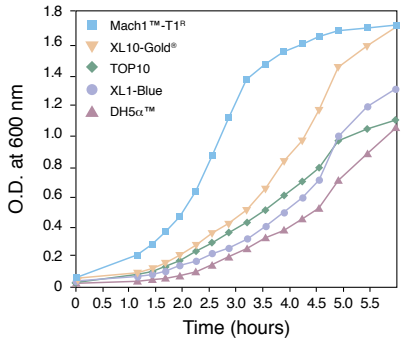


Figure 2. Mach1™ T1^R *E. coli* is the fastest-growing chemically competent strain.

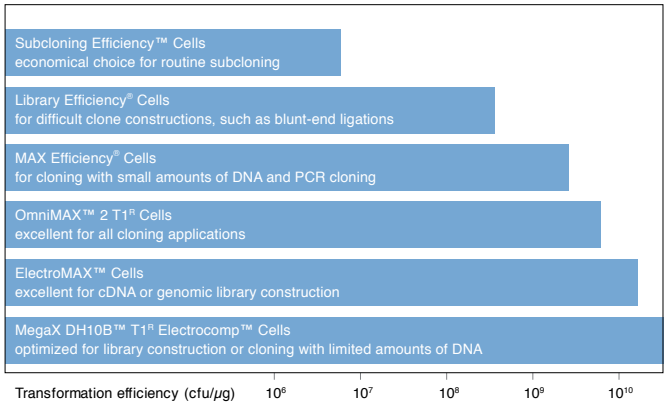


Figure 3. A range of transformation efficiencies are available to meet your every need. One Shot® cells are generally provided at >1 x 10^9 or >1 x 10^8 cfu/µg. A range of transformation efficiencies are available to meet your application needs.

For information on our complete product, offering go to www.invitrogen.com/compcells.

High-efficiency cloning using electroporation

MegaX DH10B™ T1R Electrocomp™ Cells

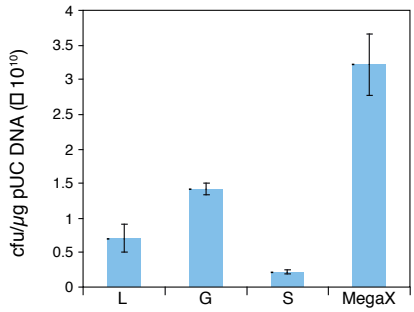
MegaX DH10B™ T1R Electrocomp™ Cells are the highest-efficiency electrocompetent cells available with a guaranteed three-fold greater number of colonies per transformation (>3 x 10^{10} cfu/µg pUC DNA). They are ideal for highly demanding cloning and library construction applications. MegaX DH10B™ T1R Cells have the same genotype as the widely used DH10B™ T1R strain, including tonA, which prevents infection by T1 and T5 lytic bacteriophages and safeguards your valuable clones and libraries.

Other cells include:

- ElectroMAX™ DH10B™ T1R Competent *E. coli*—high-efficiency electrocompetent cells derived from the widely used DH10B™ strain, includes the *tonA* genotype to prevent T1 infection and safeguard your clones and libraries
- ElectroMAX™ DH5α-E™ Cells—derived from the DH5α™ strain, suitable for transformation by electroporation

Competent cells for specialized applications

In addition to cells for cloning, we offer a number of cell lines for specific applications. Cell types have been optimized to achieve the highest performance in its noted experiment.



MegaX DH10B™ T1R Electrocomp™ Cells consistently outperform the competition.

Cloning unstable DNA

- Stbl3™ *E. coli* strain—designed for cloning direct repeats found in lentiviral expression vectors
- MAX Efficiency® Stbl2™ cells—specifically designed for cloning unstable inserts
- ElectroMAX™ Stbl4™ cells—electroporation cells, specifically designed for cloning unstable inserts

Single-stranded DNA (ssDNA) production

- DH12S™ cells—used for DNA sequencing, preparation of strand-specific probes, *in vitro* mutagenesis, and subtraction library applications

Propagating vectors with the *ccdB* gene

- One Shot® *ccdB* Survival™ 2 T1 phage-resistant (T1R) cells—designed for use with the Gateway® Vector Conversion System and for propagating Gateway® Destination, donor, and supercoiled entry vectors

Recombinant baculovirus production

- MAX Efficiency® DH10Bac™ competent cells—used for production of a recombinant bacmid in the Bac-to-Bac® Baculovirus Expression System

Competent cells for protein expression

E. coli is one of the most popular hosts for overexpression of recombinant proteins because it grows fast, is inexpensive to use, and yields high levels of protein. The most popular strains for recombinant protein expression from T7 expression systems are BL21 and its derivatives. BL21 strains have two important attributes that make them great for protein expression: key genetic markers and inducibility of protein expression.

Competent cells commonly used for protein expression from T7 promoter–containing vectors.

Product	Application	Efficiency (cfu/µg)	Quantity	Cat. No.
BL21 Star™ (DE3) One Shot® Cells	Extremely high expression of nontoxic proteins	>1 x 10^8	20 x 50 µL	C601003
BL21-AI™ One Shot® Cells	Tight regulation and strong expression of toxic proteins	>1 x 10^8	20 x 50 µL	C607003
BL21(DE3) One Shot® Cells	Expression of nontoxic proteins	>1 x 10^8	20 x 50 µL	C600003
BL21(DE3)pLysS One Shot® Cells	Expression of toxic or insoluble proteins	>1 x 10^8	20 x 50 µL	C606003
BL21(DE3)pLysE One Shot® Cells	Expression of toxic or very insoluble proteins	>1 x 10^7	20 x 50 µL	C656503

Competent cells ordering information

Product	Transformation efficiency (cfu/µg)	Quantity	Cat. No.
High-efficiency cloning, routine cloning, and subcloning			
One Shot® OmniMAX™ 2 T1^R Chemically Competent Cells	>5 x 10^9	20 x 50 µL	C854003
One Shot® Mach1™ T1^R Chemically Competent Cells	>1 x 10^9	20 x 50 µL	C862003
One Shot® TOP10 Chemically Competent Cells	>1 x 10^9	10 x 50 µL 20 x 50 µL 40 x 50 µL	C404010 C404003 C404006
One Shot® MAX Efficiency® DH10B™ T1^R Chemically Competent Cells	>1 x 10^9	20 x 50 µL	12331013
One Shot® MAX Efficiency® DH5α™ T1^R Chemically Competent Cells	>1 x 10^9	20 x 50 µL	12297016
MAX Efficiency® DH5α™ Chemically Competent Cells	>1 x 10^9	5 x 200 µL	18258012
Library Efficiency® DH5α™ Chemically Competent Cells	>1 x 10^8	5 x 200 µL	18263012
Subcloning Efficiency™ DH5α™ Chemically Competent Cells	>1 x 10^6	4 x 500 µL	18265017
cDNA or genomic library construction using electroporation			
MegaX™ DH10B™ T1^R Electrocomp™ Cells	>3 x 10^{10}	25 x 50 µL	C640003
ElectroMAX™ DH10B™ T1^R Electrocomp™ Cells	>1 x 10^{10}	5 x 100 µL	12033015
ElectroMAX™ DH10B™ Electrocomp™ Cells	>1 x 10^{10}	5 x 100 µL	18290015
ElectroMAX™ Stbl4™ Electrocomp™ Cells	>5 x 10^9	5 x 100 µL	11635018
Genomic and cDNA library construction using chemically competent cells			
One Shot® OmniMAX™ 2 T1^R Cells	>5 x 10^9	20 x 50 µL	C854003
High-throughput cloning			
MultiShot™ StripWell Mach1™ T1^R Cells	>1 x 10^9	1 plate	C869601
MultiShot™ StripWell TOP10 Chemically Competent Cells	>1 x 10^9	1 plate 5 plates	C409601 C40005
Cloning unstable DNA			
ElectroMAX™ Stbl4™ Electrocomp™ Cells	>5 x 10^9	5 x 100 µL	11635018
One Shot® Stbl3™ Chemically Competent Cells	>1 x 10^8	20 x 50 µL	C737303
MAX Efficiency® Stbl2™ Chemically Competent Cells	>1 x 10^9	5 x 200 µL	10268019
Single-stranded DNA production			
ElectroMAX™ DH12S™ Electrocomp™ Cells	>1 x 10^{10}	5 x 100 µL	18312017
MAX Efficiency® DH5αF´IQ™ Chemically Competent Cells	>1 x 10^8	5 x 200 µL	18288019
Propagating unmethylated DNA			
One Shot® INV110 Chemically Competent Cells	>1 x 10^6	20 x 50 µL	C717103
Propagating vectors with the *ccdB* gene vectors			
One Shot® *ccdB* Survival™ 2 T1^R	>1 x 10^9	10 x 50 µL	A10460
Library Efficiency® DB3.1™ Chemically Competent Cells	>1 x 10^8	5 x 200 µL	11782018
Recombinant baculovirus production			
MAX Efficiency® DH10Bac™ Competent Cells	>1 x 10^8	5 x 100 µL	10361012
Protein expression			
BL21-AI™ One Shot® Chemically Competent Cells	>1 x 10^8	20 x 50 µL	C607003
BL21 Star™ (DE3) One Shot® Chemically Competent Cells	>1 x 10^8	20 x 50 µL	C601003
BL21 Star™ (DE3)pLysS One Shot® Chemically Competent Cells	>1 x 10^8	20 x 50 µL	C602003
One Shot® BL21(DE3) Cells	>1 x 10^8	20 x 50 µL	C600003
One Shot® BL21(DE3)pLysS Cells	>1 x 10^8	10 x 50 µL	C606010
One Shot® BL21(DE3)pLysS Cells	>1 x 10^8	20 x 50 µL	C606003
One Shot® BL21(DE3)pLysE Cells	>1 x 10^7	20 x 50 µL	C656503

imMedia™ growth medium

Microwavable, premixed, low-salt LB medium

- No weighing or mixing of media components
- No autoclaving
- No waiting for media to cool before adding antibiotics

imMedia™ medium is a premixed, presterilized *E. coli* growth medium that can be prepared typically in just five minutes without autoclaving. imMedia™ growth medium contains everything you need—antibiotics, IPTG, and X-gal—to prepare low-salt LB medium or agar plates. You'll save hours of medium preparation time because imMedia™ medium offers:

Fast medium preparation

Simply mix imMedia™ growth medium with water in a clean flask, heat in a microwave oven, and typically in less than 5 minutes, your medium is prepared.

The ultimate convenience

imMedia™ growth medium is available for preparing liquid medium or agar plates, with or without IPTG and X-gal. You also have a choice of three antibiotics: ampicillin, kanamycin, or Zeocin™ selection agent. imMedia™ growth medium is packaged in ready-to-use individual pouches. Each pouch contains sufficient reagents for preparing 200 mL of liquid media or 8–10 standard agar plates.

Quality results

Each type of imMedia™ growth medium is extensively tested to help ensure *E. coli* growth, antibiotic activity, and sterility. imMedia™ Blue products are tested for efficient blue/white colony screening.

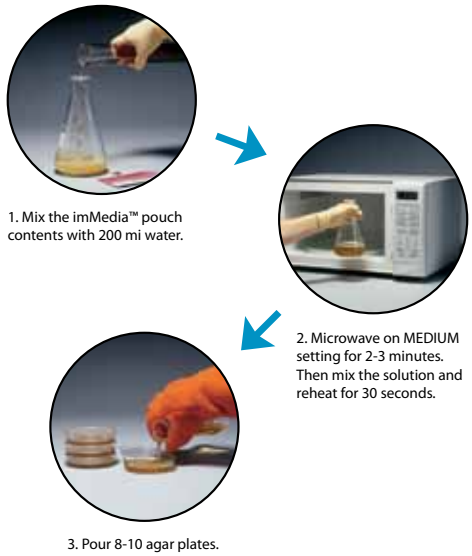


Figure 1. imMedia™ plates provide robust growth and effective selection.

Figure 2. TOP10 *E. coli* were plated on imMedia™ plates. The plates were incubated at 37°C for 16 hours. **(A)** TOP10 *E. coli* grown on an imMedia™ Amp Agar plate (negative control). **(B)** Amp^R TOP10 *E. coli* grown on an imMedia™ Amp Agar plate. **(C)** Amp^R *Lac*Z + TOP10 *E. coli* grown on an imMedia™ Amp Blue plate.

Product	Quantity	Cat. No.
For the preparation of liquid medium		
imMedia™ Amp Liquid	20 pouches	Q60020
imMedia™ Kan Liquid	20 pouches	Q61020
imMedia™ Zeo Liquid	20 pouches	Q62020
For the preparation of agar plates		
imMedia™ Amp Agar	20 pouches	Q60120
imMedia™ Kan Agar	20 pouches	Q61120
imMedia™ Zeo Agar	20 pouches	Q62120
For the preparation of agar plates with IPTG and X-gal		
imMedia™ Amp Blue	20 pouches	Q60220
imMedia™ Kan Blue	20 pouches	Q61220
Selection agents		
Zeocin™ antibiotic (100 mg/mL)	10 mL	R25001
	50 mL	R25005
X-gal	100 mg	15520034
	1g	15520018
Bluo-gal	1 g	15519028
IPTG	1 g	15529019

RNAi, epigenetics, and non-coding RNA research

5

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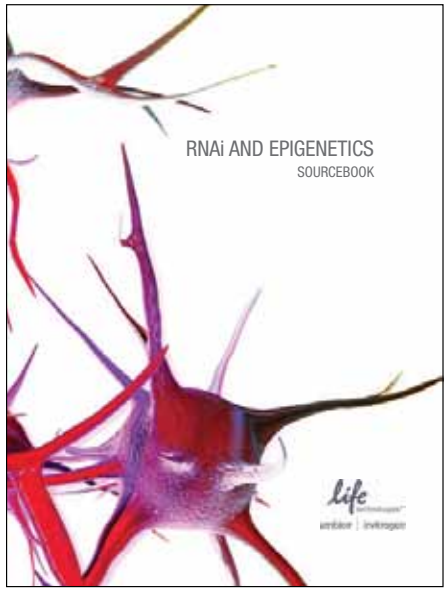
Overview of RNAi, epigenetics, and non-coding RNA research

The human genome contains all the information to determine who you are, how you look, and how you behave; however, this information is frequently further influenced by gene regulation processes such as epigenetics and regulation by non-coding RNAs (ncRNAs) such as siRNAs. As an example, many complex diseases have been linked to genetic variation in regions that control activity of genes, rather than in the regions that specify the protein code.

As a result, techniques enabling the study of gene regulation are becoming essential tools for studying growth, development, and differentiation of normal or diseased cells. These techniques allow better understanding of how, when, and where genes are activated or suppressed, and may provide the opportunity to define an entirely new family of biomarkers, define new mechanisms of regulation, or develop better diagnosis and treatment of disease.

Life Technologies has products specifically designed to study RNA interference (RNAi), epigenetics, and non-coding RNA as described here. As this area of research is currently quite dynamic, please go to www.invitrogen.com for the latest products and protocols.



RNAi interference

- Large dsRNA processed to siRNA or naturally occurring miRNAs that bind to mRNA and silences gene expression
- Directs gene expression; valuable experimental tool to silence genes *in vitro* and *in vivo*

Non-coding RNA and miRNAs

- Long and small non-coding RNAs; often processed into smaller RNAs (miRNAs)
- May control chromosome structure, epigenetic memory, transcription, RNA splicing and editing, mRNA translation and stability, protein stability and transport

Epigenetics

- DNA methylation, chromatin remodeling, and other changes in DNA structure, which are inherited
- Alters gene expression, forms basis of chromatin structure; DNA hypermethylation seen in many diseases including cancer

Overview of RNAi

Two types of small RNA molecules function in RNAi. The first are synthetic, short interfering RNA (siRNA) molecules that target mRNA cleavage effectively knocking down expression of a gene of interest. MicroRNA (miRNA) molecules are natural and regulate gene expression by binding to the 3´ untranslated regions (UTRs) of target mRNAs to inhibit their function. There are several ways to induce RNAi: synthetic molecules, RNAi vectors, and *in vitro* dicing. Your choice of tools depends on your model system, the length of time you require knockdown, and many other experimental parameters. Use the chart below to help you decide which approaches best meet your experimental needs.

Choosing an RNAi approach.

RNAi approach		Experimental system	Products		
			siRNAs	siRNA controls	siRNA delivery
siRNA analysis	RNA-based (synthetic siRNA)	Cell culture	•*Silencer*® Select Validated siRNAs •*Silencer*® Select Predesigned siRNAs •*Silencer*® Select siRNA Libraries •*mir*Vana™ miRNA Mimics and Inhibitors (page 215)	•*Silencer*® Select GAPDH Positive Control siRNA •*Silencer*® Select Negative Control siRNAs	•Lipofectamine® RNAiMAX Transfection Reagent •Lipofectamine® LTX Transfection Reagent •Neon® Transfection System
		In vivo	•Ambion® *In Vivo* siRNAs •*mir*Vana™ miRNA Mimics and Inhibitors (page 215)	•Ambion® *In Vivo* Positive and Negative siRNA Controls	•Invivofectamine® 2.0 Reagent
	DNA-based (vector)	Cell culture	•BLOCK-iT™ Lentiviral Pol II miR RNAi Expression System •BLOCK-iT™ Inducible H1 Lentiviral RNAi System •BLOCK-iT™ Adenoviral RNAi Expression System	•Controls included	•Lipofectamine® LTX Transfection Reagent •Neon® Transfection System
	Experimental objective		Products		
			miRNAs	miRNA controls	miRNA delivery
miRNA analysis	Gain of function (*in vitro or in vivo*)		•*mir*Vana™ miRNA Mimics	•*mir*Vana™ miRNA Mimics, Positive and Negative Controls	•Lipofectamine® RNAiMAX Transfection Reagent •Neon® Transfection System
	Loss of function (*in vitro or in vivo*)		•*mir*Vana™ miRNA Inhibitors	•*mir*Vana™ miRNA Inhibitors, Positive and Negative Controls	•Lipofectamine® RNAiMAX Transfection Reagent •Neon® Transfection System
				miRNA isolation	miRNA detection
	miRNA analysis			•Ambion® *mir*Vana™ miRNA Isolation Kit •Ambion® *mir*Vana™ PARIS™ Isolation Kit •TRIzol® Reagent •RecoverAll™ Total Nucleic Acid Isolation Kit	•SOLiD® Total RNA Sequencing Kit •TaqMan® Micro RNA, pri-MicroRNA, and ncRNA Assays •NCode™ miRNA Microarrays (see website)

For more details, go to www.invitrogen.com.

Silencer® Select siRNAs

The best siRNAs for *in vitro* applications

The benefits of using *Silencer*® Select siRNAs include:

- Potent—up to 100-fold more potent than currently available siRNAs and fewer off-target effects
- Most Specific—LNA® chemical modifications reduce off-target effects by up to 90%
- Reliable—demonstrated improvements in consistency and reliability of phenotypic results
- Guaranteed—100% guaranteed to silence, the best guarantee in the industry

The *Silencer*® Select siRNAs are classic 21-mers which incorporate the latest improvements in siRNA design, off-target effect prediction algorithms, and chemistry. Current siRNA design algorithms predict siRNAs that induce 70% target mRNA knockdown with only ~80% confidence. The *Silencer*® Select siRNA design algorithm was developed using a powerful machine learning method, which incorporates more than 90 different sequence and thermodynamic parameters. The result is siRNAs that are up to 100-fold more potent than other siRNAs (modified and unmodified), allowing a higher percentage of "on-target" phenotypes (see figure).

A choice of *Silencer*® Select siRNAs are available—Predesigned or Validated, as described on the following pages. *Silencer*® Select Predesigned siRNAs are guaranteed to silence based on their proven design, while the *Silencer*® Select Validated siRNAs are functionally tested to reduce target gene expression. Sets of *Silencer*® Select siRNA Libraries (page 194) are also available in tubes or plate for high-throughput screening. The *Silencer*® Select Positive and Negative Control RNAs (page 194) should be included in every siRNA experiment.

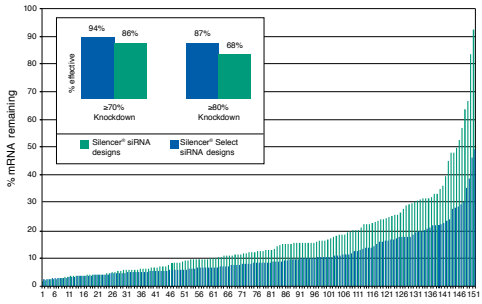
Silencer® Select Validated siRNAs

Silencer® Select Validated siRNAs are individual siRNA duplexes that have been verified experimentally to reduce the expression of their individual target genes. Each siRNA was designed using the same effective algorithm used to design *Silencer*® Select Predesigned siRNAs. However, each one has also been functionally confirmed and is guaranteed to reduce target gene expression by at least 80% when measured 48 hours post-transfection.

All *Silencer*® Select Validated siRNAs are annealed and provided ready to use. The included data sheet indicates the extent of mRNA knockdown observed during validation, the exon targeted by the siRNA, and full siRNA sequence information.

Silencer® Select Predesigned siRNAs

Silencer® Select Predesigned siRNAs, which are 100% guaranteed to silence their intended target, are immediately available for >98% of genes in the human, mouse, and rat genomes in a purified, ready-to-use format. siRNA sequence information is always provided.



***Silencer*® Select siRNA design algorithm significantly improves effective siRNA prediction accuracy.** The *Silencer*® Select siRNA design algorithm was used to design 155 siRNAs to 40 different targets. These siRNAs were tested side by side with siRNAs designed using the previous algorithm at 5 nM in HeLa cells. mRNA knockdown was measured 48 hr post-transfection via qRT-PCR using TaqMan® Gene Expression Assays. Results are expressed as percent of mRNA remaining compared to *Silencer*® Negative Control #1 siRNA–treated cells. The inset shows the percentage of siRNAs that elicited ≥70% and ≥80% mRNA knockdown.

TaqMan® siRNA Assays

TaqMan® siRNA Assays, sharing the same principle as miRNA assays (page 213), quantitate siRNAs with the specificity and sensitivity of TaqMan® Assay chemistry. A simple two-step protocol requires only reverse transcription with a siRNA-specific primer, followed by real-time PCR with TaqMan® probes. Key product features:

- Highly specific
- Sensitive—conserve limited samples: require only 1–10 ng of total RNA or equivalent
- Fast, simple and scalable—two-step quantitative RT-PCR assay provides high-quality results in less than 3 hours

TaqMan® siRNA Assays	Cat. No.	No. of RT/PCR reactions
Extra Small (XS)	4440877	25/75
Small (S)	4440878	50/150
Medium (M)	4440879	750/750
Large (L)	4440880	2,900/2,900

Silencer® Select siRNA Libraries

Some of the most popular siRNA collections are available in 96-well or 384-well plates (or tubes). The libraries enable high-throughput analysis of phenotypes resulting form target gene silencing. Three or four individual siRNAs are supplied for each target; they are not pooled, so that results are more easily discerned. Libraries are available as defined sets ready to ship, or may be customized for your research project.

Quality Control

Silencer® Select siRNA are synthesized in state-of-the-art facilities to meet the highest quality standards. As part of our rigorous quality control procedures, each RNA oligonucleotide is analyzed by MALDI-TOF mass spectrometry, and analytical HPLC is used to monitor purity. To provide the utmost in quality, each annealed siRNA is also assessed by gel electrophoresis to confirm that the strands anneal properly. The result is premium-quality siRNA that is purified and ready to use.

Custom siRNA libraries

The ultimate in flexibility

Do you have a favorite list of human genes you would like to target with a collection of siRNAs? We can prepare a custom *Silencer*® Select siRNA Library to any set of human genes. The minimum order is only 36 siRNAs, and you specify the siRNA layout on either 96- or 384-well plates. Multiple aliquots, pooling of siRNAs, and even custom siRNA design requests can all be accommodated. For additional information, or to discuss your research with one of our technical experts, contact your local sales representative or email us at rnairesearcher@invitrogen.com.

Silencer® Select GAPDH Positive Control siRNA

This validated positive control siRNA targets human, mouse, and rat GAPDH and is an ideal "test" siRNA for those just beginning siRNA experiments. In addition, because it targets GAPDH mRNA, which is commonly used as an internal control, its effects are easy to assay, and thus it provides an excellent tool to monitor siRNA transfection efficiency by real-time RT-PCR.

Silencer® Select Negative Control siRNAs

Negative control siRNAs—siRNAs with sequences that do not target any gene product—are essential for determining the effects of siRNA delivery on the cell and for providing a baseline to compare siRNA-treated samples. The extensively tested *Silencer*® Select Negative Control siRNAs include the same modifications for reducing off-target effects found in other *Silencer*® Select siRNAs and have no significant sequence similarity to mouse, rat, or human gene sequences. They have been shown to have minimal effects on gene expression, and no significant effects on cell proliferation, viability, or morphology in the cell lines tested.

Silencer® Select Predesigned and Validated siRNAs

These products are ordered using our online system; go to www.invitrogen.com/rnai. Ready-to-ship *Silencer*® Select Libraries include:

Silencer® Select siRNA Libraries

- *Silencer*® Select Human Genome siRNA Library
- *Silencer*® Select Human Kinase siRNA Library
- *Silencer*® Select Human Phosphatase siRNA Library
- *Silencer*® Select Human GPCR siRNA Library
- *Silencer*® Select Human Protease siRNA Library
- *Silencer*® Select Human Ion Channel siRNA Library
- *Silencer*® Select Human Extended Druggable Genome siRNA Library

In addition to human siRNA libraries, *Silencer*® Select siRNA Libraries are also available for mouse and rat. For detailed information and availability, go to www.invitrogen.com/RNA/libraries. For information on custom libraries, contact custom.services@invitrogen.com.

Silencer® Select Positive and Negative Controls

Product	Quantity	Cat. No.
Silencer® Select Negative Control #1 siRNA	5 nmol	4390843
Silencer® Select Negative Control #1 siRNA	40 nmol	4390844
Silencer® Select Negative Control #2 siRNA	5 nmol	4390846
Silencer® Select Negative Control #2 siRNA	40 nmol	4390847
Silencer® Select GAPDH Positive Control siRNA	5 nmol	4390849
Silencer® Select GAPDH Positive Control siRNA	40 nmol	4390850

Ambion® *In Vivo* siRNAs

Chemically modified RNAi duplexes for specificity, stability, and effective knockdown

Ambion® *In Vivo* siRNA

Ambion® *In Vivo* siRNAs, the new standard for *in vivo** RNAi applications, offer:

- High stability against nucleases
- No induction of the interferon response
- Easy tracking of administered siRNAs
- Combined knockdown effectiveness and specificity

Ambion® *In Vivo* siRNA molecules are chemically modified, 21-mer, double-stranded siRNAs that are recognized by the RNA-induced silencing complex (RISC) to mediate inhibition of a target gene. Proprietary chemical modifications allow Ambion® *In Vivo* siRNAs to overcome many *in vivo*–specific obstacles, ensuring their effectiveness and stability *in vivo*. Ambion® *In Vivo* siRNAs are at least 100x more stable in 90% mouse serum than unmodified siRNAs.

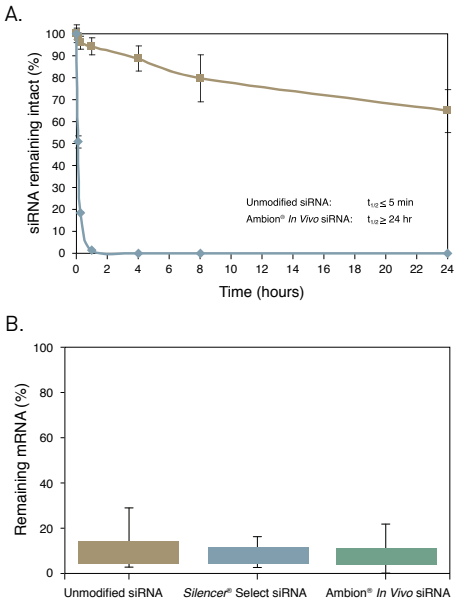
Go to www.invitrogen.com/rnai to order Ambion® *In Vivo* siRNAs using our online system.

Ambion® *In Vivo* siRNA Controls

The features of these controls include:

- Positive controls functionally tested siRNA controls for human, mouse, and rat cell lines
- Negative Controls functionally proven to have minimal effects on cell proliferation and viability
- *Silencer*® Select and *in vivo* modifications for enhanced specificity of positive controls and increased stability

Ambion® *In Vivo* Positive Control siRNAs are available targeting GAPDH or Factor VII (FVII/F7), also known as proconvertin, which is a Vitamin K–dependent serine protease that functions as a central protein in the coagulation cascade. Ambion® *In Vivo* Negative Control #1 siRNA is also available.



Ambion® *In Vivo* siRNAs are >100x more serum-stable and are just as effective at knockdown as unmodified siRNAs. (A) Eight different Ambion® *In Vivo* siRNA sequences, and the same eight sequences in an unmodified form, were incubated in 90% serum for the time indicated. The fraction of remaining full-length siRNA remaining was quantified by HPLC. **(B)** 40 different siRNA sequences with no modifications, *Silencer*® Select siRNA modifications, or Ambion® *In Vivo* siRNA modifications were transfected into HeLa cells, and target gene knockdown was measured 48 hr later using TaqMan® Gene Expression Assays. The boxes represent the middle quartile of data, with the line indicating the median percentage of mRNA remaining.

Ambion® *In Vivo* siRNAs

Product	Quantity	Cat. No.
Ambion® *In Vivo* Negative Control #1 siRNA	5 nmol	4390843
Ambion® *In Vivo* Negative Control #1 siRNA	50 nmol	4459405
Ambion® *In Vivo* Negative Control #1 siRNA	250 nmol	4457289
Ambion® *In Vivo* GAPDH Positive Control siRNA	5 nmol	4457288
Ambion® *In Vivo* GAPDH Positive Control siRNA	50 nmol	4459407
Ambion® *In Vivo* GAPDH Positive Control siRNA	250 nmol	4457291
Ambion® *In Vivo* Factor VII Positive Control siRNA	50 nmol	4459408
Ambion® *In Vivo* Factor VII Positive Control siRNA	250 nmol	4457292

**in vivo* refers to research use in small animals. This product is not intended for use in humans.

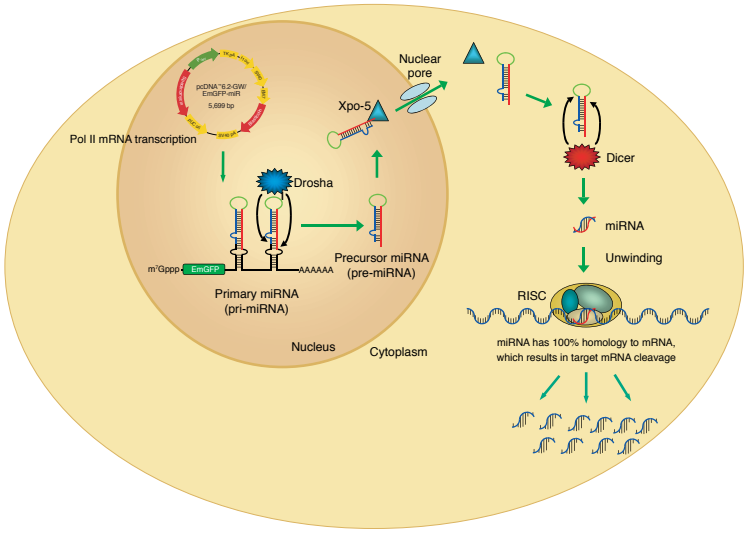
Overview of vector-mediated RNAi

Both miR RNAi and shRNA vector systems take advantage of the endogenous RNAi pathway found in all animal cells. Compared to shRNA vectors, miR RNAi vector systems make better use of the cell's machinery, resulting in more efficient processing of expressed RNA hairpins (see figure).

BLOCK-iT™ Pol II miR RNAi expression vector kits use the pcDNA™6.2GW/EmGFP-miR Expression Vector to transcribe artificial miRNA via RNA Polymerase II. The result is the cocistronic expression of Emerald Green Fluorescent Protein (EmGFP) and multiple miRNA hairpins on the same transcript. The primary miRNA (pri-miRNA) transcript contains the EmGFP sequence on the 5´ end, followed by one or more precursor miRNAs (pre-miRNAs). The enzyme Drosha recognizes pre-miRNA sequences and excises them from the pri-miRNA transcript. Each pre-miRNA is then actively transported out of the nucleus by Xpo-5.

In the cytoplasm, Dicer processes the pre-miRNA hairpins into miRNAs. Finally, the miRNAs unwind, load into the RNA-induced silencing complex (RISC), and hybridize with their mRNA target and result in target cleavage.

In contrast, short hairpin RNA (shRNA) vectors contain an RNA Polymerase III (Pol III) promoter (H1 or U6) for nuclear expression of shRNAs. Exportin-5 actively exports shRNA to the cytoplasm where it is recognized and cleaved by the RNase III enzyme Dicer to produce short interfering RNA (siRNA).



Expression of miR RNAi sequences using the BLOCK-iT™ Pol II miR RNAi expression vectors.

Choosing a lentiviral or adenoviral RNAi system.

Viral system	When to use	Products
Lentiviral RNAi delivery systems (page 197)	• Stable RNAi in any cell line, even nondividing cells • Inducible or constitutive shRNA or miR RNAi expression • Studies in animal models	• **BLOCK-iT™ Lentiviral Pol II miR RNAi Expression System**—a complete lentiviral system with all of the advantages of miR RNAi: multiple-target knockdown and a higher design success rate than conventional shRNA (contains pLenti6/V5-DEST™ vector) • **BLOCK-iT™ Lentiviral Pol II miR RNAi Expression System with EmGFP**—a system with all of the benefits listed above, plus easy expression tracking with cocistronic EmGFP (contains pLenti6/V5-DEST™ vector) • **BLOCK-iT™ Inducible H1 Lentiviral RNAi System**—complete lentiviral system for inducible or constitutive shRNA expression in any cell type (contains pLenti4/BLOCK-iT™-DEST vector) • **BLOCK-iT™ Lentiviral RNAi Expression System**—complete lentiviral system for constitutive shRNA expression in any cell type (contains pLenti6/BLOCK-iT™-DEST vector)
Adenoviral RNAI delivery System (page 198)	• High-level transient shRNA expression • Effective delivery to a wide range of human cell types • Studies in animal models	• **BLOCK-iT™ Adenoviral RNAi Expression System**—complete system for high-level transient expression of shRNA

BLOCK-iT™ Lentiviral RNAi Expression Systems

Stable and inducible expression systems for miRNA and shRNA

- Efficient and effective lentiviral delivery of miRNA and shRNA
- Greater than 75% knockdown success with simple online hairpin insert designer
- Easily track miRNA expression with co-expression of GFP
- Inducible system available

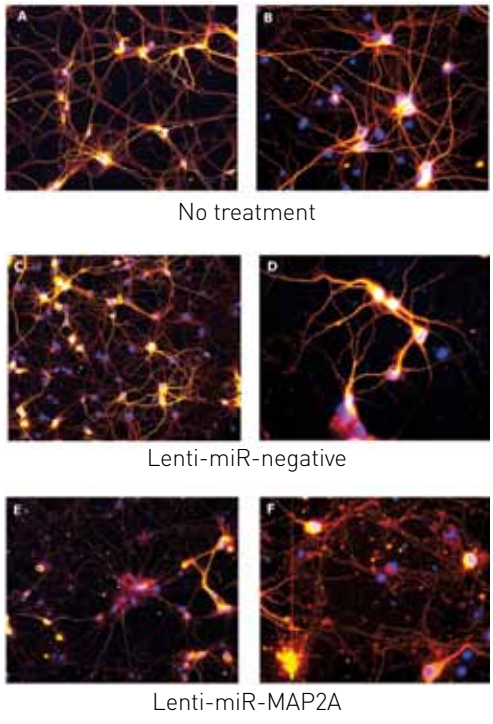
BLOCK-iT™ RNAi Expression Systems can be used to efficiently introduce and stable express short hairpin RNA (shRNA) or microRNA (miRNA) *in vivo* from a lentiviral vector. These systems provide options for long-term, as well as inducible expression, for gene knockdown studies in a wide variety of cell types.

BLOCK-iT™ Lentiviral Pol II miR RNAi Expression System

The BLOCK-iT™ Lentiviral Pol II miR RNAi Expression System combines BLOCK-iT™ Pol II miR RNAi and ViraPower™ Lentiviral technologies to facilitate creation of a replication-incompetent lentivirus that delivers a microRNA sequence of interest to dividing or nondividing mammalian cells for RNA interference (RNAi) analysis, thus broadening the potential RNAi applications beyond those of other traditional retroviral systems [1].

BLOCK-iT™ Lentiviral Pol II miR RNAi Expression System with EmGFP

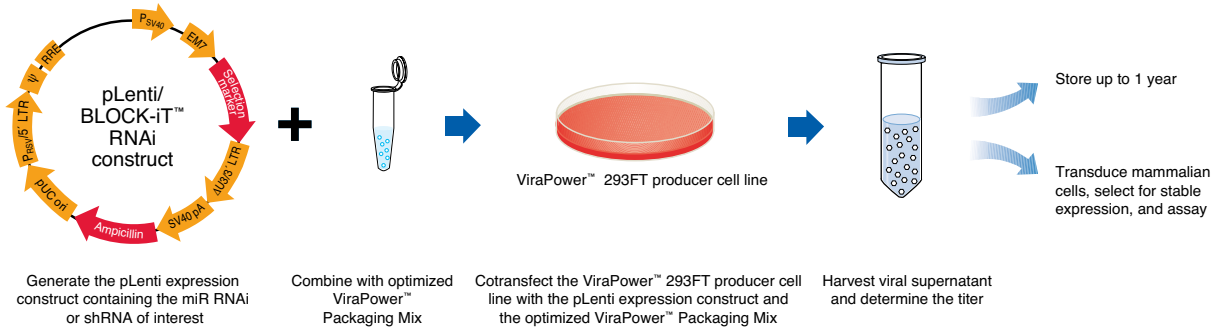
This system combines the same components as the BLOCK-iT™ Lentiviral Pol II miR RNAi Expression System (described above), plus the pLenti6/V5-DEST™ vector for easy tracking with cocistronic expression of the Emerald Green Fluorescent Protein (EmGFP).



Lentiviral transduction of miR RNAi. Untreated (**A, B**) and transduced (**C–F**) samples were stained with MAP2 antibodies (orange) and DAPI (blue). **E** and **F** clearly show less target protein expression compared to the untreated neurons and those transduced with the Lenti-miR-negative control.

Reference

1. Current Opinion Biotech 9:5 (1998).



How the BLOCK-iT™ lentiviral RNAi systems work.

BLOCK-iT™ Lentiviral RNAi Expression System

Stable expression of shRNA *in vivo* for RNAi studies

The pLenti6/BLOCK-iT™-DEST expression vector provided in the BLOCK-iT™ Lentiviral RNAi Expression System can be used to efficiently introduce and stably express short hairpin RNA (shRNA) *in vivo* from a lentiviral vector. This shRNA is expressed and then recombined into the pLenti6/BLOCK-iT™-DEST vector. After viral production and transduction, the shRNA driven by a U6 promoter becomes stably integrated as an RNAi cassette. The shRNA generated avoids the host's defense mechanism and will be effective at producing the RNAi gene knockdown response.

BLOCK-iT™ Inducible H1 Lentiviral RNAi System

Inducible RNAi—in any cell type

The pLenti4/BLOCK-iT™-DEST expression vector provided in the BLOCK-iT™ Inducible H1 Lentiviral RNAi System can be used for long-term inducible shRNA expression in virtually any mammalian cell. A novel cloning process places a a double-stranded oligonucleotide immediately following an H1/TO pol III promoter to generate an H1/TO RNAi cassette in an inducible BLOCK-iT™ H1/TO entry construct. This can be quickly and efficiently recombined from the pENTR™/H1/TO entry vector into the pLenti4/BLOCK-iT™-DEST vector via a standard Gateway® LR recombination reaction. The construct is transfected with the ViraPower™ Lentiviral packaging mix into 293FT cells with Lipofectamine® 2000 Reagent producing viral particles capable of transducing any mammalian cell. If the cell expresses the tetracycline repressor (TR) protein, inducible RNAi is possible. The pLenti4/BLOCK-iT™-DEST expression vector enables lentiviral delivery and genomic integration of DNA coding for shRNA. Once expressed, the shRNA is processed by cellular machinery and initiates target-specific RNAi.

Product	Quantity	Cat. No.
BLOCK-iT™ Lentiviral Pol II miR RNAi Expression System	20 reactions	K493700
BLOCK-iT™ Lentiviral Pol II miR RNAi Expression System With EmGFP	20 reactions	K493800
BLOCK-iT™ Inducible H1 Lentiviral RNAi System	20 reactions	K492500
BLOCK-iT™ Lentiviral RNAi Expression System	20 reactions	K494400

BLOCK-iT™ Adenoviral RNAi Expression System

Efficient delivery and transient expression of a short hairpin RNA (shRNA) *in vivo*

After efficient recombination of the entry vector into the pAd/BLOCK-iT™-DEST vector, followed by viral production and transduction, the shRNA driven by the U6 promoter can be transiently expressed in most dividing or nondividing mammalian cell types and animal models. The shRNA generated avoids the host's defense mechanism and will be effective at producing the RNAi gene knockdown response. The system contains all of the required components for efficient adenoviral packaging and delivery of the shRNA of interest.

Product	Quantity	Cat. No.
BLOCK-iT™ Adenoviral RNAi Expression System	20 reactions	K4924100

Overview of RNAi transfection

RNAi delivery is accomplished by physical or chemical means, and includes lipid-mediated transfection, virus-mediated transduction, and electroporation. Determining which of these approaches to use depends on the cell type being studied and whether transient or stable knockdown is desired. The most popular chemical transfection method, transient transfection of unmodified siRNAs, modified siRNA duplexes, or RNAi vectors, uses charged molecules to deliver siRNA across the cell membrane. Cationic lipid–based reagents are suitable for delivering these molecules across a diverse range of commonly used cell lines. For cell types not amenable to lipid-mediated transfection, viral vectors or electroporation are often employed. Adenoviral vectors work well for transient delivery in many cell types; however, for stable delivery in dividing and nondividing cells, lentiviral vectors are best. Electroporation, allows direct transfect through the membrane of the cell and introduce the siRNA directly into the cytoplasm. The table below descirbes the products available for RNAi transfection.

Choosing RNAi transfection products.

	Lipofectamine® RNAiMAX Transfection Reagent (page 200)	Lipofectamine® LTX Transfection Reagent (page 201)	Invivofectamine® 2.0 Reagent (page 202)	The Neon® Transfection System (page 203)
Optimized for transfection	siRNA, miRNA, snRNA *in vitro*	shRNA and other vectors and plasmids *in vitro*	siRNA *in vivo*	DNA and siRNA *in vitro*
Features	• Superior transfection efficiency enables use of lower siRNA with minimal nonspecific effects • Easy optimization and low cytotoxicity • Compatible with a wide range of cell types • Simple and rapid protocol for consistent results	• Easy-to-follow protocol • Convenient optimization with the included BLOCK-iT™ Fluorescent Oligo • Excellent performance in a wide variety of cell types	• Easily deliver siRNA into transgenic mice and rats • Highly effective and specific • Stable complex formation • Nontoxic and non-immunostimulatory	• Efficient for many cell types; difficult-to-transfect, primary, and stem cells • Flexible transfection into a range of cell numbers • Single reagent kit for all cell types and simple procedure • Versatile system allows easy optimization of electroporation parameters
Technology	Chemical	Chemical	Chemical	Mechanical (electroporation)

Lipofectamine® RNAiMAX Transfection Reagent

Unmatched gene silencing with reduced cytotoxicity for siRNA experiments

The Lipofectamine® RNAiMAX Transfection Reagent provides:

- Superior transfection efficiency enables use of lower siRNA concentrations and leads to more successful gene knockdown with a minimum of nonspecific effects
- Easy optimization due to low cytotoxicity across a 10-fold range of transfection reagent concentrations
- Compatible with a wide range of cell types
- Simple and rapid protocol for consistent results
- Convenient optimization of transfection conditions and efficiency with the BLOCK-iT™ Fluorescent Oligo

The procedure for using Lipofectamine® RNAiMAX Transfection Reagent consists of mixing it with siRNA, adding cells, incubating, and measuring gene knockdown (Figure 1). The simplicity and speed, combined with superior transfection efficiency (Figure 2), make Lipofectamine® RNAiMAX Transfection Reagent the best choice for high-throughput siRNA transfections. Transfection conditions can be readily established for automated or robotic systems used in such applications.

Product	Quantity	Cat. No.
Lipofectamine® RNAiMAX Transfection Reagent	0.75 mL	13778075
	0.1 mL	13778100
	1.5 mL	13778150
BLOCK-iT™ Alexa Fluor® Red Fluorescent Control	2 x 125 µL	14750100

NOTE: Lipofectamine® RNAiMAX Transfection Reagent is for use with siRNA duplexes, and not for transfecting vectors expressing RNAi sequences.

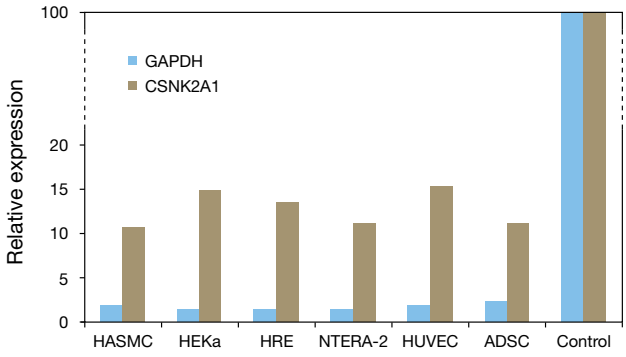


Figure 1. Superior knockdown of *Silencer*® Select siRNA with Lipofectamine® RNAiMAX Transfection Reagent. Transfection efficiency is expressed as the percentage of cells that have received the RNAi duplex or expression plasmid. With the cationic Lipofectamine® RNAiMAX Transfection Reagent, transfection efficiency can be optimized and verified easily using the BLOCK-iT™ Alexa Fluor® Red Fluorescent Control. (Email rnairesearcher@invitrogen.com for protocol details).

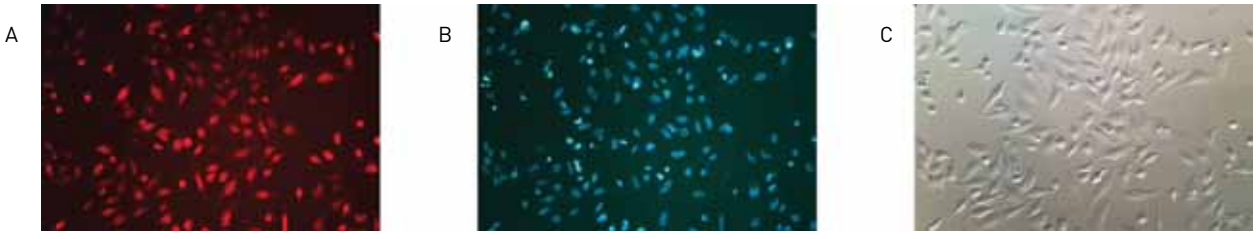


Figure 2. Assessing transfection efficiency of the Lipofectamine® RNAiMAX Transfection Reagent with the BLOCK-iT™ Alexa Fluor® Red Fluorescent Control. Lipofectamine® RNAiMAX Transfection Reagent was used to transfect HeLa cells with the BLOCK-iT™ Alexa Fluor® Red Fluorescent Control (50 nM) (**A**). Twenty-four hours after transfection, growth medium was removed and replaced with PBS containing 10 mg/mL Hoechst 33342 for visualization of cell nuclei (**B**). Nuclear localization of the red-fluorescent control oligo is seen (**A**). The bright-field image (**C**) shows that the cells retain a normal morphology after transfection.

Lipofectamine® LTX Transfection Reagent

Ideal for delivery of shRNA and miR RNAi vectors

The Lipofectamine® LTX Transfection Reagent provides:

- Effective transfection or cotransfections of shRNA and miR RNAi vectors and synthetic siRNAs
- Easy-to-follow protocols; media changes not required
- Excellent performance in a wide variety of cell types

Lipofectamine® LTX Reagent offers a new, advanced solution for gene expression studies in hard-to-transfect cells. No other plasmid DNA-specific transfection reagent can match the efficiency, convenience, and gentleness of Lipofectamine® LTX Reagent for transfection of primary, difficult-to-transfect, and sensitive cell lines.

Lipofectamine® LTX Reagent offers the highest level of reproducible, efficient protein expression in all cell types, including primary, hard-to-transfect, and disease-related cells. This enables cells to respond to your assay conditions, which is critical for relevant data from functional genomics expression studies. Lipofectamine™ LTX Reagent is synthesized from 100% animal origin–free components, making it easy to validate the absence of zoonotic diseases, such as BSE or viruses, in experiments or cell lines. Use with PLUS™ Reagent to obtain both high levels of protein expression and maximum viability in primary cells. This reagent was designed with the perfect balance of potency while still being gentle to your cells.

Product	Quantity	Cat. No.
Lipofectamine® LTX and PLUS™ Reagent	1 mL	15338100
Lipofectamine® LTX and PLUS™ Reagent	0.1 mL	A12621

For recommended transfection conditions for various cell types, go to www.invitrogen.com/transfection.

Invivofectamine® 2.0 Reagent

A breakthrough for *in vivo* RNAi delivery

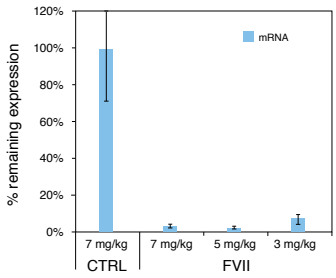
In vivo RNAi is used to achieve phenotypic alternations in animals. Historically, *in vivo* RNAi delivery reagents have been ineffective and difficult to use. Using Invivofectamine® 2.0 Reagent to deliver siRNA *in vivo*, you can transfect cells in the liver with confidence that the siRNA arrives intact and ready to perform knockdown. The features of Invivofectamine® 2.0 Reagent include:

- Easy to use—mix, equilibrate, and inject
- Effective—80% or greater knockdown in liver
- Specific—direct correlation of phenotypic response to target knockdown
- Stable complex formation
- Nontoxic and non-immunostimulatory

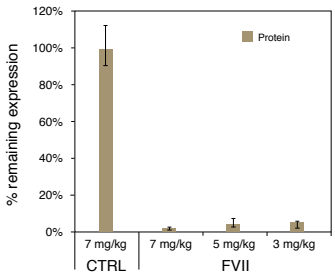
Complexes comprising Invivofectamine® 2.0 Reagent and Ambion® *In Vivo* siRNA targeting Factor VII have been successfully delivered by tail vein injection to liver tissue. We have demonstrated effective knockdown of Factor VII at the mRNA level (figure, panel A) and by loss of protein activity in serum 2 days post injection (figure, panel B). RNAi-mediated knockdown occurs through siRNA interaction with the RISC complex resulting in a site-specific cleavage of the target mRNA. The cleavage site can be detected by using a method known as RACE (Rapid Amplification of cDNA Ends). Data shown in the figure (panel C) verify that the knockdown observed was mediated through the RISC complex and is specific to the siRNA target sequence.

Product	Quantity	Cat. No.
Invivofectamine® 2.0, 1 mL Starter Kit	1 kit	1377-501
Invivofectamine® 2.0, 5 mL Kit	1 kit	1377-505

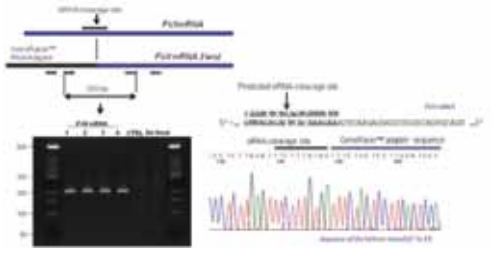
A. mRNA knockdown



B. Protein expression level



C. siRNA-mediated cleavage



Use of Invivofectamine® 2.0 Reagent results in targeted knockdown in liver after a single intravenous Injection. Invivofectamine® 2.0 Reagent complexed with siRNA targeting Factor VII mRNA (injected at doses of 1, 3, 5, and 7 mg/kg) achieved >95% knockdown of target mRNA levels (**A**) (knockdown was assessed by TaqMan® assays), and >95% reduction in protein activity (**B**) (blood serum was isolated and assayed for Factor VII activity 48 hours post-injection via a chromogenic substrate).The knockdown observed was RNAi-mediated (**C**) as shown by detection of the siRNA-induced mRNA cleavage fragment by RNA ligase-mediated rapid amplification of 5′ cDNA ends (5′ RLM-RACE). Schematic depicting the location of the predicted Factor VII-specific siRNA cleavage site and the primers used for PCR amplification of the cleavage fragment are shown above. 5′ RLM-RACE PCR amplification products from treated mice were of the expected size (208 bp band) on agarose gels.

Neon® Transfection System

High transfection efficiency and high cell viability in a broad range of cell lines

Features of the Neon® system include:

- Efficiency—up to 90% in many cell types, including difficult-to-transfect, primary, and stem cells
- Flexibility—easily transfect from 1 x 10^4 cells to 5 x 10^6 cells per reaction
- Simplicity—single reagent kit for all cell types and simple three-step procedure
- Versatility—open system allows electroporation parameters to be optimized freely

The Neon® Transfection System (Figure 1) is a novel benchtop electroporation device that uses a pipette tip as an electroporation chamber to efficiently transfect mammalian cells, including primary and immortalized hematopoietic cells, stem cells, and primary cells. The system efficiently delivers nucleic acids, proteins, and siRNA into all mammalian cell types with a high cell survival rate. The transfection is performed using as few as 2 x 10^4 or as many as 1 x 10^7 cells per reaction, in a sample volume of 10 µL or 100 µL, in a variety of cell culture formats (from 96-well to 100mm).

The Neon® Transfection System uses a single transfection kit that is compatible with various mammalian cell types, including primary and stem cells, thereby avoiding the need to determine an optimal buffer for each cell type. The system offers open and transparent protocols that are optimized for ease of use and simplicity (Figure 2).

Figure 1. The Neon® Transfection System.

Product	Quantity	Cat. No.
Neon® Transfection System 100 µL Kit	50 reactions	MPK10025
Neon® Transfection System 100 µL Kit	192 reactions	MPK10096
Neon® Transfection System 10 µL Kit	50 reactions	MPK1025
Neon® Transfection System 10 µL Kit	192 reactions	MPK1096
Neon® Transfection System	1 each	MPK5000
Neon® Transfection System Starter Pack	1 pack	MPK5000S
Neon® Transfection System Pipette	1 each	MPP100
Neon® Transfection System Pipette Station	1 each	MPS100
Neon® Transfection Tubes	1 pack	MPT100
One-Year Extended Warranty, Rapid Exchange Service	1 each	4457724
Three-Year Extended Warranty, Rapid Exchange Service	1 each	4457725

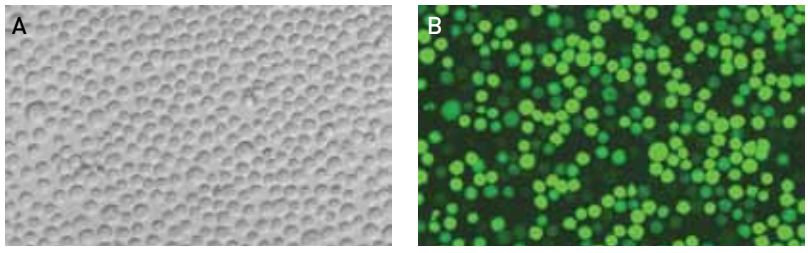


Figure 2. High transfection efficiency of Jurkat cells with the Neon® Transfection System. Intracelluar uptake of reporter vector encoded with EGFP at 24 hr following transfection of Jurkat cells using the Neon® Transfection System. B is the corresponding fluorescence image of A.

Overview of non-coding RNA analysis

It has recently been demonstrated that miRNA and non-coding RNA (ncRNA) are key to gene regulation, and their expression can be influenced by environmental and genetic factors. Long ncRNAs are transcripts that are longer than 200 nucleotides, distinguishing them from miRNAs that are typically 19 to 22 nucleotides long. These genetic elements have been shown to play a critical role in many areas, including developmental timing, cell fate, tumor progression, and neurogenesis. Scientists are just begining to understand these natural "gene-silencing" mechanism, which are helping them map genomic pathways and, in biomedicine, uncover connections to diseases and new classes of therapies.

Life Technologies provides a full suite of solutions to study the role of ncRNAs, from hypothesis-neutral research studies that utilize next-generation sequencing enabling you to discover novel ncRNA, to quantifying, profiling, and validating the levels of known ncRNA with real-time PCR or microarray platforms.

Products for non-coding RNA and miRNA analysis include:

Ambion® sample preparation for ncRNA and miRNA

- A selection of sample preparation products optimized for the isolation of small RNAs from a variety of sample types (see below)
- TaqMan® Assays for small RNA, ncRNA, pri-micro and miRNA analysis

SOLiD® Total RNA-Seq Kit

- A complete solution for the discovery of small RNAs using the next-generation SOLiD® sequencing system (page 207)

TaqMan® Non-coding RNA Assays

- Specific and reproducible quantification of short and long ncRNA expression levels, including miRNA (page 213)

miRNA analysis

- TaqMan® MicroRNA and pri-MicroRNA Assays and Arrays for specific miRNA quantitation (page 209)
- *mir*Vana™ miRNA Mimics and Inhibitors for artificial regulation of target mRNA (page 215)

Sample preparation tailored for microRNA and ncRNA experiments

Most RNA isolation kits were developed to recover mRNA and ignore smaller molecules such as miRNA. Life Technologies offers a range of produts specifically designed for optimal recovery of miRNA and other small RNAs from a wide variety of samples types. See the selection guide below for help choosing which product best fits your needs.

For more detailed information on sample preparation kits for all applications, see Chapter 2—Nucleic Acid Purification and Analysis, starting on page 71.

Choosing a miRNA or non-coding RNA sample preparation kit.

Product	Description	Sample input amounts
Ambion® *mir*Vana™ miRNA Isolation Kit (page 205)	Isolates miRNA, siRNA, snRNA, and other small RNAs from tissues and cells	10^3–10^7 cultured cells or 0.5–250 mg tissue
Ambion® *mir*Vana™ PARIS™ Kit (page 205)	Isolates mRNA, miRNA, siRNA, and protein from tissues and cells	10^2–10^7 cultured cells or up to 100 mg tissue
Ambion® RecoverAll™ Total Nucleic Acid Isolation Kit for FFPE Tissues (page 206)	Isolates total nucleic acids from FFPE samples	Up to four 20 µm FFPE sections
Ambion® TaqMan® MicroRNA Cells-to-C$_T$™ Kit (page 206)	For miRNA profiling directly from cells without RNA purification	10–10^4 cells

Ambion® *mir*Vana™ miRNA Isolation Kit

Efficient isolation of small RNA-containing total RNA

The kit offers the following features:

- Enrich for small RNA <200 nt to increase sensitivity in downstream analyses
- Simple 30 minute procedure
- Ideal for miRNA, siRNA, shRNA, and snRNA analysis
- Compatible with virtually all cell and tissue types

The Ambion® *mir*Vana™ miRNA Isolation Kit uses a rapid procedure to isolate small RNAs from tissue and cells using an efficient glass fiber filter (GFF)-based method. The method isolates total RNA ranging in size from kilobases down to 10-mers. Each kit contains sufficient reagents for up to 40 purifications.

Ambion® *mir*Vana™ PARIS™ miRNA Isolation Kit

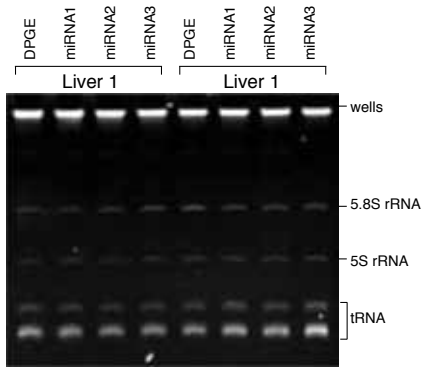
For the purification of both native protein and RNA from the same sample

The kit includes sufficient reagents for processing up to 40 samples, and offers the following features:

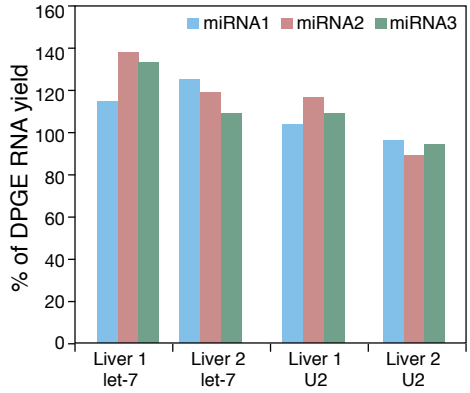
- Isolate small-RNA–containing total RNA and native protein from the same sample
- Start with 100–10^7 cultured cells or up to 100 mg of many mammalian tissues
- Enrich for small RNA <200 nt to increase sensitivity in downstream analyses
- Simple 30 minute procedure
- Ideal for correlating mRNA, miRNA or siRNA, and protein levels

The *mir*Vana™ PARIS™ Kit procedure begins with homogenization of samples with a special Cell Disruption Buffer that includes nonionic detergent. Protein remains intact, so a portion of the lysate can be used directly for common protein analysis applications. The remainder of the lysate is used for RNA isolation, using a procedure that combines the advantages of organic and solid-phase extraction, while avoiding the disadvantages of both.

A.



B.



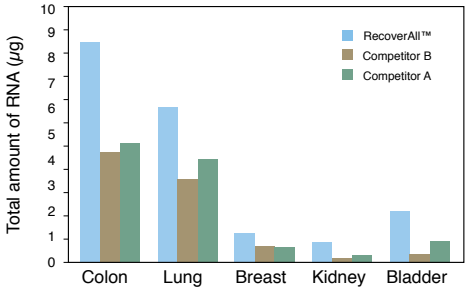
***mir*Vana™ miRNA Isolation Kit: improved RNA isolation procedure for efficient recovery of miRNA. (A)** Total RNA was isolated from the same mouse liver lysate using a double phenol/guanidinium extraction (DPGE) or the *mir*Vana™ miRNA Isolation Kit procedure in triplicate (miRNA 1 to 3). The experiment was performed with two different mouse liver lysates. 1 µg of each sample RNA was analyzed on a denaturing 15% polyacrylamide gel stained with ethidium bromide. **(B)** RNAs from the same gel were transferred to a membrane and probed for U2 snRNA and let-7 miRNA. The relative amount of small RNA in each lane was quantified with a phosphorimager. The graph shows the percentage of recovery relative to the DPGE prep.

Ambion® RecoverAll™ Total Nucleic Acid Isolation Kit for FFPE Tissues

For the extraction of total nucleic acid from formalin or paraformalin-fixed, paraffin-embedded (FFPE) tissues

Sufficient reagents are included for the 40 purifications from up to four 20 µm sections, or up to 35 mg of unsectioned core samples each. The kit features include:

- Optimized for isolation of total nucleic acids, including microRNAs, from FFPE tissue
- No overnight Proteinase K digestion required—deparaffinize in the morning and perform qRT-PCR in the afternoon
- Obtain typical yields of >50% that of unfixed tissue from the same sample source (see figure)
- Recovered nucleic acids are suitable for real-time RT-PCR, PCR, mutation screening, and microarray analyses



Yield of RNA from archived human FFPE tissue samples: RecoverAll™ kit vs. two competitor systems. A 10–20 µm section from each of the above archived human tissue blocks was isolated using each of three kits: Competitor A kit, Competitor B kit, and the Ambion® RecoverAll™ kit. Colon, lung, and breast samples were 1–2 years old; kidney was 3–5 years old; and bladder was 10–15 years old. Once the RNA was isolated, the concentration was determined via OD_{260}, and the amount of RNA recovered in micrograms was calculated. The RecoverAll™ Kit yielded the highest recovery of the three systems for all tissue types.

TaqMan® MicroRNA and Gene Expression Cells-to-CT™ Kits

miRNA and ncRNA expression profiling directly from cultured cells without RNA purification

The kit is the first to offer a simple, complete workflow for miRNA expression analysis from a few samples, or it can easily be incorporated into automated high-throughput applications. The kit features:

- Complete, validated solution—optimized workflow includes cell lysis reagents, DNase, TaqMan® RT reagents, and TaqMan® Universal PCR Master Mix
- Fast, simple, and convenient—prepare samples in 10 minutes; eliminate tedious RNA isolation
- Gold standard specificity—achieve superior results with TaqMan® reagents
- Robust performance—results equivalent to purified RNA
- Broad input dynamic range—linear detection from 10 to 100,000 cells
- TaqMan® Gene Expression Cells-to-CT™ Kit for ncRNA and TaqMan® MicroRNA Cells-to-CT™ Kit for miRNA analysis

Product	Quantity	Cat. No.
*mir*Vana™ miRNA Isolation Kit	40 purifications	AM1560
*mir*Vana™ PARIS™ Kit	40 purifications	AM1556
RecoverAll™ Total Nucleic Acid Isolation Kit for FFPE	40 purifications	AM1975
TaqMan® MicroRNA Cells-to-CT™ Kit	100 reactions	4391848
TaqMan® Gene Expression Cells-to-CT™ Kit	40 reactions	4399002

SOLiD® Total RNA-Seq Kit

Interrogate the whole transcriptome and small RNA on the SOLiD® Sequencing System

The SOLiD® Total RNA-Seq Kit is designed to be a complete solution with streamlined protocols for whole genome discovery or small non-coding RNA analysis (see figure). The kit includes all necessary reagents to enable hypothesis-neutral discovery of coding RNA, non-coding RNA, novel transcripts, alternate splicing, small RNAs, and isoforms, all while maintaining accurate sequence representation and preserving strand direction.

Small RNA analysis

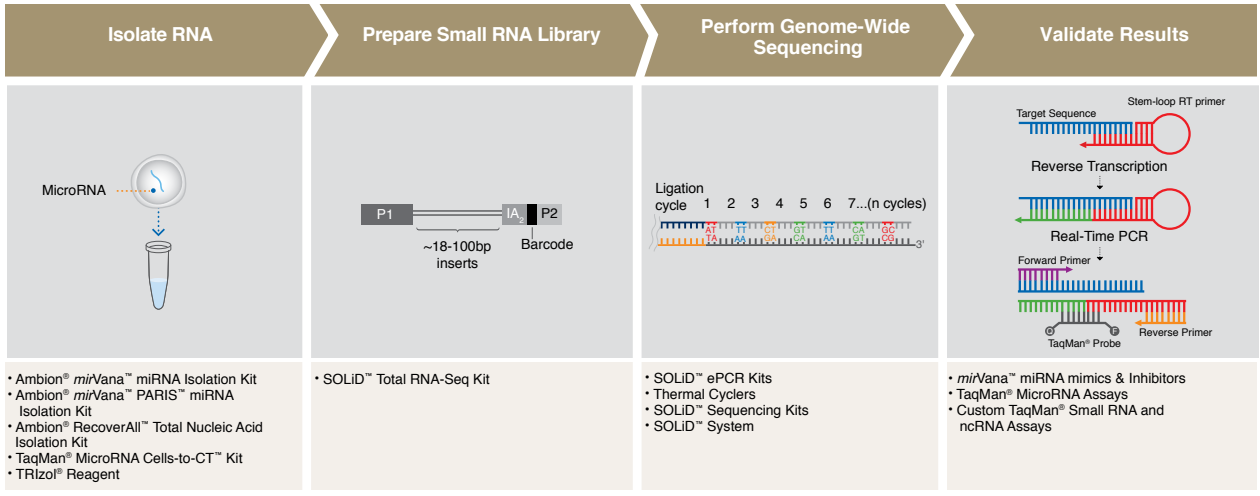
Small non-coding RNAs play a key role in regulating a variety of biological processes, including developmental timing, cellular differentiation, and tumor progression. Small RNAs are typically 18–40 nucleotides in length and belong to one of a variety of small RNA classes such as microRNA (miRNA), short interfering RNA (siRNA), and piwi-interacting RNA (piRNA).

With the SOLiD® Total RNA-Seq Kit you get:

- Hypothesis-free global expression analysis—capable of detecting all small RNA in a single assay
- Maintenance of genomic DNA strand specificity
- Highly sensitivity—interrogate low expression levels from a limited quantity of total RNA (ten-fold less than other commercially available kits)
- Simple, single-day protocol for library construction

One complete solution for RNA sequencing on the SOLiD® System

Specifically designed for use with the SOLiD® System, the SOLiD® Total RNA-Seq Kit contains enough reagents to process up to 12 samples. Either whole transcriptome libraries or small RNA libraries can be barcoded using SOLiD® RNA Barcoding Kits, allowing the respective pooling of size-selected libraries for more efficient and cost-effective analysis using a single slide.



SOLiD® RNA Barcoding Kits

Using unique barcode sequences for optimal multiplexing of up to 48 libraries, SOLiD® RNA Barcoding Kits enable the assignment of a unique identifier to templated beads made from a single library. Once the identifiers are assigned, multiple batches of templated beads may be pooled together for emulsion PCR and then sequenced. Three kits are available, each containing 16 different barcodes.

Product	Quantity	Cat. No.
SOLiD® Total RNA-Seq Kit	12 preps	4445374
SOLiD® RNA Barcoding Kit, Module 1–16	16 libraries	4427046
SOLiD® RNA Barcoding Kit, Module 17–32	16 libraries	4453189
SOLiD® RNA Barcoding Kit, Module 33–48	16 libraries	4453191

TaqMan® Non-coding RNA, MicroRNA and Custom TaqMan® Assays

Comprehensive collection of real-time PCR assays for profiling and quantitation of ncRNAs

The Life Technologies TaqMan® Assay portfolio includes a broad selection of non-coding RNA (ncRNA) assays. These ncRNA Assays are an essential tool to better understand transcriptional regulation, messenger RNA stability and translation, epigenetic regulation, and assembly of macromolecular complexes. By making novel adaptations in assay design specifically for the detection of ncRNAs (Figure 1), TaqMan® Assays are the gold standard for specificity, sensitivity, and reproducibility in real-time PCR.

TaqMan® Non-coding RNA and MicroRNA Assays feature:

- Reproducible quantification of small and long non-coding RNA, including mature and precursor miRNA
- Sensitive detection of high- and low-expressing RNAs in a single experiment
- Simultaneously detect multiple targets under universal conditions

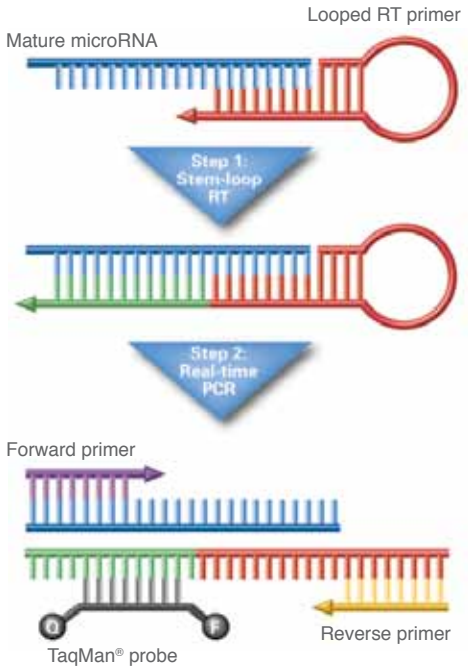


Figure 1. TaqMan® MicroRNA and Custom TaqMan® Small RNA Assay Design. TaqMan® MicroRNA and Custom TaqMan® Small RNA Assays incorporate a target-specific stem–loop reverse transcription primer to address a fundamental problem in small RNA quantitation: the short length prohibits conventional design of a random-primed RT step followed by a specific real-time assay.

TaqMan® ncRNA and miRNA Assays	Description
TaqMan® MicroRNA Assays and TaqMan® pri-MIcroRNA Assays (page 209)	•Real-time PCR assays for mature and precursor miRNAS •Available for all human and rodent miRNAs •>10,000 made-to-order assays for all other miRNAs in the Sanger registry •Available in single-tube assays, 384-well microfluidic cards, and OpenArray® plates
Custom TaqMan® Small RNA Assays (page 213)	•Custom designed real-time PCR assays for any small RNA (17–200 nucleotides) •TaqMan® siRNA Assays matched to *Silencer*® Select siRNAs
TaqMan® Long ncRNA Assays (page 214)	•Real-time PCR assays for ncRNA >200 nucleotides

TaqMan® MicroRNA and pri-MicroRNA Assays and Arrays

Convenient, scalable solutions for miRNA quantitation, validation, and profiling

MicroRNAs (miRNAs) are naturally occurring non-coding RNAs that play a role in gene regulation. They are highly conserved, single-stranded RNAs (~22 nucleotides) cleaved from larger hairpin precursor transcripts. miRNAs are involved in the RNA interference pathway and affect gene regulation by cleaving or, more often, repressing the translation of their messenger RNA (mRNA) targets.

TaqMan® Assays feature:

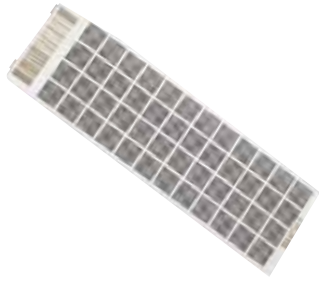
- Specificity—quantitate only biologically active mature miRNAs
- Sensitivity—minimal total RNA input requirements conserve limited samples
- Wide dynamic range—up to 9 logs—to detect high and low expressors in a single experiment
- Fast, simple, and scalable—two-step RT-qPCR assay helps to quickly provide high-quality results
- Megaplex™ Primer Pools—an ideal solution for human, mouse, and rat miRNA profiling

TaqMan® MicroRNA Assays—a real-time PCR revolution

TaqMan® MicroRNA Assays incorporate a target-specific stem–loop reverse transcription primer to address a fundamental problem in miRNA quantitation: the short length of mature miRNAs (~22 nucleotides) prohibits conventional design of a random-primed RT step followed by a specific real-time assay. The stem–loop structure provides specificity for only the mature miRNA target and forms an RT primer/mature miRNA chimera that extends the 3´ end of the miRNA. The resulting longer RT product presents a template amenable to standard real-time PCR using TaqMan® Assays. TaqMan® pri-MicroRNA Assays are available for specific detection of the primary form of RNA.

The flexibility to help meet your research needs

Individual TaqMan® MicroRNA Assays are available for targeted quantitation work¬flows, while TaqMan® Array MicroRNA Cards and TaqMan® OpenArray® MicroRNA Panels enable profiling of hundreds of miRNAs in a single experiment using human, mouse, or rat samples. TaqMan® Array MicroRNA Cards are ideal for studies containing a limited number of samples and deliver the broad dynamic range for which TaqMan® chemistry is known. For larger studies requiring high sample throughput, TaqMan® OpenArray® MicroRNA Panels enable profiling of up to 36 samples per 8-hour working day.



TaqMan® formats. TaqMan® MicroRNA Assays provide superior sensitivity and specificity in single-tube, 384-well microfluidic card, and OpenArray® Plate formats.

Applied Biosystems

	Synthetic template			
Assay	let-7a	let-7b	let-7c	let-7d
let-7a	100%	0%	3%	1%
let-7b	0%	100%	7%	0%
let-7c	0%	2%	100%	0%
let-7d	3%	0%	1%	100%

Vendor 1

	Synthetic template			
Assay	let-7a	let-7b	let-7c	let-7d
let-7a	100%	4%	51%	3%
let-7b	0%	100%	23%	0%
let-7c	2%	83%	100%	0%
let-7d	Assay not available			

Vendor 2

	Synthetic template			
Assay	let-7a	let-7b	let-7c	let-7d
let-7a	100%	0%	0%	1%
let-7b	0%	100%	0%	0%
let-7c	0%	2%	100%	0%
let-7d	24%	0%	0%	100%

hsa-let-7a UGAGGUAGUAGGUUGUAUAGUU
hsa-let-7b UGAGGUAGUAGGUUGUGUGGUU
hsa-let-7c UGAGGUAGUAGGUUGUAUGGUU
hsa-let-7d AGAGGUAGUAGGUUGGAUAGUU

Indicates a perfect match
Indicates low cross-reactivity
Indicates high cross-reactivity

Single-base discrimination of individual TaqMan® MicroRNA Assays compared with two competitors. Relative detection (%) is calculated based on Ct difference between perfectly matched and mismatched assays, where a relative detection of 50% is assigned for each C_t difference of 1.

Product	Description	Profiling	Targeted quantitation
TaqMan® miRNA Assays	Single-tube miRNA assays		●
TaqMan® Array miRNA Cards	384-well microfluidic cards	●	
TaqMan® Array miRNA Panels	OpenArray® Panels; run up to 3 samples in parallel per plate; up to 36 samples per 8-hour day.	●	
Megaplex™ Primer Pools	•Megaplex™ RT Primers in single-reaction solution or Megaplex™ RT Primer Pools. •Megaplex™ PreAmp Primers for detection of small sample miRNA; matched to content in RT Primers.	●	
TaqMan® miRNA Assay Controls	Control assays for human, mouse, rat, *Drosophila*, *C. elegans*, and *Arabidopsis*		●
TaqMan® miRNA Reverse Transcription Kit	Kit with all components needed to convert miRNA to cDNA; can be used with a single RT primers provided with each assay, or with Megaplex™ RT primers	●	●

Also available in 384-well arrays

TaqMan® MicroRNA Arrays and OpenArray Plates provide all the advantages of TaqMan® MicroRNA Assays in a convenient, preconfigured 384-well microfluidic card, reducing experimental variability and the effort required to run 384 assays in parallel.

Combine with Megaplex™ Primer Pools to simplify and accelerate miRNA profiling by reducing hands-on time and the amount of sample you need to as little as 1 ng of total RNA—up to 100 times less material than other methods.

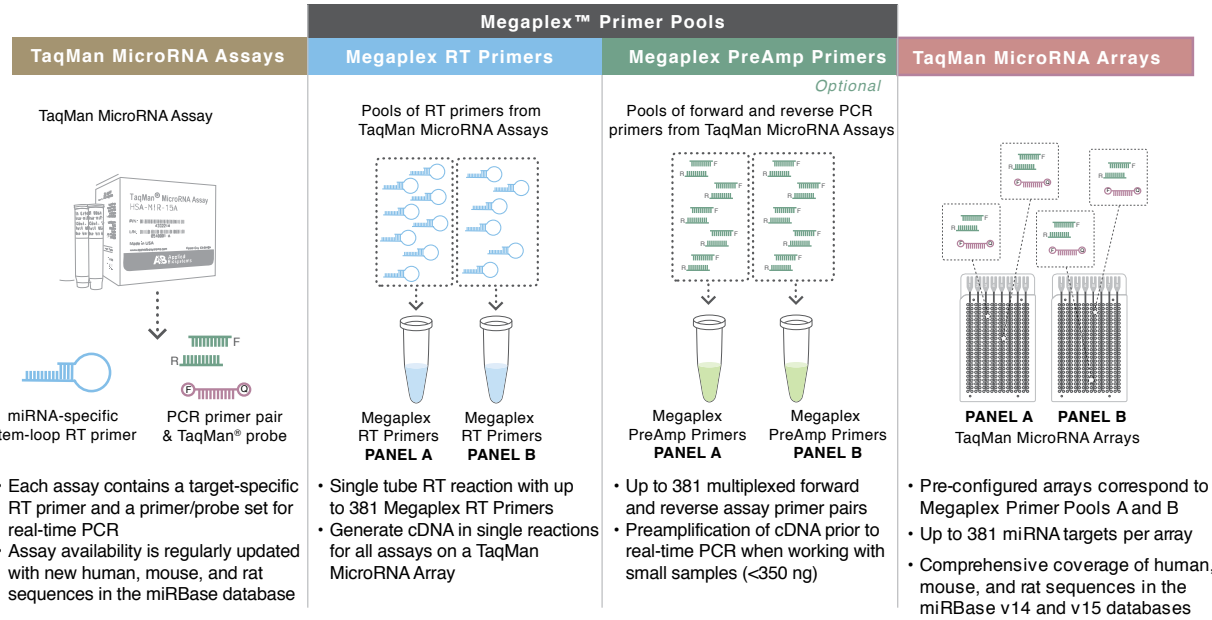
Megaplex™ Primer Pools

Megaplex Primer Pools provide comprehensive coverage of Sanger miRBase v14 for human and v15 for rodent species, and when used with TaqMan® MicroRNA Arrays, offer an ideal microRNA profiling solution.

Megaplex™ Primer Pools offers:

- High specificity—quantitate only the biologically active mature miRNAs
- Highly streamlined workflow reduces the number of RT (reverse transcription) reactions per sample
- Significant reduction in sample consumption, particularly when adding the optional preamplification step
- Comprehensive coverage is ideal for human, mouse and rat miRNA profiling

Megaplex™ RT Primers are sets of two predefined pools (Pool A and Pool B) of up to 380 stem-looped RT primers per pool that enable the simultaneous synthesis of cDNA for mature miRNAs. Megaplex™ PreAmp Primers are sets of two pools (Pool A and Pool B) of gene-specific forward and reverse primers for use with very small quantities of starting material. These primers significantly enhance the ability to detect low expressed miRNAs enabling the generation of a comprehensive expression profile from as low as 1 ng of input total RNA. Megaplex™ Primer Pools are matched to 381 unique TaqMan® MicroRNA Assays, reducing setup time and experimental variability.



Megaplex™ Primer Pools bring the power of real-time PCR to microRNA profiling experiments.

Product ordering

TaqMan® MicroRNA and pri-MicroRNA Assays

Choose from a collection of inventoried assays for human and rodent miRNAs, and over 10,000 made-to-order assays for all other miRNAs in the Sanger registry. Each assay provides 50 RT (15 µL) and 150 real-time PCR (20 µL) reactions. Order online at www.appliedbiosystems.com.

TaqMan® MicroRNA Cards

Ideal for smaller studies or when sample amounts are limiting in human and rodent species. Choose the full 2-card set for comprehensive cov¬erage or either card individually for a more focused view, depending on study needs. Each card enables up to 381 TaqMan® MicroRNA Assays to be run in parallel. Order online at www.appliedbiosystems.com.

TaqMan® OpenArray® MicroRNA Panels

Provides TaqMan® MicroRNA Assays in an OpenArray® Plate for a relevant human miRNA profile. Ideal for medium to large study sizes.

TaqMan® MicroRNA Endogenous Controls

Each endogenous control includes a 20X TaqMan® Assay and a 5X reaction RT primer, providing 50 RT (15 µL) and 150 real-time PCR (20 µL) reactions.

Megaplex™ Primers

Required for up-front reverse transcription of miRNA prior to analysis using either TaqMan® Array MicroRNA Cards or TaqMan® OpenArray® MicroRNA Panels.

Product	Quantity	Cat. No.
Human		
Megaplex™ PreAmp Primers, Human Pool A v2.1	50 reactions	4399233
Megaplex™ PreAmp Primers, Human Pool B v3.0	50 reactions	4444303
Megaplex™ PreAmp Primers, Human Pool Set (A+B) v3.0	50 reactions	4444748
Megaplex™ RT Primers, Human Pool A v2.1	50 reactions	4399966
Megaplex™ RT Primers, Human Pool B v3.0	50 reactions	4444281
Megaplex™ RT Primers, Human Pool Set (A+B) v3.0	50 reactions	4444745
Megaplex™ Primer Pools, Human Pools A v2.1	50 reactions	4401009
Megaplex™ Primer Pools, Human Pools B v3.0	50 reactions	4444749
Megaplex™ Primer Pools, Human Pools Set (A+B) v3.0	50 reactions	4444750
Rodent		
Megaplex™ PreAmp Primers, Rodent Pool A	50 reactions	4399203
Megaplex™ PreAmp Primers, Rodent Pool B v3.0	50 reactions	4444308
Megaplex™ PreAmp Primers, Rodent Pool Set (A+B) v3.0	50 reactions	4444747
Megaplex™ RT Primers, Rodent Pool A	50 reactions	4399970
Megaplex™ RT Primers, Rodent Pool B v3.0	50 reactions	4444292
Megaplex™ RT Primers, Rodent Pool Set (A+B) v3.0	50 reactions	4444746
Megaplex™ Primer Pools, Rodent Pools A	50 reactions	4401090
Megaplex™ Primer Pools, Rodent Pools B v3.0	50 reactions	4444752
Megaplex™ Primer Pools, Rodent Pools Set (A+B) v3.0	50 reactions	4444766

Custom TaqMan® Small RNA and siRNA Assays

Matched to *Silencer*® Select siRNAs or custom-designed for any small ncRNA

Custom TaqMan® Small RNA Assays feature:

- Specificity—distinguish mature miRNAs from precursors
- Sensitivity—require as little as 1–10 ng of input RNA
- Accurately quantitate small RNAs across wide dynamic range, in the same experiment
- Results in as few as 3 hours

Custom TaqMan® Small RNA Assays are ideal for analysis of any *Silencer*® Select siRNAs or other small nucleic acid 17–200 bases in length. Because of their small size, miRNAs and other small RNAs present a challenge for PCR amplification. The design of Custom TaqMan Small RNA Assays is based on novel adaptations used to design TaqMan® miRNA and siRNA Assays making them ideal for the analysis of any small nucleic acid.

Target application

Custom TaqMan® Small RNA and siRNA Assays can be used for novel small RNA discovery and to detect and quantitate endogenous small RNAs. Also, they may be used to facilitate RNAi experiments by evaluating delivery and half-life of exogenous small RNAs, such as siRNA or shRNA, and to measure siRNA knockdown efficacy by running in parallel with *Silencer*® Select siRNAs.

Custom TaqMan® Small RNA and siRNA Assays include a gene-specific stem-loop reverse transcription (RT) primer, in addition to forward and reverse PCR primers, and a TaqMan® probe. The stem-loop structure of the RT primer provides specificity for mature miRNA or processed shRNA genes. It also serves to extend the 5´ end of the small RNA, providing a template that is amenable to standard TaqMan® Assay–based real-time PCR. This target-specific approach to driving reverse transcription sets apart the Life Technologies strategy for small RNA amplification from other commercially available products.

Like TaqMan® MicroRNA Assays, Custom TaqMan® Small RNA Assays are provided in two tubes: one containing the RT primer, and the other containing the specific preformulated TaqMan® Assay (TaqMan® probe and PCR primer set).

Product	Quantity	Cat. No.
Custom TaqMan® Small RNA Assays (extra small scale)	75 reactions	4440418
Custom TaqMan® Small RNA Assays (small scale)	150 reactions	4398987
Custom TaqMan® Small RNA Assays (medium scale)	750 reactions	4398988
Custom TaqMan® Small RNA Assays (large scale)	2,900 reactions	4398989

Custom TaqMan® Small RNA and siRNA Assays are available at www.appliedbiosystems.com

TaqMan® Non-coding RNA Assays

Specific and reproducible quantification of long ncRNA expression levels

Features of the TaqMan® Non-coding RNA Assays include:

- Based on proven TaqMan® Assay technology and use the existing assay design pipeline
- Enable specific and reproducible quantification of long non-coding RNA expression levels
- Available for human, mouse, and rat species

These assays are based on proven TaqMan® Assay technology, leveraging the existing TaqMan® Gene Expression Assay design pipeline to generate gold standard assays to accurately detect ncRNA targets. Only Life Technologies offers TaqMan® Assays for measuring expression of long ncRNAs. TaqMan® Non-coding RNA Assays are available as arrays or individual assays. The table below shows the number of available TaqMan® Non-coding RNA Assays for human, mouse, and rat species.

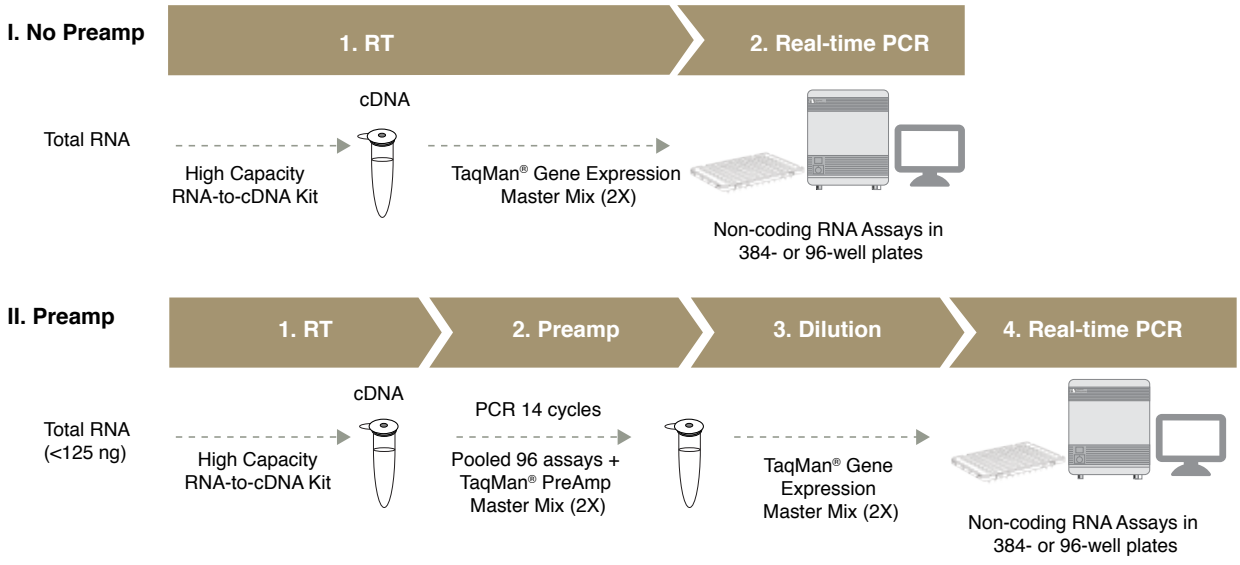
ncRNA Assay design and selection

TaqMan® Non-coding RNA Assays are designed using design algorithms similar to those used for TaqMan® Gene Expression Assays. One of the main differences between our gene expression assays and the new non-coding RNA assays is that the non-coding assays were designed to only detect non-coding transcript targets. In contrast, coding assays are designed to detect one or more coding transcripts of the same gene. Annotation for the new assays comes from NCBI and the RNAdb (http://research.imb.uq.edu.au/rnadb/), the most comprehensive non-coding RNA database available. In addition, to help facilitate the microarray validation workflow, we have designed a number of assays for transcripts that are available on Invitrogen™ NCode™ Non-coding RNA Arrays, for more information, go to www.invitrogen.com.

Species	TaqMan® Long ncRNA Assays*
Human	13,650
Mouse	10,248
Rat	47

* Quantity of assays as of June 2011. Custom assays available. Inquire at www.appliedbiosystems.com.

Product	Quantity	Cat. No.
TaqMan® Non-coding RNA Assays	360 x 20-µL reactions	4426961
	750 x 20-µL reactions	4426962
	2,900 x 20-µL reactions	4426963



*mir*Vana™ miRNA Mimics and Inhibitors

For artificial up- and down-regulation of target mRNA translation

- Versatile—functionally study specific miRNAs in cell-based or *in vivo* systems
- Powerful—validatate miRNA regulation of gene expression
- High-throughput—libraries for effective screening of multiple miRNAs simultaneously
- Current—content regularly updated with Sanger miRBase sequence database

*mir*Vana™ Mimics and Inhibitors are chemically modified, synthetic nucleic acids designed to either mimic mature miRNAs, or to bind to and inhibit endogeneous miRNAs. These *mir*Vana™ products provide a means to functionally study the role of specific miRNAs within cellular systems, or to validate the role of miRNAs in regulating target genes. *mir*Vana™ miRNA Mimics and Inhibitors can be used *in vitro* and *in vivo* and have been validated with Lipofectamine® RNAiMAX Transfection Reagent for use in cell-based systems, and with Invivofectamine® 2.0 Transfection Reagent for *in vivo* delivery (Figure 2). *In vivo* ready *mir*Vana™ miRNA Mimics and Inhibitors have been purified by HPLC and dialysis, making them ready for immediate *in vivo* use.

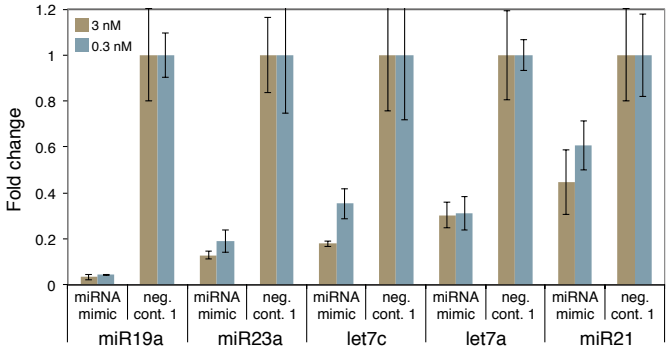


Figure 2. *mir*Vana™ miRNA Mimics display high functionality in reporter assays. *mir*Vana™ miRNA Mimics or *mir*Vana™ miRNA Mimics, Negative Control #1, along with corresponding reporter constructs, were co-transfected using Lipofectamine® 2000 reagent into HeLa cells at 3 and 0.3 nM concentrations. Activity was measured 24 hr later by luciferase assay; down-regulation for miR21 >2-fold, for let7a >3-fold, for let7c >7-fold, for miR23a >7-fold, and for miR19a >5-fold (at 3 nM).

*mir*Vana™ miRNA Mimics

*mir*Vana™ miRNA Mimics are small, chemically modified double-stranded RNA molecules that are designed to mimic endogenous mature miRNAs with maximum specificity. The chemical modifications in *mir*Vana™ miRNA Mimics inactivate the star strand, thus ensuring the guide strand (representing the desired mature miRNA) is taken up into RISC-like responsible for miRNA activity. *mir*Vana™ miRNA Mimics give maximum and consistent effects at concentrations as low as 0.3 mM (Figure 2). *mir*Vana™ miRNA Mimics are available individually or as libraries.

*mir*Vana™ miRNA Mimic Positive and Negative Controls

The *mir*Vana™ miRNA Mimic miR-1 Positive Control is used in experiments utilizing *mir*Vana™ miRNA Mimics. This mimic effectively down regulates the expression of twinfilin-1, also known as PTK9, at the mRNA level (60–95% reduction). *mir*Vana™ miRNA Mimic Negative Control #1 is a random-sequence miRNA mimic molecule that has been extensively tested in human cell lines and tissues and validated to not produce identifiable effects on known miRNA function.

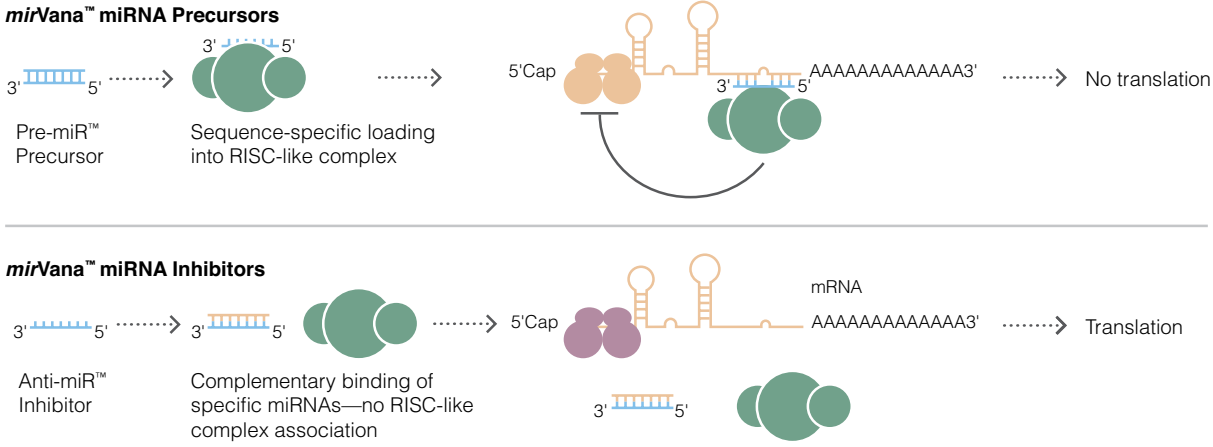


Figure 1. Activity of *mir*Vana™ miRNA Mimics and *mir*Vana™ miRNA Inhibitors.

*mir*Vana™ miRNA Inhibitors

*mir*Vana™ miRNA inhibitors are chemically modified, single-stranded nucleic acids designed to specifically bind to and inhibit endogenous miRNAs. They have the highest potency *in vitro* inhibition at the lowest miRNA inhibitor concentration of available miRNA mimics. *mir*Vana™ miRNA Inhibitors are available individually or as libraries.

*mir*Vana™ miRNA Inhibitor Positive and Negative Controls

The *mir*Vana™ miRNA inhibitors let-7c Positive Control miRNA inhibitor provides a convenient, validated positive control for experiments using *mir*Vana™ miRNA inhibitors. Endogenous let-7 miRNA negatively regulates HMGA2, a ubiquitously expressed nonhistone chromatin protein that modulates gene expression through changes in chromatin architecture. The *mir*Vana™ miRNA Inhibitor Negative Control #1 is a random sequence that has been extensively tested in human cell lines and tissues and validated to not produce identifiable effects on known miRNA function.

*mir*Vana™ miRNA Mimic and *mir*Vana™ miRNA Inhibitor Libraries

Libraries of the *mir*Vana™ miRNA Mimics and *mir*Vana™ miRNA Inhibitors are available. Each library is derived from miRNAs catalogued in version 17 of the miRBase sequence database. Each *mir*Vana™ miRNA Mimic or Inhibitor included in the respective library is provided in a quantity of 0.25 nmol, dried in individual wells of a 96-well plate. *mir*Vana™ miRNA Mimics are supplied in a quantity sufficient for 250 transfections when used at 10 nM; while each *mir*Vana™ miRNA Inhibitor is supplied in a quantity sufficient for 50 transfections when used at 50 nM each.

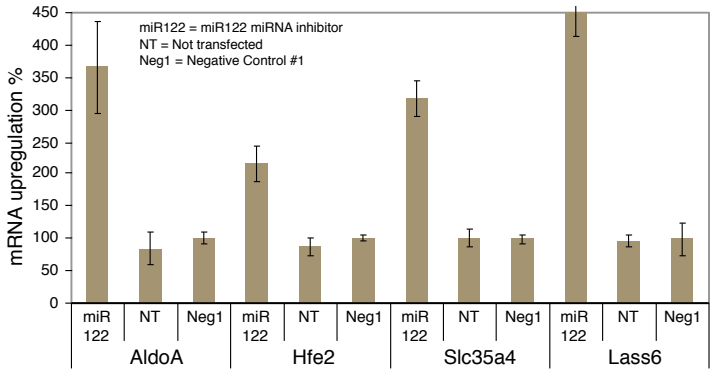


Figure 3. *mir*Vana™ miRNA Inhibitors effectively suppress miRNA *in vivo*. miR122 or Negative Control #1 *mir*Vana™ miRNA inhibitors were complexed with Invivofectamine® 2.0 Reagent and delivered to Balb-C mouse liver via tail vein injection on three consecutive days at a dose of 7 mg/kg body weight. Expression of four mRNA targets (AldoA, Hfe2, Slc35a4 and Lass6), natural targets of miR122, were measured in transfected livers of mice injected with (miR122 miRNA inhibitor or Negative Contol #1 (Neg 1)) and livers of mice that were not transfected (NT) using TaqMan® MicroRNA Assays. Significant up-regulation mRNA was detected in livers of mice treated with miR122, compared to untreated and negative control-treated mice; >3.5-fold for AldoA, >2-fold for Hfe2, >3-fold for Slc35a4, and >4-fold for Lass6. This indicates that mirVana™ miRNA inhibitors are efficiently delivered to the liver with Invivofectamine® 2.0 Reagent and inactive miR122, leading to up-regulation of genes naturally suppressed by miR122.

Individual *mir*Vana™ miRNA Mimics, *mir*Vana™ miRNA Inhibitors, and libraries

To order *mir*Vana™ miRNA Mimics, *mir*Vana™ miRNA Inhibitors, and libraries through our online system, go to www.appliedbiosystems.com.

*mir*Vana™ miRNA mimics, *mir*Vana™ miRNA inhibitors Positive and Negative Controls

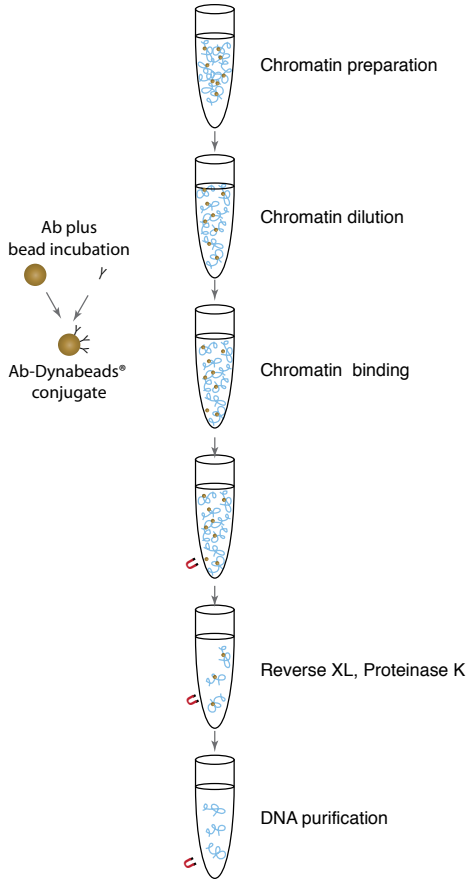
Product	Quantity	Cat. No.
*mir*Vana™ miRNA mimic, Negative Control #1	5 nmol	4464058
*mir*Vana™ miRNA mimic, Negative Control #1	20 nmol	4464059
*mir*Vana™ miRNA mimic, Negative Control #1, *in vivo* ready	50 nmol	4464060
*mir*Vana™ miRNA mimic, Negative Control #1, *in vivo* ready	250 nmol	4464061
*mir*Vana™ miRNA mimic, miR-1 Positive Control	5 nmol	4464062
*mir*Vana™ miRNA mimic, miR-1 Positive Control	20 nmol	4464063
*mir*Vana™ miRNA mimic, miR-1 Positive Control, *in vivo* ready	50 nmol	4464064
*mir*Vana™ miRNA mimic, miR-1 Positive Control, *in vivo* ready	250 nmol	4464065
*mir*Vana™ miRNA inhibitor, Negative Control #1	5 nmol	4464076
*mir*Vana™ miRNA inhibitor, Negative Control #1	20 nmol	4464077
*mir*Vana™ miRNA inhibitor, Negative Control #1, *in vivo* ready	50 nmol	4464078
*mir*Vana™ miRNA inhibitor, Negative Control #1, *in vivo* ready	250 nmol	4464079
*mir*Vana™ miRNA inhibitor, let-7c Positive Control	5 nmol	4464080
*mir*Vana™ miRNA inhibitor, let-7c Positive Control	20 nmol	4464081

Overview of DNA methylation and chromatin remodeling

Chromatin is a complex of DNA and histone proteins where DNA is compacted, folded, and organized in the nuclei of eukaryotic cells. Changes to the chromatin structure (chromatin remodeling) may be the result of epigenetic changes and effect gene transcription by loosening the complex, allowing other proteins such as transcription factors access to the DNA. Alternatively, if remodeling causes the complex to become more closed, it may block transcription resulting in loss of gene expression. Several epigenetic mechanisms introduce variation to chromatin structure, including covalent histone modifications, histone variant composition, DNA methylation, and non-coding RNA. Chromatin remodeling studies are key in understanding the regulation of gene expression and are accomplished through two main mechanisms: DNA methylation and posttranslational modification of the amino acids that make up histone proteins.

There are many methods used to study DNA methylation. Some methods depend upon enrichment of methylated DNA, while others require treatment of DNA with bisulfite that converts unmethylated cytosines to uracils (methylated cytosines remain unchanged). Both methods prepare samples compatible with various platforms for detection and analysis, including capillary sequencing, next-generation sequencing, DNA microarrays, PCR, and real-time PCR.

Methods to study post-translational modifications of histone proteins include standard protein detection techniques such as western blotting and immunoassays, as well as chromatin Immunoprecipitation (ChIP) and high resolution melt (HRM) analyses. ChIP is an immunoprecipitation technique used to investigate the interaction between proteins and DNA in the cell and can be used to determine the specific location of histone modifications (see figure). HRM analysis is a real-time PCR technique that uses the differences in melting temperature of methylated and unmethylated DNA samples to detect methylation.



The ChIP technique. A chemical fixation step with UltraPure™ reagents is used to covalently crosslink a transcription factor to a promoter sequence, followed by sonication/fragmentation of DNA for bead-based immunoprecipitation.

Life Technologies provides several products for the analysis of DNA methylation and chromatin remodeling, including:

DNA methylation system	Features	Downstream detection	Page
MethylMiner™ Methylated DNA Enrichment Kit	• Enriches and fractionates double-stranded DNA based on CpG methylation density • Upstream of bisulfite conversion • Analyze methylation densities of interest	• Real-time PCR, PCR, next-generation sequencing, direct sequencing, DNA microarray	218
Cells-to-CpG™ Bisulfite Conversion Kit	• Identify specific methylation patterns in DNA • Bisulfite conversion of DNA samples • Converts unmethylated cytosines to uracils but does not change methylated cytosines	• PCR, next-generation sequencing, capillary electrophoresis sequencing	219
MeltDoctor™ HRM real-time PCR reagents	• For post-PCR analysis of methylation • differences in DNA samples	• Real-time PCR	220

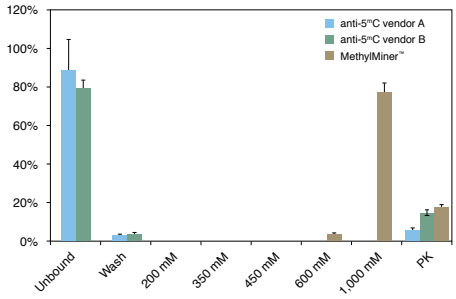
MethylMiner™ Methylated DNA Enrichment Kit

Enrichment of methylated DNA for downstream analysis

The MethylMiner™ Methylated DNA Enrichment Kit is for highly sensitive enrichment of methylated DNA for downstream analysis including PCR/qPCR based assays, bisuflite conversion followed by amplification, cloning, and sequencing, direct sequencing, library preparation for high-throughput sequencing, and sample-prep for DNA microarray analysis. Advantages of MethylMiner™ Methylated DNA Enrichment Kit include:

- Precise answers—distinctions for methylation status and density
- Greater sensitivity than antibody-based methods (see figure)
- Fast protocol—completed in less than 4 hours
- Ligate double-stranded adaptors for next-generation sequencing
- No need for proteinase K treatment or phenol/chloroform extraction

Patterns of DNA methylation may influence development and diseases such as cancer. MethylMiner™ Methylated DNA Enrichment Kit enables superior enrichment and differential fractionation of double-stranded DNA based on CpG methylation density, with increased sensitivity over antibody-based methods. Fractionation permits important comparisons between samples and enables researchers to focus analysis on only the methylation densities of interest.



MethylMiner™ Methylated DNA Enrichment Kit captures more heavily methylated DNA compared to an antibody-based method. As shown, ~100% of the heavily methylated DNA sequence was captured by the MethylMiner™ beads, and ~80% could be eluted with 1 M NaCl. In contrast, the anti-5mC antibody from two different vendors could only capture ~10–20% of this heavily methylated DNA, and elution could only be achieved with proteinase K treatment. Experiments were conducted using the provided protocols.

Product	Quantity	Cat. No.
MethylMiner™ Methylated DNA Enrichment Kit	1 kit	ME10025

Methyl Primer Express® Software v1.0

This free software enables you to design high-quality PCR primers for methylation mapping experiments. Simply cut and paste your region of interest. The tool searches for CpG islands and simulates bisulfite modification of DNA *in silico*. Methyl Primer Express® Software v1.0 enables you to:

- Easily design robust primers for methylation studies
- Annotate the transcription start base and translation start codon
- Highlight target regions of interest and examine these boundaries in your primer design reports
- Predict CpG islands in your DNA sequence
- Simulate the bisulfite modification of DNA
- Create a "reference" sequence for downstream sequencing analysis

Create information-rich reports

Create primer design reports containing your sample's initial DNA sequence, its bisulfite-modified sequence, a CpG density representation, the location of primers in the initial sequence, and both the primer sequence and its associated characteristics.

Primer design for MS-HRM analysis

Unmethylated fragments are amplified more efficiently during PCR than methylated fragments, affecting assay sensitivity, particularly at the lower limits of detection. To accurately detect methylation in the 0–2% range, CpG dinucleotides should be included in the PCR primer sequences. CpG sequences bias the amplification towards the methylated fragments of DNA but can negatively affect the sensitivity of detection. Using Methyl Primer Express® Software v1.0, primers can be designed with a specified number of CpG dinucleotides for optimized reactions.

To download this free software, go to www.appliedbiosystems.com.

Cells-to-CpG™ Bisulfite Conversion Kit

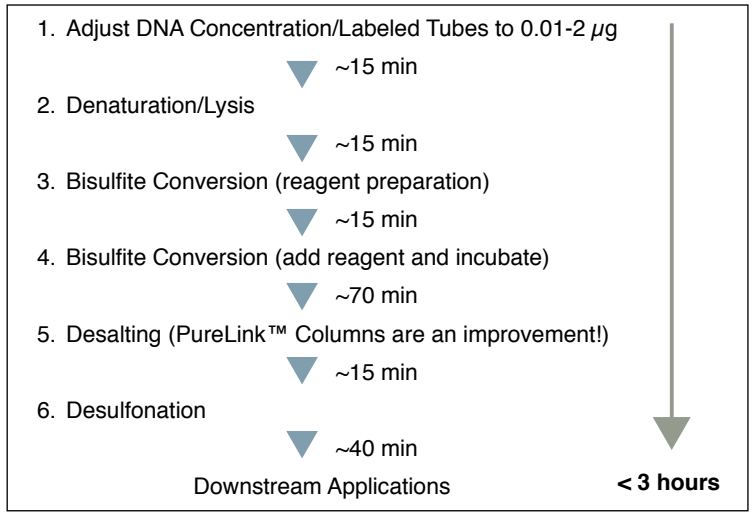
For the identification of methylation patterns in DNA

Cells-to-CpG™ Bisulfite Conversion Kit offers:

- Flexible DNA conversion—convert unmethylated cytosines to uracils without optimization
- Quantitative reliability—confidence in results with available controls to easily monitor conversion rate
- Streamlined workflow—ability to start with various sample input types including cell, tissue, blood, and FFPE samples
- Efficient application—reduced time and labor through direct conversion of cytosines in samples without need for purifying genomic DNA
- Versatile downstream applications—converted DNA suitable for analysis using real-time PCR, next generation sequencing, PCR, or capillary electrophoresis

The bisulfite method is the most commonly used technique for identifying specific methylation patterns within a DNA sample. It consists of treating DNA with bisulfite, which converts unmethylated cytosines to uracils but does not change methylated cytosines. Bisulfite conversion has been utilized in DNA methylation research for the last 20 years, with few improvements to the technology until now. With thorough optimization, the Cells-to-CpG™ Bisulfite Conversion Kit provides a quick, streamlined method (see figure) for bisulfite conversion to reveal methylated cytosines in either loci-specific or genome-wide analyses. DNA treated with the kit is suitable for high resolution melting (HRM) analysis or sequencing.

Sufficient materials are supplied in the Cells-to-CpG™ Bisulfite Conversion Kit (2 x 96) to perform bisulfite conversion of 192 samples.



Streamlined bisulfite conversion workflow with Cells-to-CpG™ Bisulfite Conversion Kit. Starting with cells, tissues, blood, FFPE samples, or purified DNA, directly lyse and denature sample for subsequent bisulfite conversion, desalting, and desulfonation. The protocol takes less than 3 hours, which is a significant reduction in time compared to other available methods that require at least 18 hours.

Product	Quantity	Cat. No.
Cells-to-CpG™ Bisulfite Conversion Kit	50 reactions	4445555
	2 x 96 preps	4445554
Cells-to-CpG™ Bisulfite Conversion and Quantitation Control Kit	50 µL	4445553
Cells-to-CpG™ Methylated and Unmethylated gDNA Control Kit	20 reactions	4445552

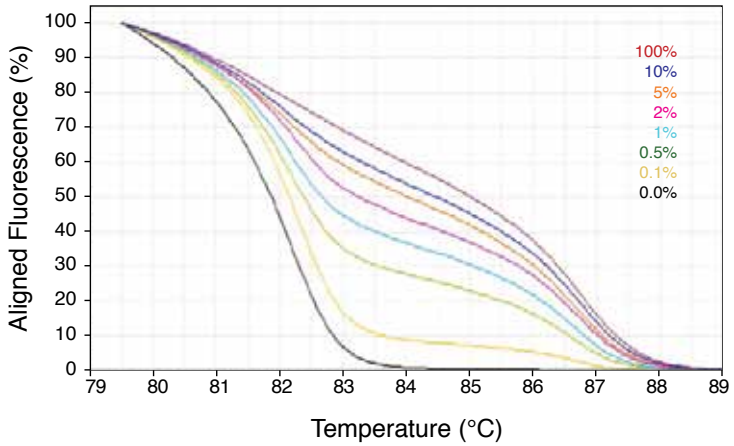
MeltDoctor™ HRM reagents

Simple and fast post-PCR identification of genetic variation

High resolution melting (HRM) analysis is a post-PCR method used for identifying genetic variation in nucleic acid sequences, such as methylation differences between samples and mutation analysis. Simple and fast, this method is based on PCR melting (dissociation) curve techniques. HRM analysis can discriminate DNA sequences based on their composition, length, GC content, or strand complementarity.

MeltDoctor™ HRM reagents include MeltDoctor™ HRM dye, a stabilized form of the dsDNA-binding dye SYTO®-9. This next-generation dye delivers sharp, clean melt profiles. The MeltDoctor™ HRM Master Mix is simple to use and requires only the addition of template DNA and a PCR primer pair before starting the PCR. MeltDoctor™ HRM Master Mix employs hot start–enabled DNA polymerase, minimizing nonspecific product formation and enabling reactions to be set up at room temperature.

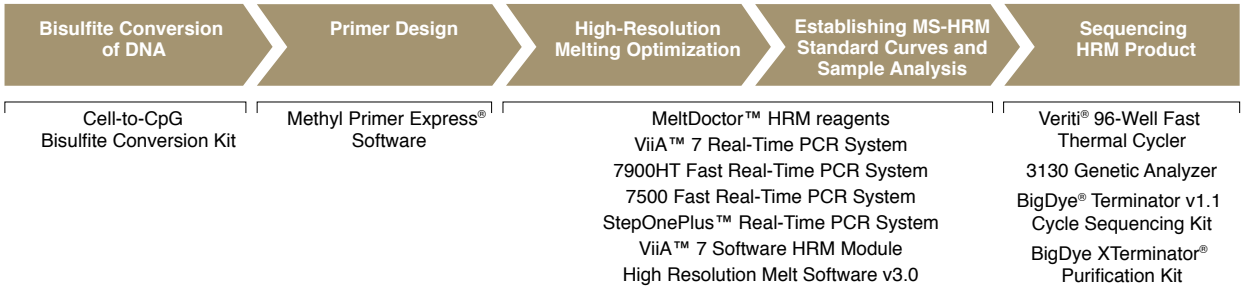
MeltDoctor™ HRM Reagent Kit provides individually packaged PCR components and the MeltDoctor™ HRM Dye. MeltDoctor™ HRM Master Mix is a pre-mix of all components (excluding template and primers).



MS-HRM standard curves with high resolution between 0 and 2%. 100% methylated DNA was mixed in different ratios with DNA from HT29 cells, in which the RASGRF1 promoter is 0% methylated. The standard curves for methylation were generated with a 110 bp PCR fragment containing 9 CpG dinucleotides.

Methyl Primer Express® Software is ideal for primer design when using methylation-sensitive high resolution melt analysis (MS-HRM). For more information, see page 218. To download the free software, go to www.appliedbiosystems.com.

Product	Quantity	Cat. No.
MeltDoctor™ HRM Reagent Kit	1 kit	4425557
MeltDoctor™ HRM Master Mix	5 mL	4415440
	5 x 5 mL	4415452
	10 x 5 mL	4415450
	50 mL	4409535
MeltDoctor™ HRM Positive Control Kit	1 kit	4410126
MeltDoctor™ HRM Calibration Standards	1 tube	4425562



Applied Biosystems Workflow for Methylation-Sensitive High-Resolution Melting (MS-HRM) Analysis Followed by DNA Sequencing.

MAGnify™ Chromatin Immunoprecipitation (ChIP) System

Fast and efficient preparation of ChIP DNA for downstream analysis

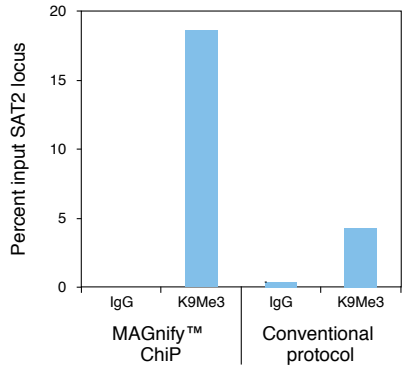
Features of the MAGnify™ Chromatin Immunoprecipitation System include:

- Reduced overall ChIP protocol time by one day
- Reduced input cell number per ChIP experiment (10,000–300,000 cells required)
- Decreased background and improved reproducibility
- Easily increased throughput with small volumes, magnetic protocol, and magnet compatible with multi-channel pipetting
- Reduced experimental error with Dynabeads® Protein A/G Mix—worry less about antibody compatibility

The MAGnify™ Chromatin Immunoprecipitation System provides a streamlined, optimized assay for the enrichment of chromatin/protein complexes and DNA recovery using magnetic bead capture technology. The isolated DNA is ready for downstream analysis by methods such as PCR- or qPCR-based assays, or massive parallel DNA sequencing. This kit enables researchers to start with lower sample amounts than traditional ChIP workflows, thereby preserving precious samples, and the protocol can be completed in a single day, compared with 2–3 days for a traditional ChIP assay. The kit can be used with the suite of ChIP-validated antibodies from Life Technologies (see page 223).

DynaMag™-PCR

The MAGnify™ ChIP system utilizes a novel magnet that is compatible with 0.2-mL PCR strip tubes. This magnet is critical for enabling the use of reduced starting cell numbers and volumes for immunoprecipitation and washing steps. This novel magnetic separator allows you to achieve results quickly and easily while offering the freedom to simultaneously perform multiple ChIP assays in PCR tubes with a multichannel pipettor.



MAGnify™ ChIP vs. conventional protocols. Sheared chromatin from 293Gt cells was prepared, and 50,000 cells were used for MAGnify™ ChIP and 10^6 cells per ChIP for the conventional protocol. Antibody (1 µg of IgG included in kit: H3-K9Me3, Cat. No. 49-1008) was used for each ChIP experiment. 1 % of the sonicated chromatin was set aside as input control. Optimized qPCR primers were used to amplify the SAT2 locus (Cat. No. 49-2026), a known target of heterochromatin-associated H3-K9Me3. Key elements of a conventional protocol use an overnight immunoprecipitation step of antibody and chromatin, followed by enrichment with Protein A sepharose beads and extensive washes with buffers containing variable salt concentrations. Reverse crosslinking was done overnight at 65°C and DNA purification performed by phenol/chloroform extraction.

Comparison of MAGnify™ ChIP to a conventional ChIP protocol.

Workflow step	MAGnify™ ChIP timeline	Conventional ChIP timeline
Preclearing	NA	1–2 hr
Antibody/chromatin incubation	2 hr	Overnight
Bead pulldown	1 hr	2 hr
Washes	30 min (2 buffers)	1–3 hr (4 buffers)
Reverse crosslinking	1.5 hr	Overnight
Proteinase K digestion		2 hr
DNA elution from beads		15–30 min
DNA purification		2 hours–overnight
Average time	5 hours	36–48 hr

Product	Quantity	Cat. No.
MAGnify™ Chromatin Immunoprecipitation System	1 kit	49-2024
DynaMag™-PCR	1 each	49-2025

SOLiD® ChIP-Seq Kit

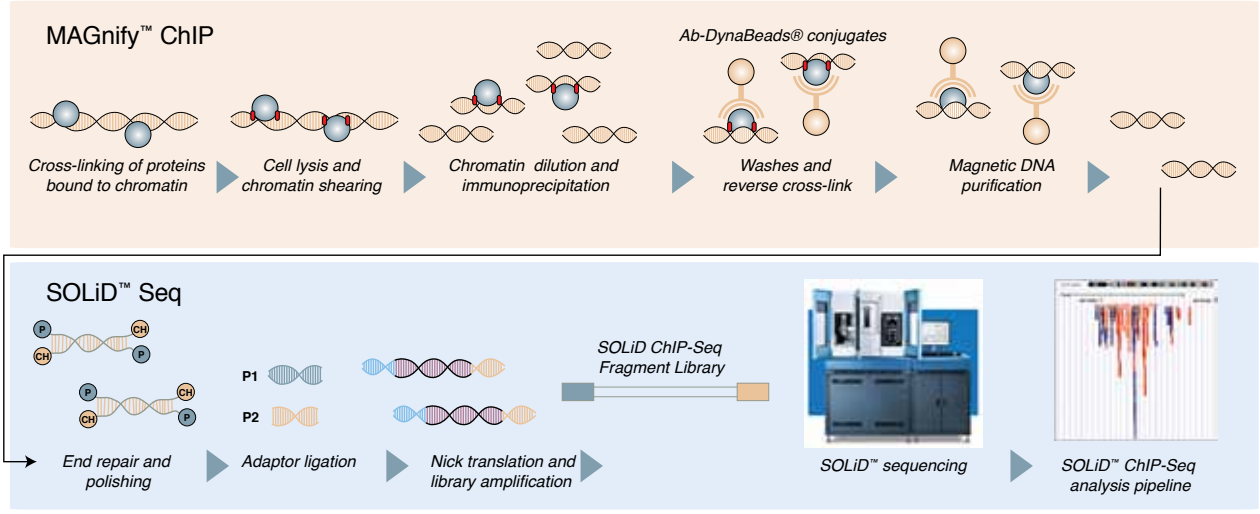
For the sequencing of chromatin immunoprecipitation (ChIP) products

The SOLiD® ChIP-Seq Kit provides:

- Optimized workflow—save an entire day compared to traditional ChIP protocols
- Reliable purification—improved reproducibility due to optimized magnetic DNA purification
- Higher sensitivity—use of fewer cells per ChIP experiment to minimize variability between experiments
- Lower input DNA—library preparation with 1–10 ng of DNA

The SOLiD® ChIP-Seq Kit is an optimized system for genome-wide ChIP analysis on SOLiD® sequencers, which includes ChIP preparation reagents and library generation reagents for 20 samples along with a unique optimized protocol. The simplified workflow involves cross-linking of proteins, cell lysis, and subsequent chromatin shearing and immunoprecipitation utilizing Dynabeads®. The result is purified DNA which can be utilized for DNA fragment library construction utilizing best-in class reagents from the SOLiD® sequencing system.

Product	Quantity	Cat. No.
SOLiD® ChiP-Seq Kit	1 kit	4449640



Antibodies qualified for chromatin immunoprecipitation (ChIP)

Because high-quality antibodies are crucial for a successful ChIP experiment, it is important to select an antibody that has high affinity for the protein or proteins of interest. Unfortunately, the successful use of a specific antibody in other applications (i.e., western blotting) may not guarantee success in ChIP analysis. To ensure success, we have qualified a number of antibodies directed against histones, as well as other chromatin-associated proteins, for use in ChIP analysis.

Product	Host	Quantity	Cat. No.
CIITA (Human)	Rabbit	50 µg	491001
DNMT3B (Mouse)	Rabbit	5 µg	491028
Estrogen Receptor alpha (Human)	Mouse	50 µg	491002
Ezh2 (Human, Mouse)	Rabbit	50 µg	491043
Glucocorticoid Receptor (Human)	Mouse	50 µg	491026
H3 Pan (Human)	Rabbit	100 µL	491038
H3K27me1 (Human)	Rabbit	50 µg	491012
H3K27me2 (Human)	Rabbit	50 µg	491013
H3K27me3 (Human)	Rabbit	100 µL	491014
H3K27me3S28p (Human)	Rabbit	100 µL	491015
H3K36me1 (Human)	Rabbit	50 µg	491016
H3K36me2 (Human)	Rabbit	100 µL	491039
H3K36me3 (Human, Mouse)	Rabbit	50 µg	491017
H3K4me1 (Human)	Rabbit	100 µL	491003
H3K4me2 (Human)	Rabbit	100 µL	491004
H3K4me3 (Human, Mouse)	Rabbit	24 µg	491005
H3K79me1 (Human)	Rabbit	100 µL	491018
H3K79me2 (Human)	Rabbit	100 µL	491019
H3K79me3 (Human, Mouse)	Rabbit	50 µg	491020
H3K9andfrasl;14ac (Human)	Rabbit	44 µg	491010
H3K9ac (Human)	Rabbit	44 µg	491009
H3K9acS10p (Human)	Rabbit	100 µL	491011
H3K9me1 (Human)	Rabbit	100 µL	491006
H3K9me2 (Human)	Rabbit	50 µg	491007
H3K9me3 (Human,Mouse)	Rabbit	50 µg	491008
H3K9me3S10p (Human)	Rabbit	100 µL	491040
H3R17me2 (Human)	Rabbit	100 µL	491021
H3S10p (Human)	Rabbit	100 µL	491041
H4K20me1 (Human)	Rabbit	100 µL	491023
H4K20me3 (Human)	Rabbit	100 µL	491024
H4K8ac (Human)	Rabbit	100 µL	491022
HDAC1 (Human, Mouse)	Rabbit	50 µg	491025
HDAC3 (Human)	Mouse	100 µL	491042
MBD1 (Human)	Rabbit	50 µg	491027
MeCP2 (Human)	Rabbit	50 µg	491029
NF-YB (Human)	Rabbit	100 µg	491030
p53 (Human)	Rabbit	50 µg	491031
p63 (Human)	Rabbit	50 µg	491032
Pol II (Human)	Mouse	100 µL	491033
RFX-AP (Human)	Rabbit	50 µg	491034
Suz12 (Human, Mouse)	Rabbit	50 µg	491035
TBP (Human)	Mouse	100 µg	491036
TIP5 (Human, Mouse)	Rabbit	100 µL	491037

For an up-to-date listing of ChIP-qualified antibodies, go to www.invitrogen.com.

Microarray labeling kits for array gene expression profiling

For siRNA knockdown, miRNA functional, and other gene expression analyses

Life Technologies' microarray analysis reagents deliver reliable and fast RNA amplification and labeling for use with the most popular array platforms. Our reagents come in easy-to-use formats that work with a wide range of RNA inputs and sample types. And because they consistently deliver higher yields and more efficient sample labeling with less hands-on time, you can count on faster, more consistent results.

Ambion® WT Expression Kit

Faster, more sensitive, and reproducible results with Affymetrix® whole transcriptome microarrays

The kit is designed to generate amplified sense-strand cDNA ready for fragmentation and labeling using the Affymetrix® GeneChip® WT Terminal Labeling Kit. The kit has been optimized specifically for use with Affymetrix® GeneChip® Human, Mouse, and Rat 1.0 ST (sense target) Arrays. Ambion® WT Expression Kit offers:

- A novel, patent-pending reverse transcription (RT) priming method eliminating an rRNA depletion step
- Proprietary RT primers designed using an algorithm that eliminates known homologous ribosomal RNA sequences
- Complete and unbiased coverage of the transcriptome, with significantly less rRNA amplification compared to other methodologies
- Consistent results with less input RNA

Product	Quantity	Cat. No.
Ambion® WT Expression Kit	10 reactions	4411973
	30 reactions	4440536
Ambion® WT Expression Kit (for high-throughput robotics)	24 reactions	4440537
	96 reactions	4440536

BioPrime Labeling Systems

BioPrime® Total Genomic Labeling System

A complete DNA labeling for array-based comparative genomic hybridization (aCGH) the BioPrime® Total Genomic Labeling Systems offer users the following advantages:

- Better call rates with precious samples
- Higher signal to noise with Alexa Fluor® 3 and 5 dyes
- Less channel bias due to novel reaction formulation
- Widest range of input material (50 ng–3 µg)
- Simplified workflow with master mix formulation

BioPrime® aCGH Genomic Labeling System

Available in both indirect and direct labeling formats, the BioPrime® Plus Array CGH Genomic Labeling Systems provide a flexible solution for array-based comparative genomic hybridization (aCGH). Using the systems, you can expect:

- High yields of fluorescently labeled genomic DNA
- Signal-to-noise ratios
- Detection of gene copy number variations

BioPrime® Total for Agilent® aCGH

Optimized for the Agilent aCGH workflow, arrays, and scanners, BioPrime® Total for Agilent® aCGH is designed to improve call rates with precious samples. The kit offers users the following advantages:

- Improved call rates from higher signal to noise ratios with Alexa Fluor® 3 and 5 dyes
- Less channel bias on the Agilent aCGH platform due to novel reaction formulation
- Widest range of input material (50 ng–3 µg)
- Cleanup with PureLink™ purification eliminates the need for complicated volume-reduction steps
- Simplified workflow includes master mix formulation and restriction enzymes, AluI and RSAI

Product	Quantity	Cat. No.
BioPrime® Total Genomic Labeling System with purification	30 reactions	18097011
	10 reactions	18097010
BioPrime® Total Genomic Labeling System without purification	30 reactions	18097012
BioPrime® aCGH Genomic Labeling System with purification	30 reactions	18095011
BioPrime® aCGH Genomic Labeling System without purification	30 reactions	18095012
BioPrime® Total FFPE Genomic Labeling System with purification	30 reactions	A10965011
	10 reactions	A10965010
BioPrime® Total for Agilent® aCGH with purification	30 reactions	A10963011
	10 reactions	A10963010

Illumina® TotalPrep™ RNA Amplification Kits

Complete systems for generating biotinylated, amplified RNA for use with Illumina® Sentrix® arrays

Ambion® Illumina® TotalPrep™ RNA Amplification Kits (patent pending) incorporates extensive improvements to the previous Illumina® RNA Amplification Kit. The *in vitro* transcription (IVT) reaction has been optimized for the most effective biotin labeling. An NTP mix containing biotinylated UTP has been added to the kit, allowing ease of use and optimal labeling. These modifications have significantly increased sensitivity, providing increased Percent Present calls and single round amplification with just 50 ng of input RNA.

The Ambion® Illumina® TotalPrep™-96 RNA Amplification Kit is also available. It is a complete system for generating biotinylated, amplified RNA for hybridization with Illumina® Sentrix® arrays. The kit includes sufficient reagents for 96 reactions. The features of kit include:

- Perform amplification from just 50 ng of input RNA
- Developed for expression profiling using the Sentrix® BeadChips
- Bio-16-UTP now included in optimized NTP mix, reducing setup time and maximizing labeling

Product	Quantity	Cat. No.
Illumina® TotalPrep™ RNA Amplification Kit	24 reactions	AMIL1791
Illumina® TotalPrep™-96 RNA Amplification Kit	96 reactions	4393543

For information on our complete selection of microarray labeling kits, go to our selection guide at www.invitrogen.com.

Cot-1 DNA

Human Cot-1 DNA® is commonly used to block nonspecific hybridization in microarray screening.

Product	Quantity	Cat. No.
Human Cot-1 DNA®	500 µg	15279011
Human Cot-1 DNA®-Fluorometric QC 1 mL at 1 mg/mL	1 mg	15279101
Mouse Cot-1 DNA®	500 µg	18440016

protein expression, isolation, and analysis

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Overview of transfection

Reagent-mediated transfection is a fast, simple, and reproducible means for easily introducing DNA, RNA, siRNA, or oligonucleotides into eukaryotic cells. Cationic lipid–mediated gene delivery allows highly efficient transfection of a broad range of cell types, including adherent, suspension, insect, as well as primary cultures. All transfection reagents from Life Technologies include streamlined protocols for maximum convenience and ease of use. Many reagents include protocols for high-throughput use. Life Technologies is the most cited and trusted supplier of transfection reagents with over 45,000 citations for the Lipofectamine® brand alone.

Transfection is usually used in conjunction with an appropriate selection agent to establish stable eukaryotic cell lines.

Choose the transfection reagent optimized for your application.

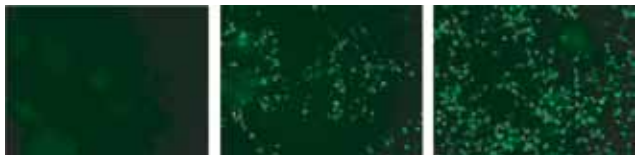
Product	Application and cell type	References
Lipofectamine® LTX Transfection Reagent	High-efficiency transfection of plasmid DNA	See below and page 201
Lipofectamine® RNAiMAX Transfection Reagent	High-efficiency transfection of siRNA into nearly every cell type	See pages 200 and 230
Invivofectamine® 2.0 Reagent	*In vivo* transfection of siRNA into mouse liver cells through a simple systemic injection	See pages 202 and 233
Lipofectamine® 2000 Transfection Reagent	Transfection of both plasmid DNA and siRNA into a broad spectrum of adherent and suspension cell lines	See page 232
FreeStyle™ MAX Transfection Reagent	Animal origin–free reagent for transient transfection of CHO and HEK 293 cells in suspension culture	See page 244

Lipofectamine® LTX Reagent

Maximum protein expression with plasmid delivery

- Superior delivery—transfect a broad range of cell types, including primary and hard-to-transfect cells, with high efficiency
- Superior performance—combine high transfection efficiency with maximal (>90%) cell viability
- Less optimization—rapid, simple protocols provided for many cell lines
- High-efficiency transfection—more stable cell lines

Lipofectamine® LTX Reagent offers the highest level of reproducible, efficient protein expression in all cell types, including primary, hard-to-transfect, and cells of clinical interest (see figure). This enables cells to respond to your assay conditions, which is critical for relevant data from functional genomics expression studies. Lipofectamine® LTX Reagent is synthesized from 100% animal origin–free components, making it easy to validate the absence of zoonotic diseases, such as BSE or viruses, in experiments or cell lines. Use with PLUS™ Reagent to obtain both high levels of protein expression and maximum viability in primary cells. This reagent was designed with the perfect balance of potency while still being gentle to your cells. Lipofectamine® LTX Reagent was tested in >14 primary and disease-related cell types. It delivered higher expression in >90% of them compared to a competitor's products.



0 ng 500 ng 1,000 ng

Lipofectamine® LTX Reagent with PLUS™ Reagent, efficiently transfects primary, neural progenitor cells. Lipofectamine® LTX Reagent (1.5 µL) was used to transfect murine embryonic primary neural progenitor cells with the indicated quantities of a GFP-expressing plasmid in the presence of PLUS™ Reagent (2.5 µL). GFP expression was analyzed 24 hr posttransfection. Data courtesy of the Beverly Davidson laboratory, University of Iowa.

Simple and rapid protocol

Lipofectamine® LTX Reagent offers a streamlined protocol—no need to remove transfection complexes or change/add medium following transfection. You'll spend less hands-on time and get better results faster.

Product	Quantity	Cat. No.
Lipofectamine® LTX and PLUS™ Reagent	1 mL	15338100
Lipofectamine® LTX and PLUS™ Reagent Sample Size	0.1 mL	A12621

For optimized protocols for transfection of a wide range of cell lines, go to www.invitrogen.com/transfection.

Lipofectamine® RNAiMAX Transfection Reagent

Delivers potent gene knockdown with less siRNA

- Compatibility with a broad range of cell types
- Easy optimization due to minimal cytotoxicity across a 10-fold concentration range of transfection reagent
- Simple and rapid protocol for consistent and reproducible results

Lipofectamine® RNAiMAX Transfection Reagent is a proprietary, siRNA-specific, cationic-lipid formulation that offers high transfection efficiencies on a wide variety of cell types (see table and figure) for siRNA-mediated gene knockdown experiments. Lipofectamine® RNAiMAX Transfection Reagent gives maximal knockdown and excellent cell viability across a 10-fold concentration range of the reagent, making this reagent easy to optimize for the lowest siRNA concentration while reducing cytotoxicity in your experimental system. Transfection-mediated cytotoxicity can mask the true phenotype of the target gene being studied, so minimizing the amount of reagent used in your transfections is a critical factor for successful RNAi experiments.

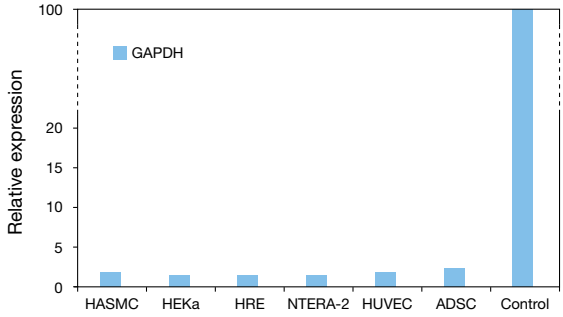
Simply mix Lipofectamine® RNAiMAX Transfection Reagent with siRNA, add your cells, and after the desired incubation time, measure your gene knockdown. Simplicity and speed combined with high transfection efficiency make Lipofectamine® RNAiMAX Transfection Reagent ideal for high-throughput siRNA transfections. Transfection conditions can be easily established for automated or robotic systems used in such applications.

For optimized protocols for transfection of a wide range of cell lines, go to www.invitrogen.com/RNAiMAX.

Product	Quantity	Cat. No.
Lipofectamine® RNAiMAX Transfection Reagent	0.75 mL	13778-075
	1.5 mL	13778-150
Lipofectamine® RNAiMAX Transfection Reagent Sample Size	0.1 mL	13778-100

Partial list of cell lines successfully transfected with Lipofectamine™ RNAiMAX Transfection Reagent.

Cell line	Cell type
293	Human kidney
A549	Human lung carcinoma
ADSC	Human adipose-derived stem cells
Cardiomyocytes	Rat heart
HAMSC	Human aortic smooth muscle cells
HCT116	Human colon carcinoma
HEKa	Human epidermal keratinocytes
HeLa	Human cervical cancer
HeLa S3	Human cervical cancer
HepG2	Human liver carcinoma
HRE	Human renal epithelial cells
HT1080	Human fibrosarcoma
HUVEC, HUAEC	Human umbilical vein endothelial cells
m-IMCD	Mouse kidney
MCF-7	Human breast/mammary cancer
MDA-MB-435	Human melanoma
MDCK	Dog kidney
ME-180	Human cervical carcinoma
MSC	Human bone marrow (mesenchymal stem cell)
N2A	Mouse neuroblastoma
Neuro 2a	Mouse neuroblastoma
NIH 3T3, 3T3-L1	Mouse fibroblast
NTERA-2	Human embryonal carcinoma
SK-N-SH	Human neuroblastoma
TIG	Human fibroblast



***Silencer*® Select siRNAs delivered by reverse transfection using Lipofectamine® RNAiMax reagent induce strong GAPDH knockdown in multiple cell types.** *Silencer*® Select siRNAs for GAPDH were transfected at 30 nM using 0.15–0.3 µL of Lipofectamine® RNAiMAX reagent per well in 96-well plates, following reverse transfection for the various cell types. More than 95% knockdown was obtained for all cell types.

Neon® Transfection System

Next generation electroporation system

- Efficiency—up to 90% in many cell types, including difficult-to-transfect cells, primary cells, and stem cells
- Flexibility—easily transfect from 1 x 10^4 cells to 5 x 10^6 cells per reaction with easy-to-use protocols
- Versatility—open system allows electroporation parameters to be optimized freely

Neon® Transfection System

Unlike standard cuvette-based electroporation chambers, the Neon® Transfection System uses a pipette tip chamber that generates a more uniform electric field and allows you to transfect cells directly in the pipette tip. This design allows better maintenance of physiological conditions, resulting in very high cell viability and transfection efficiency compared to conventional electroporation.

The transfection processes couldn't be simpler. Take up the cells and plasmid mix into the Neon® Pipette tip, plug into the pipette station, and press start. Then, just pipette the transfected cells into your culture vessel. No more filling and pulling sample from the cuvette or losing precious samples. The Neon® Transfection System uses a simple 3-step transfection procedure, and the transfection occurs in the Neon® Pipette tip.

Partial list of cell lines successfully transfected with the Neon® Transfection System.

Cell line	Cell type	Transfection efficiency (%)[1]	Viable cells (%)
MEF Primary	Embryonic fibroblast	80	75
293A	Kidney	90	90
3T3-L1	Mouse adipose	85	80
A549	Lung	75	92
Macrophages	Human (peritoneal)	60	60
MCF-7	Breast	70	80
HeLa	Cervical carcinoma	90	87
HL-60	Blood	55	70
PBMC	Blood	23	95
Primary rat cortical cells	Brain, cortical	42	98.5
Primary rat hippocampal cells	Brain, hippocampal	37	77
Raw 264.7	Blood	74	80

Product	Quantity	Cat. No.
Neon® Transfection System 100 µL Kit	50 reactions	MPK10025
	192 reactions	MPK10096
Neon® Transfection System 10 µL Kit	50 reactions	MPK1025
	192 reactions	MPK1096
Neon® Transfection System	1 each	MPK5000
Neon® Transfection System Starter Pack	1 pack	MPK5000S
Neon® Transfection System Pipette	1 each	MPP100
Neon® Transfection System Pipette Station	1 each	MPS100
Neon® Transfection Tubes	1 pack	MPT100
One-year Extended Warranty, Rapid Exchange Service	1 each	4457724
Three-year Extended Warranty, Rapid Exchange Service	1 each	4457725

Customers from the following countries or regions must go to www.invitrogen.com/instrumentregistration to place orders: Singapore, Korea, Hong Kong, Thailand, Cambodia, Vietnam, European Union countries, Turkey, Middle East countries, Canada, and Latin American countries.

Reference

1. Kim JA, Cho K, Shin MS et al. (2008) Transfection efficiency calculated from total live and dead cells. *Biosens Bioelectron* 23(9):1353–1360.

For more information and to access the complete library of fail-safe optimized protocols, go to www.invitrogen.com/neon and www.protocolexchange.com.

Lipofectamine® 2000 Transfection Reagent

Superior transfection results with DNA and siRNA

- Exceptional transfection efficiency and high expression in a broad range of cell lines and high levels of recombinant protein expression
- Proven efficiency in the presence of serum—eliminates the need to change media following transfection
- Superior performance for both plasmid and siRNA delivery [1]—most cited transfection reagent

Lipofectamine® 2000 Transfection Reagent is a proprietary cationic-lipid formulation that offers one of the highest transfection efficiencies on the widest variety of cell lines, with high protein expression levels or gene knockdown (RNAi). It is a leading transfection reagent for simple, effective, and optimal nucleic acid delivery.

Simply mix Lipofectamine® 2000 Reagent with nucleic acid, add to cell culture, and express protein or knockdown gene expression. The simplicity and speed combined with high transfection efficiency make Lipofectamine® 2000 Reagent ideal for transient protein expression or high-throughput RNAi experiments [2]. Transfection conditions can be easily established for automated or robotic systems.

References

1. Gitlin L et al. (2002) *Nature* 418:430–433.
2. Pichet JP, Ciccarone V (1999) *Focus*® 21:58.

Product	Quantity	Cat. No.
Lipofectamine® 2000 Transfection Reagent*	1.5 mL	11668019
	0.75 mL	11668027

*Bulk reagents are also available.

For protocols, citations, and additional product information about Lipofectamine® 2000 Reagent, go to the Cell Lines Database at www.invitrogen.com/transfection.

Invivofectamine® 2.0 Reagent

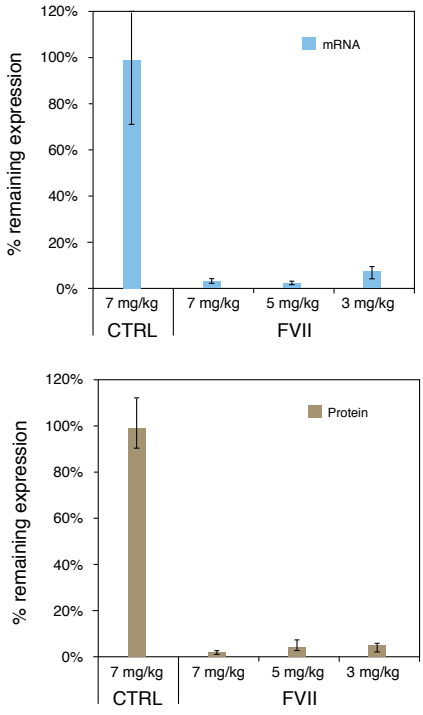
Easy-to-use, effective reagent for siRNA delivery *in vivo*

- Specific, effective knockdown (>80%) in mouse liver
- Easy-to-use procedure—just mix, equilibrate, and inject
- Nontoxic and non-immunostimulatory

Invivofectamine® 2.0 is a proprietary, lipid-based delivery reagent for *in vivo* siRNA delivery to mouse liver cells. The protocol involves an easy siRNA:lipid complex preparation (just mix, equilibrate, and inject) and a simple systemic injection, resulting in high levels of mRNA silencing (see figure) with low toxicity. A single injection of Invivofectamine® 2.0-siRNA complex results in a dose-dependent duration of knockdown at the protein level that can extend more than two weeks.

To study the biodistribution of Invivofectamine® 2.0 in mice, Alexa Fluor® 647-labeled siRNAi was injected and analyzed. Results indicated that most of the siRNA was delivered to liver cells after intravenous injection; however, we did detect some delivery to the spleen and kidney. In the kidney, cryosectioning revealed that the labeled siRNA was delivered to the glomeruli. Interestingly, a different biodistribution was observed after intraperitoneal injection. We observed delivery of labeled siRNA to spleen and pancreas cells, as well as in the liver, although to a lesser degree than with intravenous injection.

Product	Quantity	Cat. No.
Invivofectamine® 2.0, 1 mL Starter Kit	1 kit	1377-501
Invivofectamine® 2.0, 5 mL Kit	1 kit	1377-505



Use of Invivofectamine® 2.0 Reagent results in targeted knockdown in liver after a single intravenous injection. Invivofectamine® 2.0 Reagent complexed with siRNA targeting Factor VII mRNA (injected at doses of 1, 3, 5, and 7 mg/kg achieved >95% knockdown of target mRNA levels (A: knockdown was assessed by TaqMan® assays), and >95% reduction in protein activity (B: blood serum was isolated and assayed for Factor VII activity 48 hr post-injection via a chromogenic substrate).

For additional product information about Invivofectamine® 2.0 Reagent, go to www.invitrogen.com/invivofectamine.

Selection antibiotics for eukaryotic cells

Life Technologies offers high-quality selection reagents to complement its wide variety of selectable mammalian expression vectors. These antibiotics provide unique solutions for your research needs, such as dual selection and rapid, stable cell line establishment.

Geneticin® Selection Antibiotic

Reliable, trusted, and convenient selection

Geneticin® reagent is commonly used for the selection of mammalian, plant, or yeast cells. The higher purity of Geneticin® reagent means that 15–30% lower concentrations are required compared to other G418 products; therefore, surviving clonal colonies may arise faster, and cells appear healthier.

Zeocin™ Selection Antibiotic

Effective selection in multiple organisms

Zeocin™ reagent is effective in mammalian cell lines, yeast, insect cells, and bacteria. Resistance is conferred by the *Sh ble* gene. In cells expressing the protein, Zeocin™ reagent is prevented from binding and cleaving cellular DNA. The concentration required for selection ranges between 50–2,000 µg/mL (typically 300 µg/mL), depending on the cell type.

Puromycin Dihydrochloride Selection Antibiotic

Potent selection agent

Puromycin, a translational inhibitor in both prokaryotic and eukaryotic cells, is an aminonucleoside antibiotic from *Streptomyces alboniger*. Resistance is conferred by the puromycin N-acetyltransferase gene (*pac*) from *Streptomyces*. Puromycin has a fast mode of action, causing rapid cell death at low antibiotic concentrations. Adherent mammalian cells are sensitive to concentrations of 2–5 µg/mL, while cells in suspension are sensitive to concentrations as low as 0.5–2 µg/mL. Puromycin-resistant stable mammalian cell lines can be generated in less than one week.

Blasticidin S HCl Selection Antibiotic

Super-fast generation of stable cell lines

Blasticidin, a potent translational inhibitor in both prokaryotic and eukaryotic cells, is a nucleoside antibiotic from *Streptomyces griseochromogenes*. Resistance is conferred by the *bsd* gene product from *Aspergillus terreus*. *E. coli* strains are generally sensitive to concentrations of 50 µg/mL, while mammalian cells are sensitive to concentrations as low as 2–10 µg/mL. Cell death occurs rapidly, and blasticidin-resistant, stable mammalian cell lines can be generated in less than one week at low concentrations.

Hygromycin B Selection Antibiotic

Excellent for dual selection

Hygromycin B is an aminoglycosidic antibiotic that inhibits protein synthesis by disrupting translocation and promoting mistranslation of the 80S ribosome. Because it uses a different mode of action than Geneticin® or Zeocin™ reagents, it can be used in dual-selection experiments. Resistance is conferred by the *E. coli* hygromycin resistance gene (*hyg* or *hph*). The concentration for selection ranges from 100 to 1,000 µg/mL (typically 200 µg/mL) and should be optimized for each cell line.

Product	Quantity	Cat. No.
Geneticin® Selective Antibiotic, powder	1 g	11811023
	5 g	11811031
	25 g	11811098
Geneticin® Selective Antibiotic, liquid (50 mg/mL)	20 mL	10131019
	100 mL	10131027
Zeocin™ Selection Reagent, liquid (100 mg/mL)	10 mL	R25001
	50 mL	R25005
Puromycin Dihydrochloride Selection Antibiotic, liquid (10 mg/mL)	10 x 1 mL	A1113803
	20 mL	A1113802
Blasticidin S HCl, powder	50 mg	R21001
Blasticidin S HCl, liquid (10 mg/mL)	10 x 1 mL	A1113903
	20 mL	A1113902
Hygromycin B, liquid (50 mg/mL)	20 mL	10687010

Overview of protein expression systems

Recombinant protein expression technology enables analysis of gene regulation and protein structure and function. Utilization of recombinant protein expression varies widely—from investigation of function *in vivo* to large-scale production for structural studies and biotherapeutic drug discovery. Using the right expression system for your specific application is key to your success. Protein solubility, functionality, speed, and yield are often the most important factors to consider when choosing an expression system. With the wide variety of expression systems available from Life Technologies, you're sure to find one that meets your needs. The following table summarizes some of the characteristics of the most popular expression hosts. Several expression systems from Life Technologies are detailed on the following pages. For a complete list of expression systems, go to www.invitrogen.com/proteinexpression.

Expression hosts and their applications.

Host organism	Most common applications	Advantages	Challenges
Cell-free prokaryotic (pages 254–255)	•Rapid expression screening •Toxic proteins •Incorporation of unnatural labels or amino acids •Functional assays •Protein interactions	•Rapid expression directly from plasmid •Open system—easily add components to enhance solubility/functionality •Simple format •Scalable	•Large-scale expression over 3 mg
Prokaryotic (pages 252–253)	•Structural analysis •Antibody generation •Functional assays •Protein interactions	•Scalable •Low cost •Simple culture conditions	•Protein solubility •Minimal posttranslational modifications •May be difficult to express functional mammalian proteins
Yeast (page 251)	•Structural analysis •Antibody generation •Functional assays •Protein interactions	•Eukaryotic protein processing •Scalable up to fermentation (g/L) •Simple media requirements	•Fermentation required for very high yields •Growth conditions may require optimization
Insect (pages 248–250)	•Functional assays •Structural analysis •Antibody generation	•Posttranslational modifications similar to mammalian systems •Greater yield than mammalian systems	•More demanding culture conditions
Mammalian (pages 240–247)	•Functional assays •Protein interactions •Antibody generation	•Highest level of correct posttranslational modifications •Highest probability of obtaining fully functional human proteins	•Multimilligram per liter yields only possible in suspension cultures •More demanding culture conditions

Protein expression custom services

Let the experts help you express your protein at the quality and quantity you need

- Expression optimization
- Protein production and scale-up
- Protein purification

Life Technologies offers a variety of protein expression and purification services tailored to meet your individual needs. Our experienced, highly trained scientists can work with you to select the expression systems that best suit your applications and then perform the steps necessary for obtaining high-quality preparations of your protein of interest. For additional information or price quotations, contact a Custom Services Representative at www.invitrogen.com/customservices.

Overview of cloning technologies for protein expression

Life Technologies offers a variety of unique cloning technologies to greatly simplify cloning procedures and help accelerate protein expression.

TOPO® cloning technology

- Proven technology with thousands of citations
- Fast 5-minute, room-temperature reaction
- Efficient procedure yields up to 95% recombinants

TOPO® cloning provides fast and easy cloning of PCR amplified genes (with or without A-overhangs) into "off-the-shelf" TOPO® expression vectors. It requires just three easy steps: combine your PCR product and a TOPO® cloning vector, wait 5 minutes, then transform *E. coli* (Figure 1). This speed and efficiency helps save hours of time over other methods of cloning.

For an in-depth look at how TOPO® technology works, go to www.invitrogen.com/topo.

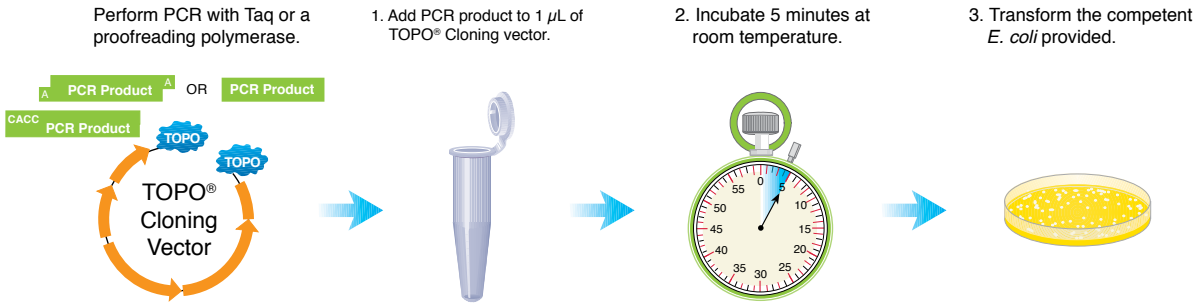


Figure 1. The TOPO® cloning protocol.

Gateway® recombination cloning technology

- Transfer DNA fragments across multiple systems and into multiple vectors
- One-hour, room-temperature reaction with >95% efficiency
- Eliminates the need for restriction enzymes and ligases

Gateway® technology provides an innovative and highly efficient method for transferring DNA fragments across multiple systems and into multiple vectors (Figure 2), replacing tedious and time-consuming cloning and subcloning steps. Orientation and reading frame are maintained with high efficiencies (typically >95%), effectively eliminating the need for secondary sequencing or subcloning. Once your DNA fragment is cloned into a Gateway® vector, you can shuttle it into as many expression systems as you like, with a simple one hour, room-temperature reaction.

For an in-depth look at how Gateway® technology works, go to www.invitrogen.com/gateway.

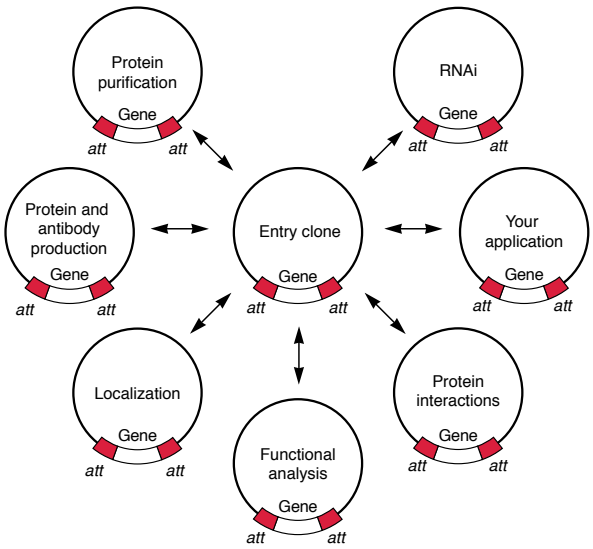


Figure 2. Rapidly move from one application to the next with Gateway® technology.

GeneArt®—Gene Synthesis and Services

Synthesize DNA constructs quickly and with 100% accuracy

Gene synthesis has become the most cost-effective, time– and resource-saving method for obtaining nearly any desired DNA construct (Figure 1), outperforming conventional molecular biology techniques in many aspects from time and economization to expression performance, stability, and quality.

GeneArt® gene synthesis tools go beyond traditional synthesis and enable you to:

- Improve protein expression with proprietary GeneOptimizer® technology
- Gain access to hard-to-clone constructs; long, complex DNAs; and customized vectors
- Create unlimited numbers of mutants for screening experiments
- Overcome RNAi inactivation
- Engineer proteins to improve enzymes, and humanize and/or increase binding affinities of antibodies

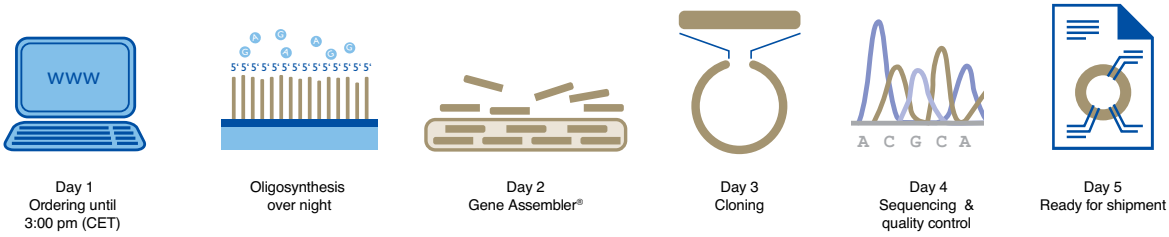


Figure 1. GeneArt® Gene Synthesis and Services can provide you with your gene of interest in 5 days.

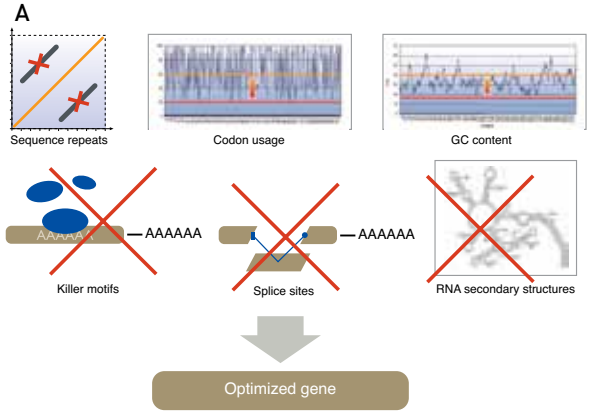
GeneArt® GeneOptimizer® Technology

Gene optimization to maximize protein expression

Our proprietary GeneOptimizer® algorithm delivers proven increases in protein yield through sequence optimization (Figure 2). By evaluating many important expression parameters in parallel, GeneOptimizer® technology generates up to 500,000 variants of your target sequence in an evolutionary approach and selects the best match for your specific requirements.

Select GeneOptimizer® technology when you need:

- Removal of introns
- Knockout of cryptic splice sites and RNA-destabilizing sequence elements
- Increased RNA stability
- Adaptation of codon usage
- Extensive mutagenesis
- Flexible combination of functional domains
- Introduction of restriction sites
- Epitope shuffling
- Consideration of immunomodulatory CpG motifs



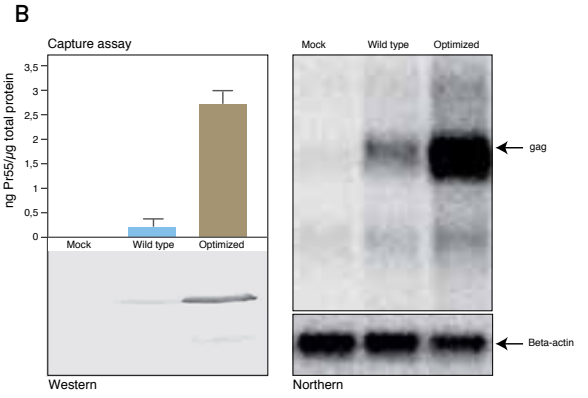


Figure 2. The GeneOptimizer® algorithm increases protein yields (B) by optimizing DNA sequences (A).

To learn more about gene synthesis and place your order, go to www.invitrogen.com/genesynthesis.

GeneArt® Seamless Cloning and Assembly System

Quickly assemble up to four DNA fragments in one vector

- Speed—clone up to four DNA fragments simultaneously in a single tube at room temperature typically in 30 minutes
- Flexibility—use our linear vector or any vector of your choice
- Precision and efficiency—no extra sequences; just clone what you want, where you want it
- Simplicity—online DNA Oligo Designer designs oligos and assembles DNA molecules *in silico*

The GeneArt® Seamless Cloning and Assembly System enables cloning of up to four DNA fragments simultaneously into virtually any linearized vectortypically in 30 minutes, without extra DNA sequences, restriction endonucleases, or ligation. The GeneArt® Seamless Cloning and Assembly System uses a proprietary enzyme mix to recognize and precisely assemble DNA fragments sharing a 15-bp end homology that is created by PCR amplification using primers designed to generate an overlap between the adjacent DNA fragments to be assembled. The online DNA Oligo Designer (www.invitrogen.com/designdnaassembly) guides users through experimental design, including oligo design and ordering.

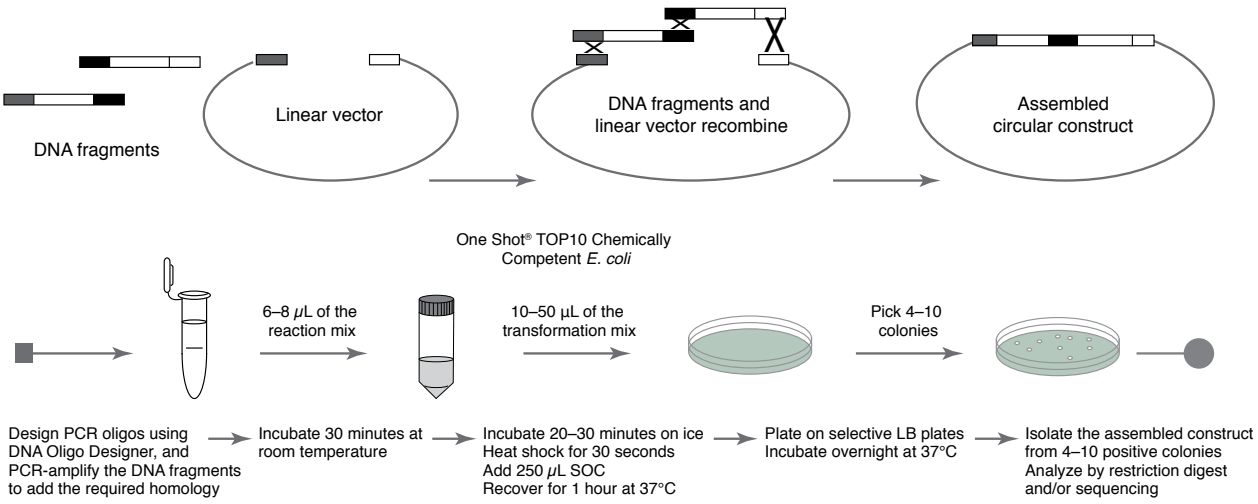


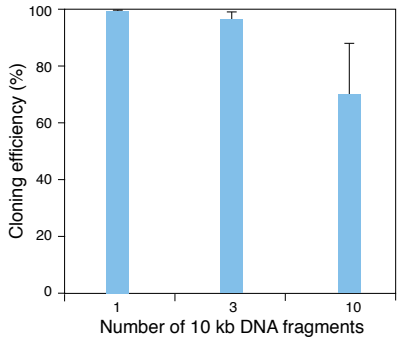
Figure 1. GeneArt® Seamless Cloning and Assembly workflow.

GeneArt® High-Order Genetic Assembly System

Easily assemble ten or more DNA fragments in a single vector

- Speed—clone ten or more DNA fragments simultaneously in a single vector (up to 110 kb)
- Flexibility—use our linear vector or a vector of your choice; oligonucleotide stitching feature allows end-editing and reuse of preexisting DNA fragments without the need for re-amplification
- Precision and efficiency—no extra sequences; just clone what you want, where you want it
- Simplicity—online DNA Oligo Designer designs oligos and assembles DNA molecules *in silico*

The GeneArt® High-Order Genetic Assembly System is a highly efficient, vector-independent system for the simultaneous and seamless assembly of up to 10 preexisting or synthetic DNA fragments *in vivo* up to 110 kb total construct size (see figure). The GeneArt® High-Order Genetic Assembly System relies on yeast's ability to take up and recombine DNA fragments with high efficiency via transformation associated recombination, greatly reducing *in vitro* handling of DNA and eliminating the need for restriction digestion and ligation. The online DNA Oligo Designer (www.invitrogen.com/designdnaassembly) guides users through experimental design, including oligo design and ordering.



Cloning efficiency of the GeneArt® High-Order Genetic Assembly System with increasing numbers of 10-kb PCR fragments assembled. Even as 100-kb PCR fragments are assembled, the cloning efficiency remains over 65%, proving the system is a reliable solution for assembly of complex DNA molecules.

To learn more about GeneArt® cloning kits, go to www.invitrogen.com/dnaassembly.

Selecting a mammalian expression system

There are many options in selecting a mammalian expression system matched to your specific needs. Ask yourself the following three questions, then use the table below to find the best system for your needs.

1. What delivery method will work best for you?

You can introduce your gene of interest into mammalian cells using viral transduction with recombinant viruses or transfection of plasmid DNA. The choice of delivery method depends on what mammalian cell type you are using and the experimental effects desired. Transfection works well for a wide variety of cell types, while viral transduction is the ideal choice for cell types that are difficult to transfect or for nondividing cell types.

- For viral transduction, see page 245
- If you selected transfection, continue to the next question

2. Will you be making stable cell lines?

Mammalian expression experiments are performed either transiently or with stable cell lines that express the protein of interest.

- Transient expression, which typically results in high levels of expression for a few days, is ideal for rapid protein production and quick data generation
- Stable expression requires generating cell lines in which your expression construct is integrated into the host genome for a stable cell line that can be used over a long experimental time course or used over many experiments. Cells that have integrated the construct are selected by the addition of a selection agent to the media (pages 241–245). If you want to make cell lines, we recommend the Jump-In™ System. For random stable cell lines, any pcDNA™ vector or the ViraPower™ Lentiviral Expression System may be used.

3. Do you need inducible or constitutive expression?

- If you are working with a nontoxic gene and the timing of expression is not important, choose a constitutively expressing promoter
- An inducible promoter allows you to control the timing of gene expression. In the absence of an inducer, your gene is not expressed. Add inducer to turn on expression. This option is ideal for expressing toxic proteins. Choose from the T-REx™ Inducible Expression System or the Flp-In™ T-REx™ System. For more information on inducible promoters, go to www.invitrogen.com/proteinexpression.

Selected mammalian expression systems from Life Technologies.

Application	System	Description
Constitutive CMV expression	pcDNA™ Vectors	Constitutive CMV expression with choices of epitope tags and selection markers
Viral expression	ViraPower™ Lentiviral and Adenoviral Expression Systems	High-level gene expression in any dividing or nondividing mammalian cell type Choice of stable gene expression (lentiviral) or transient gene expression (adenoviral)
Generating homogeneous stable cell lines	Jump-In™ Fast Gateway® System	Rapid generation of homogeneous stable cell lines
	Jump-In™ TI™-Gateway® System	Generation of stable, isogenic cell lines
Inducible expression	T-REx™ System	Regulated expression from CMV promoter
	Flp-In™ T-REx™ System	Rapid generation of regulated stable expression cell lines
	ViraPower™ Lentiviral T-REx™ System	Regulated expression in any mammalian cell type

For information on systems not discussed in this guide, go to www.invitrogen.com/proteinexpression.

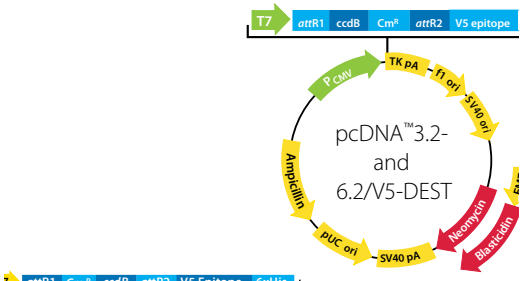
pcDNA™ Gateway® vectors

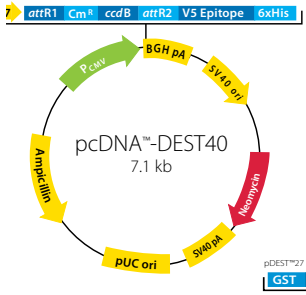
High-level expression from the CMV promoter

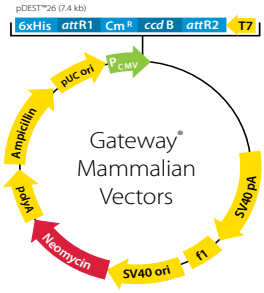
- Rapid cloning
- Most widely used vectors
- High-level, constitutive expression

Life Technologies offers the largest collection of mammalian expression vectors for efficient expression, selection, and analysis of recombinant proteins. The pcDNA™ Gateway® destination vectors are designed for rapid cloning with a Gateway® entry clone for high-level, constitutive expression in a variety of mammalian cells. These destination vectors employ a cytomegalovirus (CMV) enhancer-promoter for high-level expression and contain *attR* sites for efficient recombination from any *attL* Gateway® entry vector.

Additional pcDNA™ vectors are available that offer either the CMV or human elongation factor-1α (EF-1α) promoter for high-level expression, a variety of different epitope tags, standardized detection or purification across a range of proteins, and several selection markers available for creating stable cell lines.







For more information and complete list of Gateway® products, go to www.invitrogen.com/gateway. For more information and assistance for selecting the best vector for your experiments, go to the Vector Selection Tool at www.invitrogen.com/vectors.

Mammalian pcDNA™ expression vectors with the CMV promoter.

Promoter	Selection marker			Fusion tag				Cloning	Vector	Cat. No.
	Neo	Bsd	Zeo	V5	6xHis	GST	Xpress™			
CMV	●			●				GW*/D-TOPO®**	pcDNA™ 3.2/GW/D-TOPO®	K244020
		●		●				GW/D-TOPO®	pcDNA™ 6.2/GW/D-TOPO®	K246020
	●			●				Gateway®	pcDNA™ 3.2/V5-DEST	12489019
		●		●				Gateway®	pcDNA™ 6.2/V5-DEST	12489027
	●			●				Gateway®	pcDNA™ 3.1/nV5-DEST	12290010
	●			●	●			Gateway®	pcDNA™-DEST40	12274015
	●				●			Gateway®	pDEST™26	11809019
	●					●		Gateway®	pDEST™27	11812013
		●		●				Gateway®	pcDNA™ 6/BioEase™-DEST	K98001
	●			●	●			D-TOPO®	pcDNA™ 3.1D/V5-His-TOPO®	K490001 K490040
	●			●	●			TOPO® TA	pcDNA™ 3.3 TOPO®	K830001
	●			●	●			TOPO® TA	pcDNA™ 3.1/V5-His-TOPO®	K480001 K480040
			●		●		●	TOPO® TA	pcDNA™ 4/HisMax-TOPO®	K86420
EF-1α		●		●	●			TOPO® TA	pEF6/V5-His-TOPO®	K961020

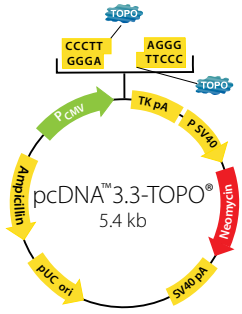
All destination vectors are provided lyophilized and supercoiled. * GW denotes Gateway® recombination cloning. ** D-TOPO® denotes Directional TOPO® Cloning.

pcDNA™ 3.3-TOPO® TA Cloning® Kit

A powerful mammalian expression vector for high protein yields

- Two- to five-fold higher protein yields
- Five–minute TOPO® TA ligation and >85% cloning efficiency
- Express native (or tagged) proteins without extraneous amino acids

The pcDNA™ 3.3-TOPO® vector contains a modified, enhanced CMV promoter that enables extremely high protein expression. The vector is ideal for large-scale protein production, especially when used in combination with the FreeStyle™ MAX CHO or 293 Expression Systems. Multiple proteins have been expressed at 8–30 mg/L in the FreeStyle™ systems (Figure 1). This vector is also well suited for superior transient or stable protein expression. Luciferase and ß-galactosidase expression were measured in adherent cultured mammalian cells following transient transfection with pcDNA™ 3.3-TOPO® constructs. Luciferase activity was approximately 5-fold higher than that from the pCI vector (Promega) and the pCMV-Script® vector (Agilent), and ß-galactosidase activity was approximately 2-fold higher (Figure 2).



Product	Quantity	Cat. No.
pcDNA™ 3.3-TOPO® TA Cloning® Kit*	20 reactions	K830001

* The pcDNA™ 3.3-TOPO® TA Cloning® Kit is supplied with the TOPO®-adapted plasmid vector, all reagents for cloning, and One Shot® TOP10 competent cells.

For more information and a complete list of TOPO® products, go to www.invitrogen.com/topo. For more information and assistance selecting the best vector for your experiments, go to the Vector Selection Tool at www.invitrogen.com/vectors.

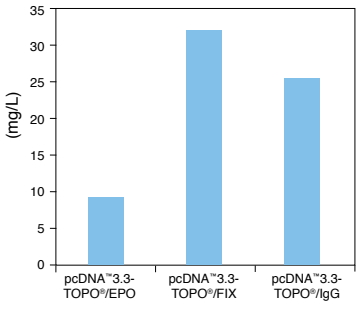


Figure 1. Using the pcDNA™ 3.3-TOPO® vector to express multimilligram levels of erythropoietin (EPO), Factor IX (FIX), and IgG in FreeStyle™ CHO-S cells. pcDNA™ 3.3-TOPO® constructs containing the relevant PCR-derived gene sequences were transfected into FreeStyle™ CHO-S cells in 30-mL volumes. Heavy and light chain genes were co-transfected for IgG expression. Four to six days posttransfection, EPO and FIX were quantified by ELISA, and IgG was quantified using immobilized goat anti-human IgG antibody. Each bar shows the average of two expression runs.

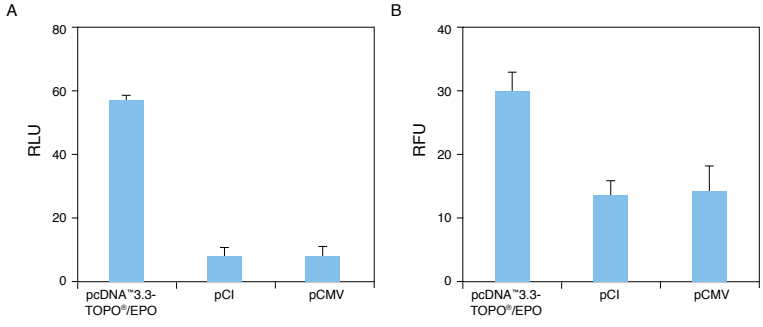


Figure 2. The pcDNA™ 3.3-TOPO® vector outperforms competitors' vectors for protein expression in adherent cells. Open reading frames encoding luciferase (A) and ß-galactosidase (B) were TOPO® cloned into the pcDNA™ 3.3-TOPO® vector. The same sequences were also cloned into pCI and pCMV-Script® by standard restriction enzyme digestion and ligation. Plasmids were transiently transfected into adherent GripTite™ 293 MSR cells. Luciferase activity is shown as relative luminescence units (RLU), and ß-galactosidase activity is reported as relative fluorescence units (RFU). Bars show the standard deviation (n = 6).

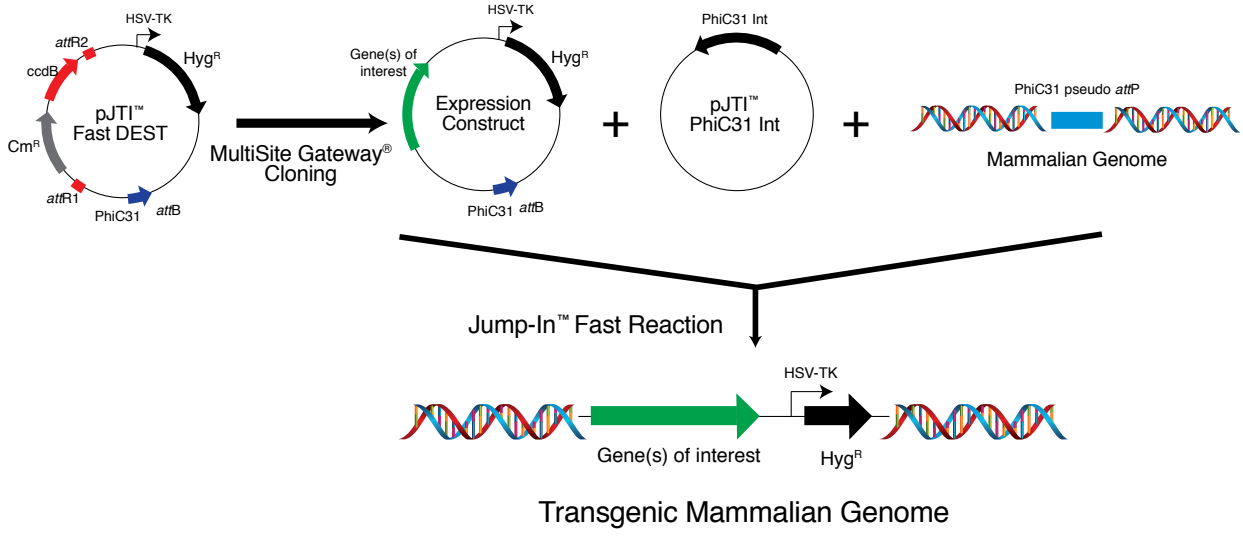
Jump-In™ Targeted Integration Technology

Rapid and efficient generation of stable cell lines

- Minimizes clonal variation
- Produces homogeneous expression levels
- Unidirectional and virtually irreversible integration

The Jump-In™ Targeted Integration (TI) technology uses PhiC31 integrase-mediated recombination to stably integrate your choice of DNA sequence at specific genomic locations, called pseudo-*attP* sites, in mammalian cells. Unlike the better-known recombinases, such as Cre and Flp, PhiC31 integrase catalyzes recombination between two non-identical sites and combined with the lack of a corresponding excisionase enzyme, makes the integration events unidirectional and virtually irreversible. In the Jump-In™ Fast Gateway® System, the integrated DNA sequences include your genetic elements of interest (such as promoter–reporter pairs) from the targeting expression construct that is generated using the pJTI™ Fast DEST vector and the MultiSite Gateway® Pro Plus Vector Module. Since the pJTI™ Fast DEST vector also encodes the Hygromycin resistance gene, transformants containing stably integrated sequences are selected using Hygromycin B and expanded for downstream applications. The figure below schematically depicts the workflow for generating your well-expressing mammalian cell line using the Jump-In™ Fast Gateway® targeted integration technology.

In addition to the Jump-In™ Fast Gateway® System, which enables the rapid creation of well-expressing mammalian cell lines, Life Technologies also offers the Jump-In™ TI™ Gateway® Targeted Integration System, which facilitates the generation of isogenic, stable cell lines expressing your genetic element(s) of interest, and allows you to eliminate chromosomal positioning effects from your experiments.



The simple workflow for using the Jump-In™ Fast Gateway® targeted integration technology.

Product	Quantity	Cat. No.
Jump-In™ Fast Gateway® System	20 reactions	A10893
Jump-In™ Fast Gateway® Core Kit	1 kit	A10894
Jump-In™ TI™ Gateway® System	20 reactions	A10895
Jump-In™ TI™ Gateway® Vector Kit	1 kit	A10896
Jump-In™ TI™ Platform Kit	1 kit	A10897

FreeStyle™ MAX systems

Rapid, high-yield protein production from transiently transfected suspension cultures

- Optimized protocols for suspension-adapted CHO and HEK293 cells
- Functional proteins with mammalian posttranslational modifications
- Scalable process to generate milligram to gram quantities of protein typically in two to seven days

The FreeStyle™ MAX System is a breakthrough protein production technology for large-scale, rapid generation of post-translationally modified functional proteins (Table 1), reliably producing up to hundreds of milligrams of recombinant protein from a liter of cultured cells. Its exceptional productivity and speed is achieved by starting with adapted CHO and 293 cells grown in suspension culture, and then transiently transfecting these cultures with the high-efficiency FreeStyle™ MAX Transfection Reagent. This system is an alternative to conventional stable cell line generation or the use of transient transfection of adherent cultures in multiple dishes, flasks, or roller bottles (Figure 1).

The FreeStyle™ 293 System for 293 cells

The FreeStyle™ 293 System for easily scalable, rapid protein production generates functional, posttranslationally modified, proteins in human HEK 293 cells. The system offers exceptional productivity and speed in the transient expression of recombinant antibody proteins (Table 2). Transfection is performed using the high-efficiency 293fectin™ Transfection Reagent. The transfection protocol can be readily scaled with minimal or no modifications for suspension culture volumes from <30 mL to >10 L while maintaining equivalent volumetric protein yields.

Convenience and speed

Large-scale, transient transfection of mammalian cells in suspension culture at the 5-mL to 25-L scale can be easily set up in most laboratories with minimal effort. This is a reliable and rapid method to generate milligram quantities of mammalian recombinant proteins for functional, biotherapeutic drug discovery, and structural studies. The FreeStyle™ MAX System achieves large-scale protein production in days, and is easily scalable from 5-mL to 25-L cultures. For detailed protocols for scale-up, go to www.invitrogen.com/transfection. This scale-up is also available as a Custom Service (go to www.invitrogen.com/customservices).

Product	Quantity	Cat. No.
FreeStyle™ MAX CHO Expression System	1 kit	K900020
FreeStyle™ MAX 293 Expression System	1 kit	K900010
FreeStyle™ MAX Transfection Reagent	1 mL	16447100
	15 mL	16447500
Gibco® FreeStyle™ 293 Expression Medium	1,000 mL	12338018
	6 x 1 L	12338026
293fectin™ Transfection Reagent	1 mL	12347019
	15 mL	12347500

FreeStyle™ components are available individually or in kit form. Bulk quantities of all individual components are also available. TheFreeStyle™ 293 System kit includes 293fectin™ Transfection Reagent, Gibco® FreeStyle™293 Expression Medium, FreeStyle™ 293 cells, and Opti-MEM® I medium.

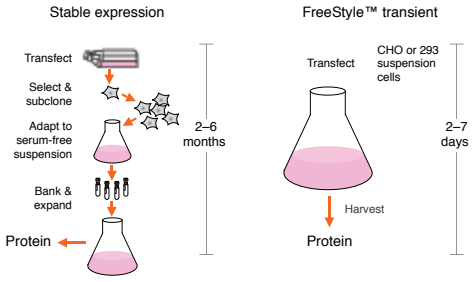


Figure 1. Transient transfection in suspension compared to stable transfection.

Table 1. High protein yield in FreeStyle™ MAX CHO suspension cells.*

Protein	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6
Human IgG	6	18	28	36	42	47
Factor IX	0.7	1.6	2.2	2.0	2.3	2.2
HGH	50	109	178	214	225	243
EPO	2.3	6.5	7.4	8.5	8.8	8.9

* Human IgG, Factor IX, human growth hormone (HGH), and erythropoietin (EPO) were produced in FreeStyle™ CHO-S cells. Cells were cultured to a density of 1 x 10^6 cells/mL in 30 mL of FreeStyle™ CHO Expression Medium in a 125-mL shaker flask. The transfection complexes were formed using 1.25 µg of plasmid DNA and 1.25 µL of FreeStyle™ MAX Reagent per mL of culture volume to be transfected, in OptiPRO™ SFM.

Table 2. High protein yield in FreeStyle™ 293 suspension cells using 293fectin™ reagent.

Protein	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6
Human Ig G	12	63	99	128	159	164
Factor IX	0.3	0.7	0.9	1.0	1.0	1.1
HGH	19	150	67	143	163	181
EPO	0.03	0.05	0.08	0.1	0.1	0.1

ViraPower™ lentiviral expression systems

Reproducible delivery and stable gene expression in any mammalian cell type

- Efficient viral delivery to both dividing and nondividing cells (e.g., stem cells)
- Ideal for analysis of long-term gene expression—even in terminally differentiated or growth-arrested cells
- High-level gene protein production

Stable gene expression is only a few steps away with the ViraPower™ Lentiviral Expression Kits (see figure). Just transfect, harvest, and titer to produce sufficient viral supernatant for performing many transduction experiments. Use the supernatant immediately or store it for up to one year.

The ViraPower™ HiPerform™ Lentiviral Kits achieve elevated protein expression even in nondividing cells such as stem cells and primary neuronal cells. These lentiviral kits offer accurate determination of functional lentivirus titers in just 2 days (FastTiter™ Kit); 4-fold increase in protein expression via WPRE (woodchuck posttranscriptional regulatory enhancer) and cPPT (polypurine tract) elements; and flexible, efficient cloning using Gateway® or TOPO® technology.

How the ViraPower™ Lentiviral System works. Your packaged gene is immediately imported into the nucleus and stably integrated into the host genome. Transient expression can be detected within the first 24 hr. In addition, you can use Blasticidin or Zeocin™ selection agents to create stable cell lines.

Product	Quantity	Cat. No.
ViraPower™ HiPerform™ Lentiviral TOPO® Expression Kit	1 kit	K531000
ViraPower™ HiPerform™ Lentiviral FastTiter™ TOPO® Expression Kit	1 kit	K532000
ViraPower™ HiPerform™ Lentiviral Gateway® Expression Kit	1 kit	K533000
ViraPower™ HiPerform™ Lentiviral FastTiter™ Kit Gateway® Expression Kit	1 kit	K534000
ViraPower™ Lentiviral Gateway® Expression Kit	1 kit	K496000
ViraPower™ Zeo Lentiviral Gateway® Expression Kit	1 kit	K498000
ViraPower™ UbC Lentiviral Gateway® Expression Kit	1 kit	K499000
ViraPower™ Lentiviral Directional TOPO® Expression Kit	1 kit	K495000
ViraPower™ Lentiviral Packaging Mix	60 reactions	K497500

The ViraPower™ lentiviral expression kits include a pLenti7.3/V5-DEST™, pLenti6.3/V5-DEST™, pLenti6/V5-DEST™, pLenti4/V5-DEST™, or pLenti6/UbC/V5-DEST™ Gateway® Vector Kit or a pLenti6/V5 Directional TOPO® Cloning Kit; the ViraPower™ Bsd or Zeo Lentiviral Support Kit; One Shot® Stbl3 Competent Cells; and the 293FT Cell Line.

MembranePro™ Functional Protein Expression Kit

Produce functional, soluble membrane proteins in mammalian cells

- Provides a concentrated population of cell surface receptors in a native cellular context
- Less labor-intensive than traditional production of cell membrane fractions—no ultracentrifugation steps, since proteins bud off in nonviral lipoparticles that are easily collected from the culture medium
- Offers a replacement for cell membrane preparations used in receptor–ligand binding assays

For researchers studying G-protein–coupled receptors (GPCRs) and other membrane proteins, the MembranePro™ Functional Protein Expression (FPE) System helps efficiently and reliably deliver enriched, functional membrane proteins that are amenable to various downstream assays used in research, antibody characterization, drug discovery, and high-throughput screening.

Lipoparticles are harvested and precipitated from culture medium 48 hours after transfection. Resuspended lipoparticles can be stored at –80°C or used directly in biochemical assays. Lipoparticles produced using the MembranePro™ FPE System have been studied using radioligand saturation and competition binding assays for multiple GPCR targets. Our performance data demonstrate that MembranePro™ particles can offer a higher receptor density and exhibit pharmacological equivalence when compared to cell membrane preparations.

Product	Quantity	Cat. No.
MembranePro™ Functional Protein Expression Kit	10 reactions	A11667
MembranePro™ Functional Protein Support Kit	10 reactions	A11668
	60 reactions	A11669
	600 reactions	A11670

Expression kit contains pEF6 TOPO® vector, MembranePro™ Reagent, 293FT cells, Lipofectamine® 2000 Transfection Reagent, MembranePro™ Precipitation Mix. Support kit contains MembranePro™ Reagent, Lipofectamine® 2000 Transfection Reagent, MembranePro™ Precipitation Mix.

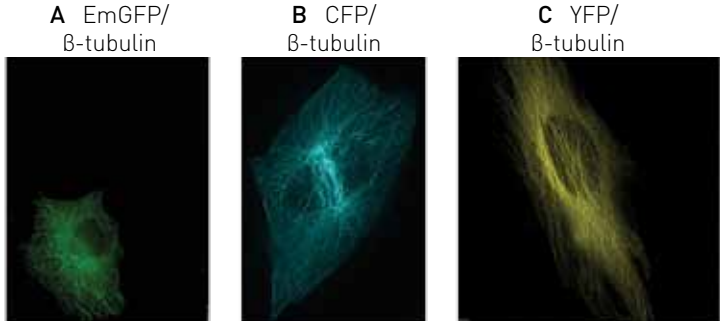
To learn more, go to www.invitrogen.com/membranepro.

Vivid Colors™ fluorescent protein vectors

The best and brightest *Aequorea victoria* fluorescent protein vectors

- Multiple colors for diverse applications
- Faster cloning using powerful and versatile Gateway® and TOPO® vectors from Life Technologies
- Most widely used autofluorescent proteins

The Vivid Colors™ fluorescent protein vectors feature the original *Aequorea victoria*–derived fluorescent proteins (FP) for simple, noninvasive detection of recombinant proteins. These FPs have been modified with critical mutations, resulting in extreme fluorescence intensity and optimal mammalian expression (Tsien RY (1998) *Annu Rev Biochem* 67:509).



CHO and HeLa cells transfected with Vivid Colors™ expression plasmids. The live cells were imaged 48 hr posttransfection. (**A**) Expression in CHO cells of EmGFP fused to ß-tubulin. (**B**) Expression in CHO cells of CFP fused to ß-tubulin. (**C**) Expression in HeLa cells of YFP fused to ß-tubulin.

Fluorescent proteins available from Life Technologies.

Protein	Color	Excitation (nm)	Emission (nm)	Mutations	Recommended filter set		
					Semrock	Omega	Chroma
EmGFP	Green	487	509	S65T, S72A, N149K, M153T, I167T	GFP-3035B	XF100	41020
YFP	Yellow	514	527	S65G, S72A, K79R, T203Y	YFP-2427A	XF1042	41028
CFP	Cyan	452	505	K26R, Y66W, N146I, M153T, V163A, N164H	CFP-2432A	XF114	31044
BFP	Blue	308–383	440–447	F64L, Y66H, Y145F, V163A , N198S	DAPI-1160A	XF10	31021
Cycle 3 GFP	Green	395, 478	507	F64L, F100S, M154T, V164A	GFP-3035B	XF76	41095

Product	Quantity	Cat. No.
Vivid Colors™ pcDNA™ 6.2 Vectors		
Vivid Colors™ pcDNA™ 6.2/C-EmGFP-DEST	6 µg	V35520
Vivid Colors™ pcDNA™ 6.2/N-EmGFP-DEST	6 µg	V35620
Vivid Colors™ pcDNA™ 6.2/C-YFP-DEST	6 µg	V35720
Vivid Colors™ pcDNA™ 6.2/N-YFP-DEST	6 µg	V35820
Vivid Colors™ pcDNA™ 6.2/EmGFP-Bsd/V5-DEST	6 µg	V36620
Vivid Colors™ pcDNA™ 6.2/C-EmGFP-GW/TOPO® Mammalian Expression Vector Kit	1 kit	K35920
Vivid Colors™ pcDNA™ 6.2/N-EmGFP-GW/TOPO® Mammalian Expression Vector Kit	1 kit	K36020
Vivid Colors™ pcDNA™ 6.2/C-YFP-GW/TOPO® Mammalian Expression Vector Kit	1 kit	K36120
Bacterial expression		
pRSET/CFP	10 µg	V35220
pRSET/BFP	10 µg	V35420
Lentiviral expression		
pLenti6.2-GW/EmGFP Expression Control Vector	20 µg	V36920

For a complete listing of FP-based expression vectors and antibodies for detection of GFP or a tagged gene of interest, go to www.invitrogen.com/vividcolors. For more information and assistance selecting the best vector for your experiments, go to the Vector Selection Tool at www.invitrogen.com/vectors.

Overview of insect expression systems

Expression in insect cells offers significant advantages, including high expression levels, ease of scale-up, production of proteins with proper posttranslational modifications, and simplified cell growth. Insect cells do not require CO_2 for growth and can be readily adapted to high-density suspension culture for large-scale expression. Many of the posttranslational modification pathways present in mammalian systems are also utilized in insect cells, allowing the production of recombinant protein that is antigenically, immunogenically, and functionally similar to the native mammalian protein. Baculovirus expression systems are powerful and versatile systems for high-level, recombinant protein expression in insect cells. Expression levels up to 500 mg/L have been reported using the baculovirus expression system.

- The BaculoDirect™ Baculovirus Expression System is a fast and easy method for generating recombinant baculovirus. The BaculoDirect™ Linear DNA includes *attR* sites for rapid and efficient recombinational cloning with a Gateway® entry clone. The resulting recombinant DNA is taken directly from the LR reaction mix and used to transfect insect cells, saving a significant amount of time. Purified virus can be isolated within one week. The reduction of hands-on time makes the BaculoDirect™ system ideal for high-throughput expression (page 249).
- The Bac-to-Bac® Baculovirus Expression System uses a unique bacmid shuttle vector that recombines by site-specific transposition and generates an expression bacmid in bacterial cells. The expression bacmid is then transfected into insect cells to generate recombinant baculovirus (page 250).
- The Bac-N-Blue™ Baculovirus Expression System has been used for over a decade to produce high levels of recombinant proteins.
- The *Drosophila* Expression System (DES®) uses the well-characterized *Drosophila* Schneider S2 cells and simple expression vectors to allow stable or transient expression of recombinant proteins.

Insect expression systems from Life Technologies.

			Fusion partner					
System	Host	Secretion signal	Position	Purif.	Epitope	Promoter	Expression/ inducer	Advantage
BaculoDirect™	Sf9, Sf21, or High Five™		N-term C-term	6xHis 6xHis	V5 V5	Polyhedrin	Infection	Fast and easy method for generation of recombinant baculovirus; ideal for high throughput
Bac-to-Bac® or Bac-to-Bac® HBM	Sf9, Sf21, or High Five™	Honeybee melittin	GST N-term	6xHis	pFastBacHT pDEST10	Polyhedrin or p10	Infection production	Rapid baculovirus production; easy blue/white selection of recombinant colonies
Bac-N-Blue™	Sf9, Sf21, or High Five™	Honeybee melittin	C-term	6xHis	Xpress™ V5	Polyhedrin	Infection	Classic and trusted expression system for high-level recombinant protein production
DES®	S2 cells	BIP	C-term	6xHis	V5	MT or Ac5	$CuSO_4$ or constitutive	Easy-to-use stable system, constitutive or inducible expression; uses simple plasmids for expression; extremely high integration of transfected plasmids eliminates need to select and screen for expression from clonal cell lines

For more information on insect expression systems, go to www.invitrogen.com/proteinexpression. For more information and assistance selecting the best vector for your experiments, go to the Vector Selection Tool at www.invitrogen.com/vectors.

BaculoDirect™ Baculovirus Expression System

Fast and easy method for generation of recombinant baculovirus

- Strong polyhedrin promoter for high-level expression
- Rapid cloning with Gateway® technology
- Simple, established protocol for high-throughput expression

The BaculoDirect™ Baculovirus Expression Kits use Gateway® technology to enhance the speed of baculovirus generation and is an easy method for producing recombinant baculovirus; purified baculovirus can be isolated typically in less than one week. Whether your application is benchtop feasibility, high-throughput expression, or scale-up, the rapid and efficient BaculoDirect™ Baculovirus Expression System is well-suited for your needs.

The BaculoDirect™ Linear DNA (the baculovirus genome) is engineered to include *attR* sites for quick and efficient recombination with a Gateway® entry clone and subsequent expression in Sf9 or Sf21 insect cells. The gene of interest is recombined from the entry clone into the BaculoDirect™ Linear DNA using a simple, 1-hour LR reaction. The resulting reaction mix contains the recombinant baculovirus carrying the gene of interest and is used directly to transfect insect cells. The need for transforming bacteria and isolating a large bacmid or cotransfection of a transfer vector and linear baculovirus DNA into insect cells is eliminated. As a result, the hands-on time is greatly reduced. Purified baculovirus can be isolated typically in less than one week.

Product	Quantity	Cat. No.
BaculoDirect™ N-Term Expression Kit	5 transfections	12562054
BaculoDirect™ N-Term Transfection Kit	5 transfections	12562062
BaculoDirect™ C-Term Expression Kit	5 transfections	12562013
BaculoDirect™ C-Term Transfection Kit	5 transfections	12562039

The BaculoDirect™ Expression Kits include BaculoDirect™ Linear DNA, Cellfectin® transfection reagent, Sf9 cells, Gateway® LR Clonase™ enzyme mix, Gibco® Unsupplemented Grace's Insect Medium, and ganciclovir. The BaculoDirect™ Transfection Kits include BaculoDirect™ Linear DNA, Cellfectin® transfection reagent, and ganciclovir.

Bac-to-Bac® Baculovirus Expression System

Generation of recombinant bacmid in *E. coli*

- Time-saving expression bacmid
- Easy colony screening
- Rapid protein purification

The Bac-to-Bac® Baculovirus Expression System relies on generation of recombinant baculovirus by site-specific transposition in *E. coli* rather than homologous recombination in insect cells to produce recombinant baculovirus. The expression cassette of the pFastBac™ vectors recombines with the parent bacmid in DH10Bac™ *E. coli* to form an expression bacmid. The parent bacmid contains the *lacZ*α complementation factor for efficient blue/white screening of positive recombinants. The bacmid is then transfected into insect cells for production of recombinant baculovirus particles. The pFastBac™ vectors offer the strong polyhedrin promoter for protein expression and a large multiple cloning site for simplified cloning. The Bac-to-Bac® HBM TOPO® Secreted Expression System enables secreted protein expression via the honeybee melittin (HBM) secretion signal, which is ideal for toxic proteins and glycoproteins that require a secretion signal to be glycosylated.

Product	Quantity	Cat. No.
Bac-to-Bac® Baculovirus Expression System	1 kit	10359016
Bac-to-Bac® Vector Kit	1 kit	10360014
Bac-to-Bac® HT Vector Kit	1 kit	10584027
Max Efficiency® DH10Bac™ Competent Cells (1 x 10^8 transformant/µg)	5 x 100 µL	10361012
Bac-to-Bac® HBM TOPO® Cloning Kit	20 reactions	A11338
Bac-to-Bac® HBM TOPO® Secreted Expression System	20 reactions	A11339
Bac-to-Bac® N-His TOPO® Cloning Kit	20 reactions	A11099
Bac-to-Bac® C-His TOPO® Cloning Kit	20 reactions	A11098
Bac-to-Bac® C-His TOPO® Expression System	20 reactions	A11100
Bac-to-Bac® N-His TOPO® Expression System	20 reactions	A11101

Each Bac-to-Bac® Baculovirus Expression System includes pFastBac™1 vector, MAX Efficiency® DH10Bac™ Chemically Competent Cells, pFastBac™ 1-Gus control vector, and Cellfectin® Reagent. The Bac-to-Bac® Vector Kit contains only the expression and control vectors.
Bac-to-Bac® TOPO® cloning kits include a vector kit for 20 TOPO® cloning reactions (pFastBac TOPO® Vector containing the TEV cleavage site and His-Tag, a control expression vector, 10x PCR, dNTP Mix, salt solution, sterile water, control PCR template, polyhedrin forward primer, SV40 pA reverse primer) and a kit containing competent cells (20 reactions, One Shot® Mach1-T1^R Chemically Competent *E. coli*). Bac-to-Bac® TOPO® expression systems include the vector kit and competent cells (described above) as well as 4 kits (5 x 0.1 mL each of MAX Efficiency® DH10Bac™ Competent *E. coli* for 20 reactions and 1 mL Cellfectin® II Reagent.

PichiaPink™ yeast expression system

Generate recombinant proteins in yeast

- Screen *Pichia* transformants rapidly by adenine auxotrophy and color selection
- Reduce protein degradation with protease-deficient *Pichia pastoris* strains
- Optimize protein secretion with one of eight secretion signal sequences

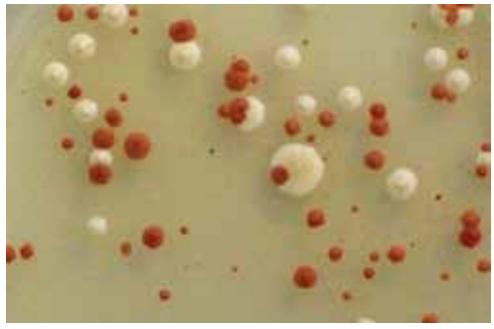
The PichiaPink™ system is a eukaryotic protein expression system based on the eukaryote *Pichia pastoris*, which can be used for high-level (g/L) and large-scale (>1,000-L) production of secreted recombinant proteins. This new system contains both low- and high-copy plasmid backbones, eight secretion signal sequences, and four yeast strains to help you optimize for the highest possible yield of recombinant protein.

P. pastoris has many of the eukaryotics subcellular machinery for post-translation modification and delivers a high probability of an active function recombinant protein. Additional advantages of *P. pastoris* include rapid growth, a well-defined genetic background, simple media formulations, and simple handling techniques—all of which make it the ideal host for producing large quantities of protein in a short period of time.

The PichiaPink™ system provides you with two different kits for secreted expression of your recombinant protein of interest:

- The PichiaPink™ Secretion Optimization Kit enables you to screen multiple signal sequences with your gene of interest in both low- and high-copy vectors (pPink-LC and pPink-HC, respectively) for optimal expression and secretion of your recombinant protein. pPink-LC and pPink-HC vectors are also available separately as the PichiaPink™ Vector Kit.
- The PichiaPink™ Secreted Protein Kit allows you to clone your gene of interest in frame with the *Saccharomyces cerevisiae* α-mating factor presequence using the pPinkα-HC plasmid for secreted expression of your recombinant protein. pPinkα-HC is also available separately as the PichiaPink™ Secreted Protein Vector Kit.

Product	Quantity	Cat. No.
PichiaPink™ Secretion Optimization Kit	1 kit	A11150
PichiaPink™ Secreted Protein Kit	1 kit	A11151
PichiaPink™ Vector Kit	1 kit	A11152
PichiaPink™ Secreted Vector Kit	1 kit	A11153
PichiaPink™ Expression Strain Set	4 x 1 mL	A11154
PichiaPink™ Secretion Signal Set	8 each	A11155
PichiaPink™ Media Kit	1 kit	A11156



Use color selection to find your transformed PichiaPink™ colonies. Transformed *Pichia* appear as white colonies while untransformed *Pichia* appear as red-pink colonies.

PichiaPink™ Secretion Optimization Kit
• The PichiaPink™ Vector kit containing a high– and low–copy number plasmid
• The PichiaPink™ Secretion Signal set containing eight secretion signals
• The PichiaPink™ Expression Strain set containing four glycerol strains
• The PichiaPink™ Media Kit containing four basic media
PichiaPink™ Secreted Protein Expression Kit
• The PichiaPink™ Secreted Protein Vector Kit containing the high-copy number plasmid with the *S. cerevisiae* α-mating factor presequence to yield secreted protein expression
• The PichiaPink™ Expression Strain Set containing four glycerol strains
• The PichiaPink™ Media Kit containing four basic media

For more information on PichiaPink™ expression systems, go to www.invitrogen.com/proteinexpression. For more information and assistance selecting the best vector for your experiments, go to the Vector Selection Tool at www.invitrogen.com/ vectors.

Overview of prokaryotic expression systems

Due to their ease of use, relatively high yields, and simple scalability, T7-regulated *E. coli* expression systems are often chosen for generating recombinant protein. Selecting the right system for your specific application is the key to your success. With the wide variety of T7 expression systems available from Life Technologies, you're sure to find one that meets your needs. The following table summarizes some of the most popular T7 expression systems. For a complete list, go to www.invitrogen.com/proteinexpression.

The most popular T7 expression systems from Life Technologies.

System	Promoter	Advantage
Champion™ pET302/NT-His and pET303/CT-His Vector Kit	T7lac	Higher expression levels than those obtained from other pET vector suppliers
Champion™ pET300/NT-DEST and pET301/CT-DEST Gateway® Vector Kit	T7lac	Convenience of Gateway® cloning and choice of N-term or C-term His6 tag
Champion™ pET Expression System	T7lac	Highest level of protein production
Champion™ pET SUMO Expression System	T7lac	Highest level expression of proteins with enhanced solubility
Champion™ pET Expression System	T7lac	Rapid, visual screening of protein expression without western blotting with Lumio™ technology
Champion™ pET BioEase™ Expression System	T7lac	Easy, efficient expression of biotinylated proteins
T7 Expression System	T7	High-level expression of nontoxic proteins
pBAD Expression System	araBAD	Tightly regulated expression for efficient expression of difficult-to-express proteins
trc Expression System	trc	Enhanced expression of eukaryotic protein

Champion™ pET Expression System

High-level expression with simplified, efficient cloning in *E. coli*

- Protein of interest constitutes >50% of total cellular protein
- 5-minute TOPO® Cloning reaction with >95% efficiency
- No ligase, post-PCR procedures, or restriction enzymes required

The Champion™ pET Expression System uses optimized vectors and a BL21 Star™ *E. coli* expression strain to produce protein yields up to ten-fold greater than any other expression system. The system takes advantage of the high activity and specificity of the bacteriophage T7 RNA polymerase for high-level transcription of the gene of interest. The *lac* operator located in the promoter provides tighter regulation than traditional T7-based vectors, improving plasmid stability and cell viability. BL21 Star™ *E. coli* are genetically engineered to reduce transcript degradation, resulting in significantly improved mRNA stability and increased protein production.

Innovative vector design

Cloning into a Champion™ pET Expression vector is easy, fast, and efficient. Available with Directional TOPO® or Gateway® Cloning Technologies, you can generate your clone typically in one day. In addition, the Champion™ pET vectors offer simplified protein purification, protein detection, enhanced solubility, and native protein expression.

Champion™ pET Directional TOPO® Expression Vector features.

Vector	Position	Tag	Cleavage protease	Antibiotic resistance	Benefit
pET100/D-TOPO®	N-term	6xHis-Xpress™	EK	Amp	Cleavable detection and purification tag
pET101/D-TOPO®*	C-term	V5-6xHis	–	Amp	Detection and purification tag
pET102/D-TOPO®	N-term	Thioredoxin	EK	Amp	Cleavable thioredoxin tag enhances protein translation and solubility
	C-term	V5-6xHis	–		Detection and purification tag
pET151/D-TOPO®	N-term	V5-6xHis	TEV	Amp	Cleavable detection and purification tag
pET160/GW/D-TOPO®	N-term	Lumio™-6xHis	TEV	Amp	Fluorescent detection and purification tag; cleavable
pET161/GW/D-TOPO®	C-term	Lumio™-6xHis	–	Amp	Fluorescent detection and purification tag
pET200/D-TOPO®	N-term	6xHis-Xpress™	EK	Kan	Cleavable detection and purification tag
Champion™ pET300/NT-DEST and pET301/CT-DEST Gateway® Vector Kit	N-term	6xHis	–	Amp	Detection and purification tag
	C-term				
Champion™ pET302/NT-His and pET303/CT-His Vector Kit	N-term	6xHis	–	Amp	Detection and purification tag
	C-term				

*Useful for native protein expression by inserting a stop codon before the C-terminal fusion sequence.

Product	Quantity	Cat. No.
Champion™ pET302/NT-His and pET303/CT-His Vector Kit	10 µg of each vector	K630203
Champion™ pET300/NT-DEST and pET301/CT-DEST Gateway® Vector Kit	6 µg of each vector	K630001
Champion™ pET200 Directional TOPO® Expression Kit	20 reactions	K20001
Champion™ pET161 Directional TOPO® Expression Kit w/Lumio™ Technology	20 reactions	K16101
Champion™ pET160 Directional TOPO® Expression Kit w/Lumio™ Technology	20 reactions	K16001
Champion™ pET151 Directional TOPO® Expression Kit	20 reactions	K15101
Champion™ pET102 Directional TOPO® Expression Kit	20 reactions	K10201
Champion™ pET101 Directional TOPO® Expression Kit	20 reactions	K10101
Champion™ pET100 Directional TOPO® Expression Kit	20 reactions	K10001

Each Champion™ pET Directional TOPO® Expression Kit contains a pET-TOPO® vector, an expression control, chemically competent *E. coli* (where appropriate), and primers for sequencing.

For a complete list of Champion™ pET Directional TOPO® Expression Kits, go to www.invitrogen.com/topo. For information and assistance selecting the best vector for your experiments go to the Vector Selection Tool at www.invitrogen.com/vectors.

Expressway™ Mini and Maxi Cell-Free Expression Systems

Cell-free synthesis of active protein typically in a few hours

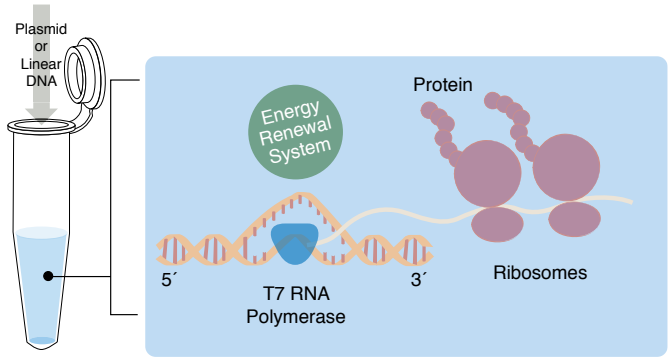
- More full-length functional protein
- Single-tube format without any specialized instrumentation
- Scalable and easily adapted for high-throughput expression

The Expressway™ Cell-Free Expression Systems use an efficient coupled transcription and translation reaction to produce high yields of full-length, functional protein from *E. coli* cell lysates. These systems eliminate the time-consuming steps of cell-based protein production including transformation, cell culture, and expression optimization. In as little as 2 hours, you can produce protein suitable for an array of downstream applications and functional studies.

The Expressway™ Mini Cell-Free Expression System offers flexibility in experimental design, allowing you to synthesize proteins from both circular plasmids and linear templates (e.g., PCR products), perform high-throughput synthesis of proteins, and express genes toxic to *in vivo* systems.

The Expressway™ Maxi Cell-Free *E. coli* Expression System uses an efficient, coupled transcription/translation reaction to produce milligram quantities of soluble, functionally active protein in 4–6 hours. The procedure can be performed in a single reaction tube and is easily scalable without the need for specialized equipment. The TOPO® TA Cloning® expression vectors (5-minute cloning reaction with 95% efficiency) are provided for optimal expression results.

The Expressway™ NMR Cell-Free *E. coli* Expression System produces milligram quantities of efficiently stable isotope–labeled recombinant proteins for NMR spectroscopy and mass spectrometry.



How the Expressway™ system works.

Product	Quantity	Cat. No.
Expressway™ Mini Cell-Free Expression System	1 kit	K990100
Expressway™ Maxi Cell-Free *E. coli* Expression System	1 kit	K990097
with pEXP5-NT/TOPO® and pEXP5-CT/TOPO®	1 kit	K990096
Expressway™ NMR Cell-Free *E. coli* Expression System	1 kit	K990099
with pEXP5-NT/TOPO® and pEXP5-CT/TOPO®	1 kit	K990098

The Expressway™ Cell-Free Expression Systems include the *E. coli*, reaction buffer, feed buffer, amino acid mix, methionine, DNase/RNase-free water, RNase A, T7 Enzyme Mix, 2-mL reaction tubes, and a positive expression control vector.

For more information about these Expressway™ Systems and complementary TOPO® and Gateway® vectors, go to www.invitrogen.com/expressway. For more information and assistance selecting the best vector for your experiments, go to the Vector Selection Tool at www.invitrogen.com/vectors.

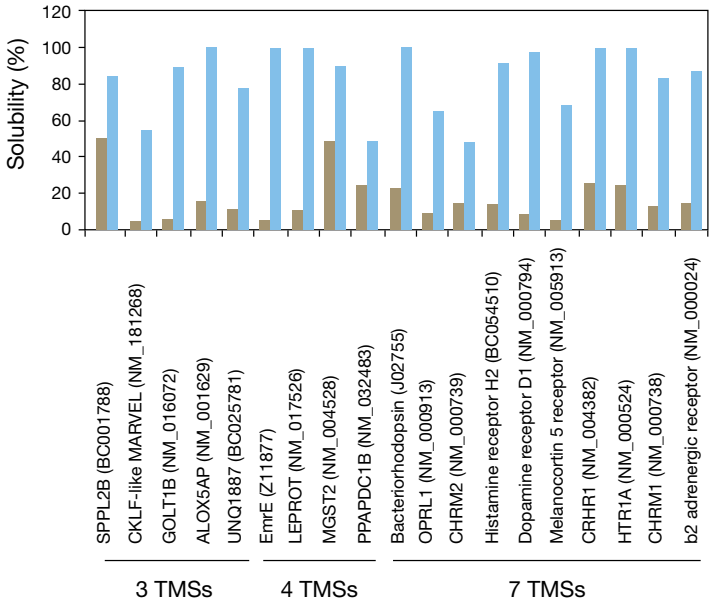
MembraneMax™ Protein Expression Kits

Optimized, cell-free expression for maximum yield, solubility, and purity

- High yields of soluble membrane proteins
- Scalable cell-free expression format
- Convenient purification scheme

Based on the MembraneMax™ reagent—a planar phospholipid membrane bilayer surrounded by a scaffold protein (also called a nanolipoprotein particle, or NLP)—the MembraneMax™ Protein Expression Kit allows you to produce high yields of soluble (dispersed), tagged membrane proteins. The MembraneMax™ *HN* Protein Expression Kit also incorporates His-tagged MembraneMax™ reagent for purification of native membrane proteins. MembraneMax™ Protein Expression Kits are simple to use—just supply your gene of interest and in one day, you can express and purify your membrane protein.

Both kits deliver scalable, cell-free expression of microgram to milligram quantities of your membrane protein, and are amenable to high-throughput protein synthesis for screening applications, as well as the expression of toxic proteins. Downstream analyses of membrane proteins synthesized with the MembraneMax™ kits include antibody production, crystallography, immunoprecipitation, ligand binding or other functional assays, mass spectrometry, nuclear magnetic resonance, and protein array construction.



Soluble membrane protein expression achieved using the MembraneMax™ *HN* kit. The *in vitro* expression and solubility of membrane proteins of different topologies, sizes, origins, and proposed roles were analyzed. Proteins were expressed in the presence (blue) or absence (tan) of MembraneMax™ *HN* reagent. For the analyzed data set, the overall solubility increased from 17.3 ± 2.2% (in the absence of NLPs) to 78.8 ± 3.4% (in the presence of NLPs). GPCRs exhibited a remarkable increase in solubility in the presence of NLPs. TMS = transmembrane segments.

Product	Quantity	Cat. No.
MembraneMax™ Protein Expression Kit	20 reactions	A10632
	100 reactions	A10633
MembraneMax™ *HN* Protein Expression Kit	20 reactions	A10634
	100 reactions	A10635

To learn more, go to www.invitrogen.com/membranemax.

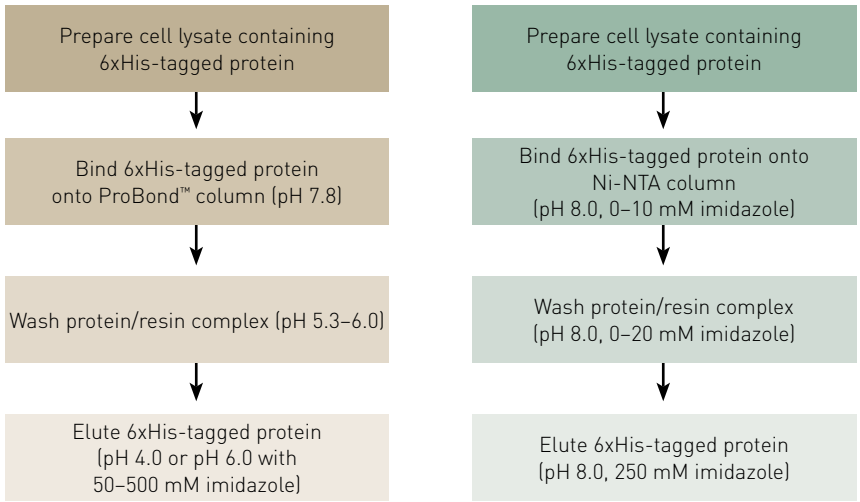
Nickel-charged affinity resins

Efficient FPLC, batch, and gravity-flow purification of 6xHis-tagged proteins

- Purify recombinant proteins with 6xHis sequence
- One-step purification
- Compatible with native and denaturing conditions

Two nickel-charged resins are available for purifying recombinant proteins containing a polyhistidine sequence. ProBond™ Nickel-Chelating Resin uses chelating ligand iminodiacetic acid (IDA) coupled to a resin that is suitable for use in FPLC, batch, and gravity-flow applications. Ni-NTA Agarose uses chelating ligand nitrilotriacetic acid (NTA) coupled to a resin that is suitable for batch and gravity-flow applications. Both resins are offered individually or as part of complete kits for convenient purification of recombinant proteins under native and denaturing conditions.

Product	Quantity	Cat. No.
ProBond™ Nickel-Chelating Resin	50 mL	R801-01
	150 mL	R801-15
ProBond™ Purification System	6 purifications	K850-01
Ni-NTA Agarose	10 mL	R901-01
	25 mL	R901-15
	100 mL	R901-10
Ni-NTA Purification System	6 purifications	K950-01
Polypropylene Columns (empty)	50 columns	R640-50



ProBond™ purification procedure | Ni-NTA purification procedure

Overview of 6xHis-tagged protein purification procedures.

Dynabeads® Magnetic Beads for protein isolation and purification

Magnetic separation of peptides, proteins, and protein complexes

- Reproducible results when isolating peptides, proteins, and protein complexes
- Extremely gentle sample handling for your target
- Simple and ideal for use in immunoprecipitation, serum profiling, and antibody purification—no columns required

Magnetic bead chromatography for fractionation and serum profiling

In profiling, peptides are captured from serum samples (or other biological fluids) by magnetic bead chromatography, then analyzed by MALDI mass spectrometry. This can now be automated for high-throughput using the new Dynal® Peptide Profiler. Once installed, you can accurately screen 96 samples per day on Tecan® robot platforms with Dynabeads® RPC-18, helping you save months of time-consuming, one-a-day sample handling with conventional techniques.

Table 1. Choose Dynabeads® Magnetic Beads for your serum or biomarker profiling application.

Application	Product	Technology	Format	Quantity	Cat. No.
Fractionation, serum profiling, and sample clean-up	Dynabeads® RPC 18	Protein/peptide adsorption and release	12.5 mg/mL	2 mL	10211D
				20 mL	10212D
	Dynabeads® RPC Protein		12.5 mg/mL	2 mL	10216D
				20 mL	10217D
	Dynabeads® WCX		12.5 mg/mL	2 mL	10511D
				20 mL	10512D
	Dynabeads® SCX		12.5 mg/m	2 mL	10513D
				20 mL	10514D
	Dynabeads® SAX		12.5 mg/mL	2 mL	10515D
				20 mL	10516D
Automated peptide profiling for biomarker discovery	Dynal® Peptide Profiler	Magnets and protocol algorithm for Tecan® platform, installed and ready to use; for use with Dynabeads® RPC-18	CD with algorithm, 2x magnet installation		12040D

Protein isolation and immunoprecipitation

Immunoprecipitation (IP) uses an antigen-specific antibody to precipitate the target antigen from solution. Magnetic beads can be used to enrich and purify a given protein or to "pull-down" a whole protein complex by co-immunoprecipitation.

Table 2. Choose Dynabeads® Magnetic Beads for your immunoprecipitation or protein isolation application.

Application	Product	Technology	Format	Quantity	Cat. No.
Immunoprecipitation and organelle isolation	Dynabeads® Protein A	Protein/peptide capture through primary antibodies	Isolate 250 µg hIgG/mg beads (30 mg/mL)	1 mL	10001D
				5 mL	10002D
	Dynabeads® Protein G		Isolate 250 µg hIgG/mg beads (30 mg/mL)	1 mL	10003D
				5 mL	10004D
				2 mL	11201D
	Dynabeads® M-280 Sheep Anti-Mouse IgG	Protein/peptide capture through mouse or rabbit specific antibody	10 mg/mL	10 mL	11202D
				2 mL	11203D
	Dynabeads® M-280 Sheep Anti-Rabbit IgG		10 mg/mL	10 mL	11204D
				2 mL	14203
	Dynabeads® M-280 Tosyl-Activated	Protein/peptide capture through primary antibody conjugated via surface active tosyl groups	30 mg/mL	10 mL	14204
				2 mL	11205D
Immunoassays, purification of DNA/RNA binding proteins, biopanning, and phage display	Dynabeads® M-280 Streptavidin	Protein/peptide capture through any biotinylated ligand	10 mg/mL	10 mL	11206D
				100 mL	60210

To find out more or to see the whole range of Dynabeads® and DynaMag™ magnets for protein analysis, go to www.invitrogen.com/dynal.

Qubit® 2.0 Quantitation Platform

Sophisticated quantitation of proteins, DNA, and RNA

- Accurate, fluorescence-based quantitation of proteins, DNA, and RNA
- High sensitivity allowing you to detect as little as 1 µL of sample
- Fast, simple add-and-read protocol
- Touch screen operation with USB port for easy data management

The Qubit® 2.0 Quantitation Platform pairs the Qubit® 2.0 Fluorometer with the Qubit® assays for the most accurate, sensitive protein quantitation. The Qubit® assays represent the most advanced quantitation systems for DNA, RNA, or protein samples. State-of-the-art Molecular Probes® assay reagents deliver high sensitivity and specificity together with a simple, streamlined protocol. All Qubit® 2.0 kits contain prepared buffers and prediluted standards—just dilute the dye in the provided buffer, add your sample, and read!

Qubit® 2.0 Fluorometer.

Product	Quantity	Cat. No.
Qubit® 2.0 Fluorometer	1 instrument	Q32866
Qubit® 2.0 Quantitation Platform Starter Kit	1 kit	Q32871
Qubit® 2.0 Quantitation Platform Lab Starter Kit	1 kit	Q32872
Qubit® Protein Assay Kit	100 assays	Q33211
	500 assays	Q33212
Qubit® assay tubes	500 tubes	Q32856

The Qubit® 2.0 Quantitation Platform Starter Kit includes 1 Qubit® 2.0 Fluorometer, 1 Qubit® dsDNA HS Assay Kit (100 assays), 1 Qubit® dsDNA BR Assay Kit (100 assays), 1 Qubit® RNA Assay Kit (100 assays), 1 Qubit® Protein Assay Kit (100 assays), and 500 Qubit® assay tubes. The Qubit® 2.0 Quantitation Platform Lab Starter Kit includes everything in the Qubit® 2.0 Quantitation Platform Starter Kit plus an additional four Qubit® 2.0 fluorometers (for a total of five). The Qubit® 2.0 Fluorometer is ideal for quantitating DNA samples for cloning and transfection (see pages 118–119), as well as protein samples (see page 259 for Qubit® Protein Assay Kits).

For more details or a demonstration, go to www.invitrogen.com/qubit.

Protein quantitation kits

Superior sensitivity, ease of use, high tolerance of contaminants, high throughput—all the features you need for fast protein quantitation in your laboratory.

Choose from the following protein quantitation kits.

Assay	No. of assays	Abs/Em (nm) *	Sensitivity and range	Speed	Throughput	Compatible with	Cat. No.
Qubit® Protein Assay Kit	100 500 1,000	485/590	250 ng–5 µg	5 min	High	• Reducing agents • Amines	Q33211 Q33212 Q33210
EZQ® Protein Assay	2,000	450/620	20 ng–5 µg	1 hr	Medium	• Reducing agents • Detergents • Ampholytes • Chaotropes • Amines	R33200
NanoOrange® Protein Assay	200–2,000	485/590	20 ng–2 µg	30 min	Medium	• Reducing agents • Amines	N6666
CBQCA Protein Assay	300–800	450/550	10 ng–150 µg	1 hr	High	• Reducing agents • Amines	C6667

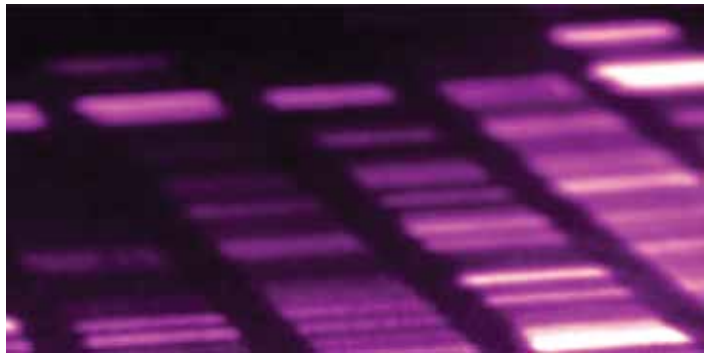
For more details and other quantitation kits, go to www.invitrogen.com/proteomics and click on "Protein Quantitation".

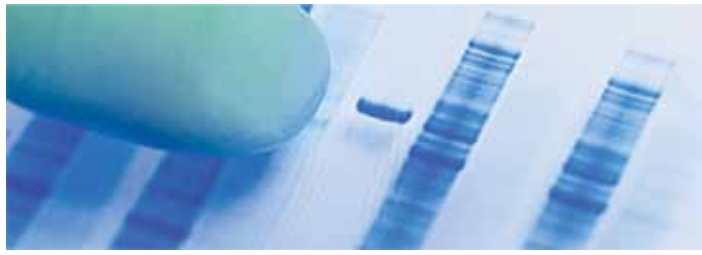
Novex® protein analysis solutions

Designed for accurate, reproducible results

- Superior protein resolution
- Ultrasensitive protein detection
- Accelerated protocols for extremely fast results

Today the Novex® brand includes more than just gels. It also covers high-quality products and reagents designed to work with the original gel product line, such as sharp, accurate prestained and unstained molecular weight standards; uniquely formulated, sensitive stains and detection reagents; the 7-minute iBlot® Dry Blotting System, and BenchPro® 4100 automated western processing system. Using these products together enables complete protein separation with tight band resolution, accurate molecular weight estimations, sensitive detection, and complete transfer. In short, using the Novex® protein analysis products helps ensure that you'll get the right answer the first time, every time.





Novex® technology

For even faster protein analysis results—complete electrophoresis, blotting, and western detection, typically in 1 hour

- Advanced technologies allow an accelerated process
- Excellent separation and resolution, typically in only 25 minutes (Figure 2)
- Complete western analyses in as little as 35 minutes—combine iBlot® Dry Blotting System 7-minute protein transfers (page 272) with iBlot® Western Detection Kits for detection (page 275)

Figure 1. The Novex® accelerated method provides a complete workflow solution from gels to blotting to detection, typically in 1 hour.

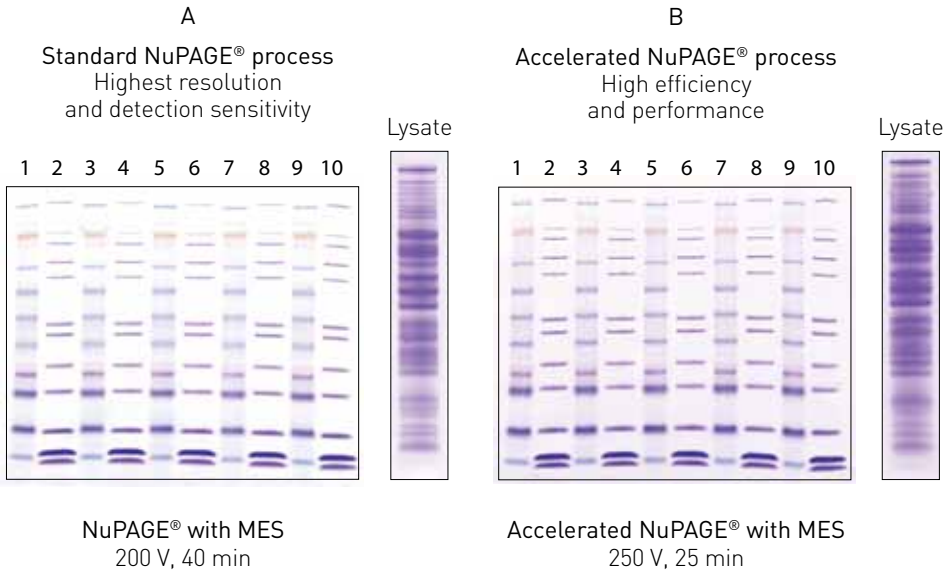


Figure 2. Novex® NuPAGE® 4–12% Bis-Tris Gels loaded with samples and run with MES SDS running buffer using (A) the standard protocol at 200 V for 40 min, or (B) the accelerated protocol at 250 V for 25 min. The gels were stained with Coomassie® R-250. Lanes 1, 3, 5, 7, 9: 5 µL SeeBlue® Plus2 Pre-stained Standard; Lanes 2, 4, 6, 8, 10: 5 µL Mark12™ Unstained Standard; Lysate: 10 µg *E.coli* lysate.

For more details, go to www.invitrogen.com/novex.

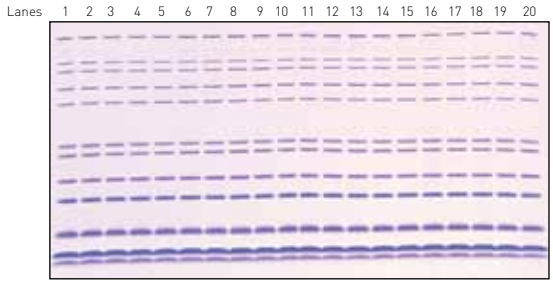
Novex® NuPAGE® gel system

High-performance, high-quality precast protein gels

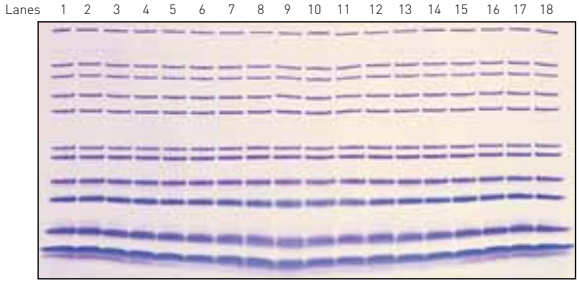
- Superior protein band resolution and stability
- Faster sample run times (typically 35 min or less)
- Longer product shelf life (up to 12 months)

The Novex® NuPAGE® Precast Gel System is a revolutionary, high-performance polyacrylamide gel system. Gels are available in both Bis-Tris and Tris-acetate formulations and in a variety of acrylamide percentages and sizes (mini: 8 x 8 cm; midi: 8 x 13 cm), so you're sure to find one that meets your electrophoresis needs. Regardless of the gel chemistry and percentage, the unique Novex® NuPAGE® gel formulations minimize the "smiles" and poor resolution of other gel types (see figure). The neutral-pH gel chemistry and buffer system avoid chemical sample modifications that occur in alkaline environments. You're enabled to get reliable separation results every time. In addition, proteins transfer from Novex® NuPAGE® gels more efficiently, leading to more sample on your membrane for higher detection signals during western analysis.

A Novex® NuPAGE® 4–12% Bis-Tris Midi Gel



B Bio-Rad Criterion™ XT 4–12% Bis-Tris Gel



Proteins resolve into straight, sharp bands on a Novex® NuPAGE® Midi Gel (A) compared to the wavy bands seen on a competitor's gel (B). Each lane contains 5 µL of Mark12™ Unstained Standard.

Cat. No. for Novex® NuPAGE® Mini Gels. Each box of Novex® NuPAGE® Mini Gels includes 10 gels.

Gel type and percentage*	1.0-mm thickness				1.5-mm thickness (for larger sample volumes)		
	10 wells	12 wells	15 wells	2D well	10 wells	15 wells	2D well
Bis-Tris 10%	NP0301BOX	NP0302BOX	NP0303BOX	NP0306BOX	NP0315BOX	NP0316BOX	NP0317BOX
Bis-Tris 4–12%	NP0321BOX	NP0322BOX	NP0323BOX	NP0326BOX	NP0335BOX	NP0336BOX	NP0337BOX
Bis-Tris 12%	NP0341BOX	NP0342BOX	NP0343BOX	NP0346BOX			
Tris-acetate 7%	EA0355BOX	EA03552BOX	EA03555BOX	EA0358BOX	EA03585BOX		
Tris-acetate 3–8%	EA0375BOX	EA03755BOX	EA0376BOX	EA0378BOX	EA03785BOX		

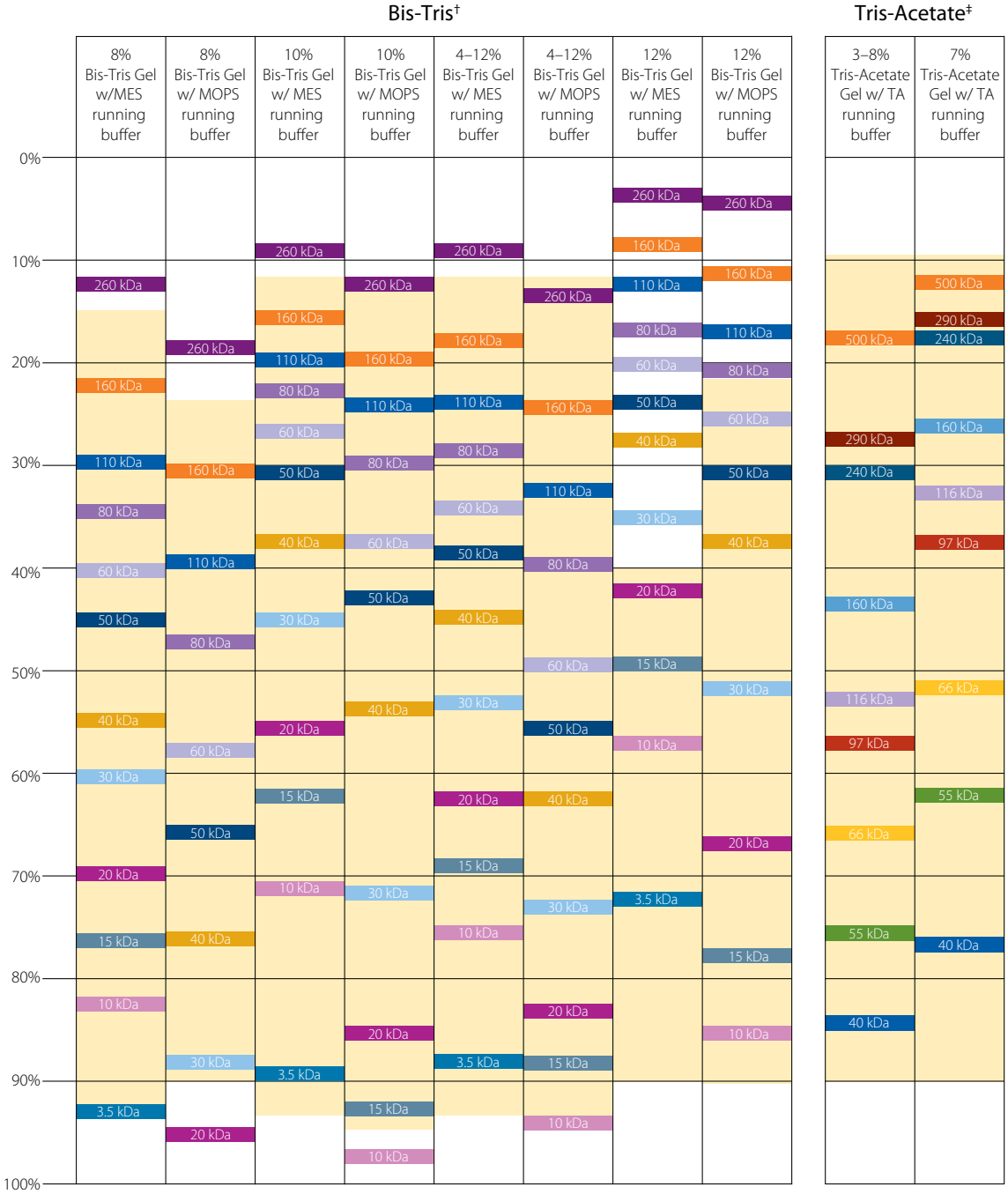
Cat. No. for Novex® NuPAGE® Midi Gels. Each box of Novex® NuPAGE® Midi Gels includes 10 gels.

Gel type and percentage*	Without Midi Gel Adapters			With Midi Gel Adapters (10)†		
	12 + 2 wells‡	20 wells	26 wells‡	12 + 2 wells‡	20 wells	26 wells‡
Bis-Tris 8%	WG1001BOX	WG1002BOX	WG1003BOX	WG1001A	WG1002A	WG1003A
Bis-Tris 10%	WG1201BOX	WG1202BOX	WG1203BOX	WG1201A	WG1202A	WG1203A
Bis-Tris 4–12%	WG1401BOX	WG1402BOX	WG1403BOX	WG1401A	WG1402A	WG1403A
Tris-acetate 3–8%	WG1601BOX	WG1602BOX	WG1603BOX	WG1601A	WG1602A	WG1603A

* For a more extensive list of gel percentages, well counts (including standard multichannel pipettor–compatible), and gel sizes, go to www.invitrogen.com/novex1d. † By attaching the Midi Gel Adapter (Cat. No. WA0999) to the gel cassette, you can run these midi gels in Bio-Rad's Criterion™ Cell. ‡ Compatible with standard multichannel pipettor. 12 + 2 well gels have 12 regular wells and 2 smaller wells for protein standards.

Choose the right Novex® NuPAGE® gel for your application

Use the migration pattern depicted in the table below to determine which Novex® NuPAGE® gel will provide the pattern most appropriate for your sample. For more selections, go to www.invitrogen.com/novex1d.



† Migration patterns of Novex® Sharp Protein Standards (Cat. No. LC5800, Pre-stained; Cat. No. LC5801, Unstained) on Novex® NuPAGE® Bis-Tris Gels.
‡ Migration patterns of HiMark™ Unstained Standard (Cat. No. LC5688) on Novex® NuPAGE® Tris-Acetate Gels.

Tris-glycine/Tricine to Novex® NuPAGE® Gel conversion chart.	
Currently using:	Recommended NuPAGE® Gel
4% Tris-glycine	3–8% NuPAGE® Tris-Acetate (+ TA Buffer)*
6% Tris-glycine	3–8% NuPAGE® Tris-Acetate (+ TA Buffer)
8% Tris-glycine	7% NuPAGE® Tris-Acetate (+ TA Buffer)
10% Tris-glycine	10% NuPAGE® Bis-Tris (+ MOPS Buffer)
12% Tris-glycine	10% NuPAGE® Bis-Tris (+ MOPS Buffer)
14% Tris-glycine	12% NuPAGE® Bis-Tris (+ MOPS Buffer)
16% Tris-glycine	12% NuPAGE® Bis-Tris (+ MES Buffer)
18% Tris-glycine	12% NuPAGE® Bis-Tris (+ MES Buffer)
4–12% Tris-glycine	3–8% NuPAGE® Tris Acetate (+ TA Buffer) or 4–12% NuPAGE® Bis-Tris (+ MOPS Buffer)
4–20% Tris-glycine	4–12% NuPAGE® Bis-Tris (+ MES Buffer)
8–16% Tris-glycine	4–12% NuPAGE® Bis-Tris (+ MOPS Buffer)
10–20% Tris-glycine	12% NuPAGE® Bis-Tris (+ MOPS Buffer)
10% Tricine	10% NuPAGE® Bis-Tris (+ MES Buffer)
16% Tricine	4–12% NuPAGE® Bis-Tris (+ MES Buffer) or 12% NuPAGE® Bis-Tris (+ MES Buffer)
10–20% Tricine	4–12% NuPAGE® Bis-Tris (+ MES Buffer)

* Resolution on a 3–8% Novex® NuPAGE® Tris-Acetate Gel is better than on a 4% Tris-glycine gel, but the molecular weight separation range is not as wide. See migration chart on previous page for migration patterns.

Novex® NuPAGE® premixed buffers and reagents

Convenient buffers for optimal results with Novex® NuPAGE® gels

Product	Quantity	Cat. No.
Novex® Gels*	Varies	Varies
NuPAGE® Sample Reducing Agent (10X)	250 µL	NP0004
	10 mL	NP0009
NuPAGE® Antioxidant	15 mL	NP0005
NuPAGE® LDS Sample Buffer (4X)	10 mL	NP0007
	250 mL	NP0008
NuPAGE® MOPS SDS Running Buffer (for Bis-Tris gels only) (20X)	500 mL	NP0001
	5 L	NP000102
NuPAGE® MES SDS Running Buffer (for Bis-Tris gels only) (20X)	500 mL	NP0002
	5 L	NP000202
NuPAGE® Tris-Acetate SDS Running Buffer (20X)	500 mL	LA0041
NuPAGE® MOPS SDS Buffer Kit (for Bis-Tris gels) contains 1 each of NP0001, NP0004, NP0005, NP0007	1 kit	NP0050
NuPAGE® MES SDS Buffer Kit (for Bis-Tris gels) contains 1 each of NP0002, NP0004, NP0005, NP0007	1 kit	NP0060
NuPAGE® Tris-Acetate SDS Buffer Kit (for Tris-acetate gels only) contains 1 each of LA0041, NP0004, NP0005, NP0007	1 kit	LA0050
NuPAGE® Transfer Buffer (20X)	125 mL	NP0006
	1 L	NP00061

* For additional gel formulations, percentages, and formats, go to www.invitrogen.com/novex1d.

Novex® Tris-Glycine Precast Gels

Precast Laemmli-style gels for protein analysis

Novex® Tris-glycine polyacrylamide gel chemistry is based on the Laemmli system [1] with minor modifications for maximum performance in the precast format. These gels do not contain SDS and can therefore be used to accurately separate both native and denatured proteins. Novex® Tris-Glycine Gels provide reproducible separation of a wide range of proteins into well-resolved bands. Novex® Tris-Glycine Gels are available in a variety of acrylamide percentages and in the mini (8 x 8 cm) or midi (8 x 13 cm) formats.

Reference

1. Laemmli UK (1970) *Nature* 227:680–685.

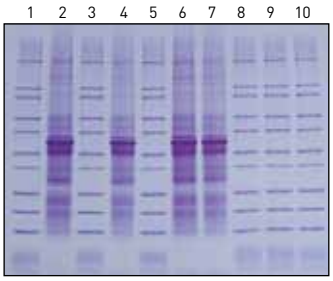


Figure 1. Separation on a Novex® 4–20% Tris-Glycine Gel (10-well, Coomassie® stained).

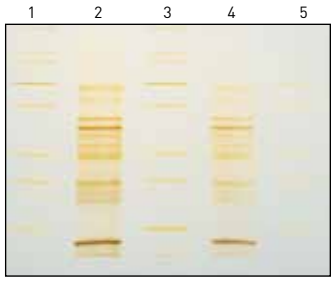


Figure 2. Separation on a Novex® 12% Tris-Glycine Gel (5-well, silver-stained).

Cat. No. for Novex® Tris-Glycine Mini Gels. Each box of Novex® Mini Gels includes 10 gels.

Tris-glycine gel percentage*	1.0 mm thickness				1.5 mm thickness (for larger sample volumes)		
	10 wells	12 wells	15 wells	2D well	10 wells	15 wells	2D well
4%	EC6055BOX	EC60552BOX	EC60655BOX		EC6058BOX	EC60585BOX	
6%	EC6065BOX	EC60652BOX	EC60655BOX		EC6068BOX	EC60685BOX	
10%	EC6075BOX	EC60752BOX	EC60755BOX	EC6076BOX	EC6078BOX	EC60785BOX	
12%	EC6005BOX	EC60052BOX	EC60055BOX	EC6006BOX†	EC6008BOX	EC60085BOX	
16%	EC6495BOX	EC64952BOX	EC64955BOX		EC6498BOX	EC64985BOX	
4–12%	EC6035BOX	EC60352BOX	EC60355BOX	EC6036BOX†	EC6038BOX	EC60385BOX	
8–16%	EC6045BOX	EC60452BOX	EC60455BOX		EC6048BOX	EC60485BOX	EC6049BOX†
4–20%	EC6025BOX	EC60252BOX	EC60255BOX	EC6026BOX	EC6028BOX	EC60285BOX	EC6029BOX
10–20%	EC6135BOX	EC61352BOX	EC61355BOX	EC6136BOX		EC61385BOX	

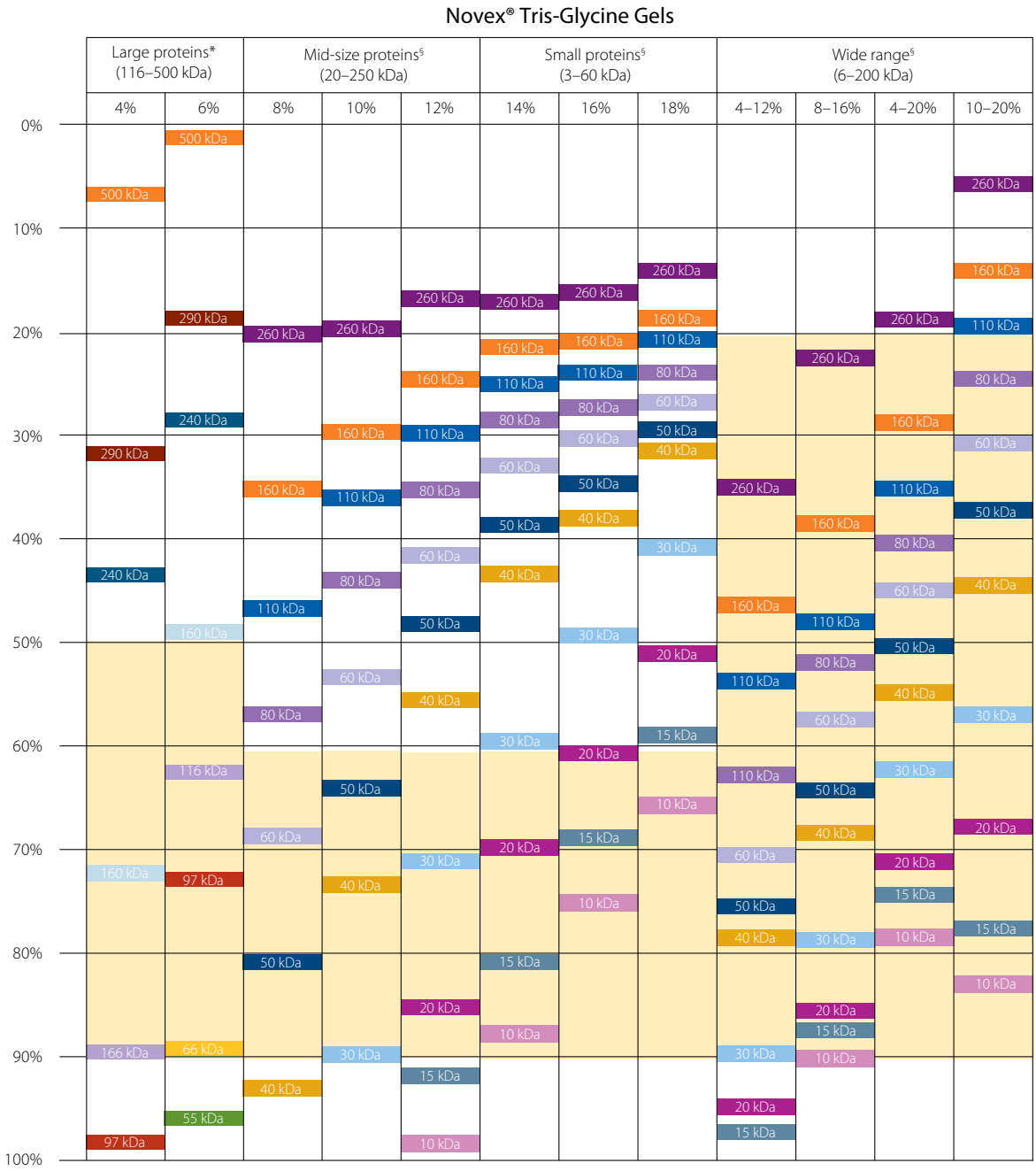
Cat. No. for Novex® Tris-Glycine Midi Gels. Each box of Novex® Midi Gels includes 10 gels.

Tris-glycine gel percentage*	Without midi gel adapters			With midi gel adapters‡		
	12 + 2 wells§	20 wells	26 wells§	12 + 2 wells§	20 wells	26 wells§
8%	WT0081BOX	WT0082BOX	WT0083BOX	WT0081A	WT0082A	WT0083A
10%	WT0101BOX	WT0102BOX	WT0103BOX	WT0101A	WT0102A	WT0103A
12%	WT0121BOX	WT0122BOX	WT0123BOX	WT0121A	WT0122A	WT0123A
4–12%	WT4121BOX	WT4122BOX	WT4123BOX	WT4121A	WT4122A	WT4123A
8–16%	WT8161BOX	WT8162BOX	WT8163BOX	WT8161A	WT8162A	WT8163A
4–20%	WT4201BOX	WT4202BOX	WT4203BOX	WT4201A	WT4202A	WT4203A

*For a more extensive list of gel percentages, well counts (including standard multichannel pipettor–compatible), and gel sizes, go to www.invitrogen.com/novex1d. †Custom order product. Please add an additional 1–2 weeks for delivery. ‡By attaching the Midi Gel Adapter (Cat. No. WA0999) to the gel cassette, you can run these midi gels in Bio-Rad's Criterion™ Cell. §Compatible with standard multi-channel pipeter. 12 + 2 well gels have 12 regular wells and 2 smaller wells for protein standards.

Choose the right Novex® Tris-Glycine Gel for your application

Use the migration pattern depicted in the table below to determine which Novex® Tris-Glycine Gel will provide the pattern most appropriate for your sample. For more selections, go to www.invitrogen.com/novex1d.



§ Migration patterns of Novex® Sharp Protein Standards (Cat. No. LC5800, Pre-stained; Cat. No. LC5801, Unstained) on Novex® Tris-Glycine Gels. * Migration patterns of HiMark™ Unstained Standard (Cat. No. LC5688) on Novex® Tris-Glycine Gels.

Premixed buffers and reagents for Novex® Tris-Glycine Gels

Convenient buffers for optimal results with Novex® gels

Product	Quantity	Cat. No.
Novex® Tris-Glycine SDS Sample Buffer (2X)	20 mL	LC2676
Novex® Tris-Glycine SDS Running Buffer (10X)	500 mL	LC2675
	4 x 1 L	LC26754
	5 L	LC26755
NuPAGE® Sample Reducing Agent (10X)	250 µL	NP0004
Novex® Tris-Glycine Native Sample Buffer (2X)	20 mL	LC2673
Novex® Tris-Glycine Native Running Buffer (10X)	500 mL	LC2672
Novex® Tris-Glycine Transfer Buffer (25X)	500 mL	LC3675

Precast gel accessories

Product	Quantity	Cat. No.
Midi-gel Adapters (for using midi gels in Bio-Rad Criterion™ Cell)	10/box	WA0999
Gel Knife	1 each	EI9010
Incubation Tray (10 x 14 cm)	8 trays and lids/pack	LC2102

For information regarding optimal loading volumes, go to www.invitrogen.com/novex1d.

Recommended loading volumes for mini gels.

		1 well	IPG well	2D well	9 well	10 well	12 well	15 well	17 well
Maximum sample volume/well	1.0 mm thickness	700 µL	7-cm IPG strip	400 µL	28 µL	25 µL	20 µL	15 µL	15 µL
	1.5 mm thickness	NA	NA	600 µL	NA	37 µL	NA	25 µL	NA
Maximum recommended load for optimal resolution†	Per protein band, Coomassie® stained	12 µg	NA	12 µg	0.5 µg	0.5 µg	0.5 µg	0.5 µg	0.5 µg

† NuPAGE® gels have a higher protein load capacity than Tris-glycine or Tricine gels.

Recommended loading volumes for midi gels.

	12 + 2 wells*	20 wells	26 wells*
Maximum sample volume/well‡	45 µL sample + 15 µL standards	25 µL	15 µL
Maximum protein load—Coomassie® stain	0.7 µg/band	0.5 µg/band	0.35 µg/band
Maximum protein load—silver stain	1.4 ng/band	1 ng/band	0.7 ng/band

‡ These volumes represent approximately 60% of the actual maximum volume of the wells. * Compatible with standard multichannel pipettor.

Novex® NativePAGE™ Bis-Tris Gel System

Reliable analysis of very large protein complexes

- Simplify resolution of large proteins (15–10,000 kDa)
- Analyze membrane-protein complexes in their native conformations
- Obtain better resolution than with Tris-glycine native electrophoresis

The Novex® NativePAGE™ Bis-Tris Gel System is a precast polyacrylamide mini-gel system that provides sensitive, high-resolution analysis of native membrane protein complexes, native soluble proteins, molecular mass estimations, and assessment of purity of native proteins. It is based on the blue native polyacrylamide gel electrophoresis (BN PAGE) technique developed by Schägger and von Jagow [1–3].

BN PAGE uses Coomassie® G-250 as the charge-shift molecule, which binds to proteins and confers a net negative charge while maintaining the proteins in their nondenatured native state. The near neutral pH of the Novex® NativePAGE™ Bis-Tris Gel System provides maximum stability of both the proteins and the gel matrix, resulting in highly sensitive analysis of native membrane-protein complexes and superior band resolution over traditional Tris-glycine gel systems.

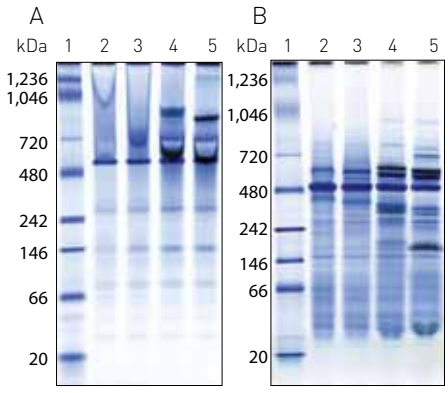


Figure 1. Improved separation with Novex® NativePAGE™ gels. Native electrophoresis was performed with Novex® 4–12% Tris-Glycine (**A**) and Novex® NativePAGE™ 4–16% Gels (**B**). Both gels were loaded with NativeMark™ standards (lane 1) and 18 mg spinach chloroplast extract solubilized in 0.25%, 0.5%, 1.0%, and 2.0% dodecylmaltoside (lanes 2–5, respectively). Stained with Colloidal Blue Staining Kit.

Product	Quantity	Cat. No.
Novex® NativePAGE™ 3–12% Bis-Tris Gels		
1.0 mm, 10 well	1 box	BN1001BOX
1.0 mm, 15 well	1 box	BN1003BOX
Novex® NativePAGE™ 4–16% Bis-Tris Gels		
1.0 mm, 10 well	1 box	BN1002BOX
1.0 mm, 15 well	1 box	BN1004BOX
NativePAGE™ Running Buffer Kit*	1 kit	BN2007
NativePAGE™ Sample Prep Kit†	1 kit	BN2008
NativePAGE™ Running Buffer (20X)	1 L	BN2001
NativePAGE™ Cathode Buffer Additive (20X)	250 mL	BN2002
NativePAGE™ Sample Buffer (4X)	10 mL	BN2003
NativePAGE™ 5% G-250 Sample Additive	0.5 mL	BN2004
10% DDM (n-dodecyl ß-D-maltoside)	1 mL	BN2005
5% Digitonin	1 mL	BN2006
NativeMark™ Unstained Protein Standard	250 µL	LC0725

*The NativePAGE™ Running Buffer Kit contains the NativePAGE™ Running Buffer (20X) and the NativePAGE™ Cathode Buffer Additive (20X). †The NativePAGE™ Sample Prep Kit includes the NativePAGE™ Sample Buffer (4X), 5% G-250 Sample Additive, 10% DDM, and 5% Digitonin.

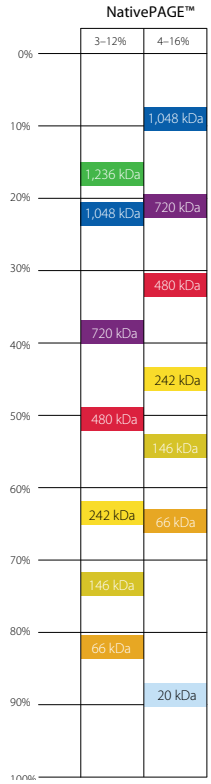


Table 2. Migration patterns of the NativeMark™ Unstained Protein Standard on Novex® NativePAGE™ gels.

References

1. Schägger H, von Jagow G (1991) *Anal Biochem* 199:223–231.
2. Schägger H, Cramer WA, von Jagow G (1994) *Anal Biochem* 217:220–230.
3. Schägger H (2001) *Meth Cell Biol* 65:231–244.

For more information on Novex® NativePAGE™ gels, go to www.invitrogen.com/native.

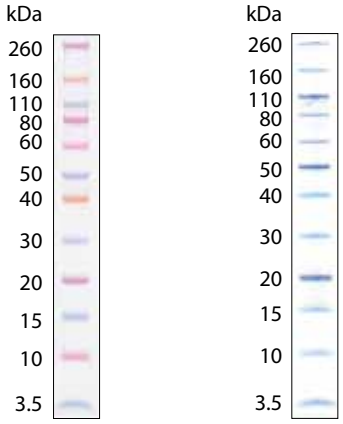
Novex® Sharp Pre-stained and Unstained Protein Standards

Accurate molecular weight estimations

- Sharpest bands—clearer estimation of MW
- Broadest MW range—estimation over a larger range (3.5 kDa to 260 kDa)
- Easy band identification—evenly spaced, ladder-like separation of bands

The Novex® Sharp Pre-stained Protein Standard consists of contrasting color bands for easy identification and allows you to monitor molecular weight separation during electrophoresis. Of all our standards it provides the sharpest bands in the broadest molecular weight range available. The Novex® Sharp Unstained Protein Standard provides bands that are unmodified by the presence of dye, for accurate molecular weight estimation for gel electrophoresis. Novex® Sharp Protein Standards are suitable for use with Novex® Tris-glycine, Tricine, and NuPAGE® gels.

Product	Quantity	Cat. No.
Novex® Sharp Pre-stained Protein Standard	2 x 250 µL	LC5800
Novex® Sharp Unstained Protein Standard	2 x 250 µL	LC5801

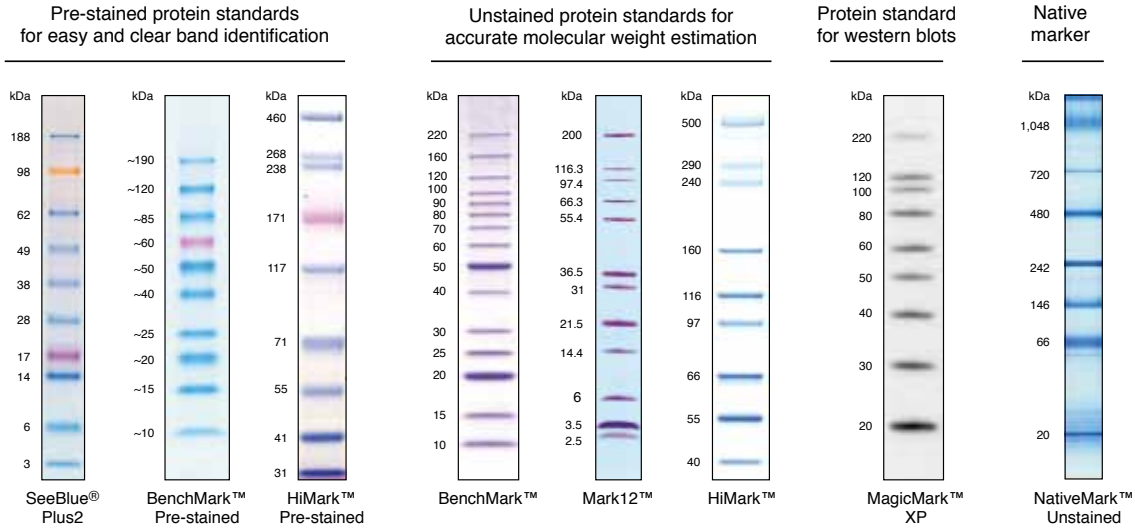


Novex® Sharp Pre-stained Protein Standard

Novex® Sharp Unstained Protein Standard

The Novex® Sharp Protein Standards. The Novex® Sharp Protein Standards contain 12 proteins that resolve into sharp, tight bands in the range of 3.5–260 kDa. The lanes show 10 µL of each standard run on a Novex® NuPAGE® 4–12% Bis-Tris Gel w/MES SDS buffer. The Novex® Sharp Unstained Standard was stained with SimplyBlue™ SafeStain.

Additional Novex® protein standards for gel electrophoresis

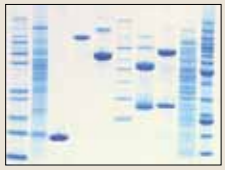
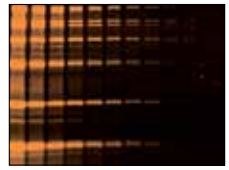


Standard	SeeBlue® Plus2	BenchMark™ Pre-stained	HiMark™ Pre-stained	BenchMark™	Mark12™	HiMark™	MagicMark™ XP	NativeMark™ Unstained
Cat. No.	LC5925	10748-010	LC5699	10747-012	LC5677	LC5688	LC5602	LC0725
Quantity	500 µL	2 x 250 µL	250 µL	2 x 250 µL	1 mL	250 µL	250 µL	250 µL

For more protein electrophoresis standards, such as NativeMark™ Standard for native gel electrophoresis, IEF Marker, and BenchMark™ His-Tagged and Fluorescent Markers, go to www.invitrogen.com/proteinstandards.

Sensitive stains for total-protein detection on gels

Life Technologies offers a number of staining reagents for total protein and posttranslationally modified proteins, including Coomassie® G-250 stain, silver stain, and a suite of compatible and highly sensitive fluorescent stains that allow direct quantitation of differential phosphorylation and glycosylation patterns as well as total-protein expression from a single sample on the same gel. Use the table below to find the stain that's right for your application.

Product	SimplyBlue™ SafeStain	SilverQuest™ Silver Stain	SYPRO® Ruby Protein Gel Stain	Coomassie Fluor™ Orange
	Fast, sensitive, and safe colloidal Coomassie® G-250–based stain for nanogram-level detection of proteins	Mass spectrometry (MS)–compatible silver stain ideal for colorimetric staining of low-abundance proteins, with sensitivity to sub-nanogram levels	Highly sensitive, ready-to-use fluorescent stain for the detection of low-abundance proteins in 1D or 2D gels. Also used for Multiplexed Proteomics® analysis	Ready-to-use fluorescent stain for fast, simple, sensitive staining of proteins typically in less than an hour—no separate fixation or destaining steps required
				
Type of stain	Ready-to-use colloidal Coomassie® G-250	Ready-to-use MS-compatible silver stain	Ready-to-use fluorescent total-protein stain	Ready-to-use fluorescent total-protein stain
Detection/ imaging	Visual-colorimetric	Visual-colorimetric	UV transilluminator, blue-light box, or a laser scanner	UV transilluminator, blue-light box, or a laser scanner
Sensitivity	To 7 ng BSA	Subnanogram (to 0.3 ng)	Subnanogram (to 0.25–0.5 ng)	To 4–8 ng/band
Linear dynamic range	7–70 ng	1–10 ng	0.25–1,000 ng	4–1,000 ng
MS compatibility	Yes	Yes	Yes	Yes
Total staining time	12 min (microwave protocol) 3 hr (standard protocol)	<60 min (microwave protocol) 90 min (standard protocol)	90 min (microwave protocol); overnight (standard protocol for large-format gels)	30–60 min (standard protocol)
Protocol summary	Rinse, stain, water-based destain	Fix, sensitize, wash, stain, wash, develop, and stop	Fix, stain, and wash	Fix and stain, in one step
Shipping/ Storage	Store at room temperature	Store at room temperature	Store at room temperature	Store at room temperature
Stability	6 months	6 months	1 year (at least 9 months)	6 months
For densitometry	Yes	Not recommended	Integration of fluorescent signal	Integration of fluorescent signal
Staining membranes	PVDF	No	Use SYPRO® Ruby Blot Stain	No
Quantity (Cat. No.)	1 L (LC6060) 3.5 L (LC6065)	1 kit (LC6070)	200 mL (S12001) 1 L (S12000) 5 L (S21900)	1 L (C33250) 5 L (C33251)

The Novex® Reversible Membrane Protein Stain Kit allows complete, reversible staining of proteins on nitrocellulose and PVDF membranes, providing highly sensitive detection (<10 ng) with minimal background. For more details on this and other other stains like SYPRO® Ruby Blot Stain, Colloidal Blue Staining Kit, SilverXpress® Silver Staining Kit, SYPRO® Tangerine Protein Gel Stain, SYPRO® Orange and SYPRO® Red, as well as other Molecular Probes® stains like Multiplex Proteomics® Technology Stains (Pro-Q® Diamond Phosphoprotein Gel Stain and Pro-Q® Emerald Glycoprotein Gel Stain), go to www.invitrogen.com/proteinstains. For information on Qdot® nanocrystals, go to www.invitrogen.com/qdots.

Characterizing glycoproteins?

Harnessing the power of "click" chemistry, the Click-iT® Protein Analysis Detection Kits are used for detecting new protein synthesis or subclasses of glycoproteins modified with azido groups. This detection method is compatible with downstream mass spectrometry (MS) analyses including LC-MS/MS and MALDI-MS and can be used for differential analyses of glycoprotein subclasses, total glycoproteins, phosphoproteins, and total protein. Go to www.invitrogen.com/clickchemistry click to learn more.

Gel electrophoresis equipment

Product	Specifications	Quantity	Cat. No.
XCell *SureLock*® Mini-Cell for Mini Gels 	The XCell *SureLock*® Mini-Cell incorporates a gel tension wedge in place of the rear wedge used on earlier models, making it the easiest-to-use mini-cell on the market. • Gel capacity: Up to two Novex® mini-gels, or one XCell II™ Blot Module • Cell dimensions: 12.5 cm (l) x 14.4 cm (w) x 16 cm (h) • Upper buffer chamber requirement: 200 mL for Novex® mini gels • Lower buffer chamber requirement: 600 mL for Novex® mini gels • Electrical limits: 1,500 VDC or 75 W • Material: Polycarbonate • Accessories: XCell II™ Blot module is available for semi-wet protein transfers.	XCell *SureLock*® Mini-Cell, 1 unit* CE XCell *SureLock*® Mini-Cell w/ XCell II™ Blot Module Kit, 1 unit* CE	EI0001 EI0002
XCell6™ MultiGel Unit for Mini Gels 	Flexible apparatus for increased electrophoresis throughput with mini gels • Gel capacity: Up to six Novex® mini gels • Cell dimensions: 28 cm (l) x 16 cm (w) x 19 cm (h) • Upper buffer chamber requirement: 3 x 250 mL • Lower Buffer chamber requirement: 670 mL • Electrical limits: 600 VDC or 160 W	XCell6™ MultiGel Unit, 1 unit* CE	EI0006
XCell4 *SureLock*® Midi Cell for Midi Gels 	Advanced apparatus for easier, more reliable electrophoresis with midi gels • Gel dimensions: 87 mm (l) x 133 mm (w) x 1 mm (t) • Cassette dimensions: 10.7 cm (l) x 15.0 cm (w) x 0.53 cm (t) • Gel capacity: Up to 4 gels • Cell dimensions: 21.1 cm (l) x 19.2 cm (w) x 16.3 cm (h) • Upper buffer chamber volume: 175 mL per gel • Lower buffer chamber volume: 540–700 mL • Electrical limits: 600 VDC or 160 W • Operating temperature: 4–40°C • Materials: Acrylic, delrin, high-density polyethylene, nylon, platinum, polycarbonate, gold-plated copper, plasticized silicone	XCell4 *SureLock*® Midi Cell, 1 unit* CE	WR0100
ZOOM® Dual Power 	The ZOOM® Dual Power is a microprocessor-controlled, programmable power supply capable of running both high-voltage/low-current and high-current/low-voltage applications concurrently. The high-voltage/low-current section is ideal for sample fractionation, IPG strips, IEF, and DNA sequencing applications. The low-voltage/high-current section is for SDS-PAGE, native PAGE, second-dimension SDS-PAGE, DNA/RNA electrophoresis, and blotting applications. Four sets of output jacks on each side of the unit allow for multiple apparatuses to run simultaneously, offering greater workflow efficiency and higher sample throughput.	ZOOM® Dual Power (220/240 VAC 47 60 Hz) OE (for sale in the EU only)* CE ZOOM® Dual Power (220/240 VAC 47 60 Hz) OE (for sale in the UK only)* CE	ZP10002 ZP10002UK
PowerEase® 500 Power Supply 	The PowerEase® 500 Power Supply is designed specifically for mini-gel electrophoresis. It offers extensive programming capabilities, including eight preset and four custom methods for you to set to your own preferences. The simple, intuitive PowerEase® interface is faster and easier to use than traditional "dial-in" power supplies. In addition, PowerEase® 500 features: constant voltage, current, or power settings; buffer temperature monitoring capability; "on the fly" program editing; and compatibility with most printers using a parallel port.	PowerEase® 500 Power Supply (100–120 VAC, 50–60 Hz), 1 unit* CE	EI8700EU Europe EI8700UK UK EI8700AG Switzerland

For gel electrophoresis accessories like StainEase® Staining Tray, DryEase® Mini-gel Drying System, Empty Plastic Gel Cassettes and Combs, and Gel Loading Tips, go to www.invitrogen.com/novex.

XCell *SureLock*® Mini Cell, XCell6™ MultiGel, and XCell4 *SureLock*® Midi Cell units chemical resistance: Impervious to alcohol, but not compatible with chlorinated hydrocarbons (e.g., chloroform), aromatic hydrocarbons (e.g., toluene, benzene) or acetone.

* The CE mark symbolizes that the product conforms to all applicable European Community provisions for which CE marking is required.

Specialty, one–dimensional, gel electrophoresis products and kits

High-throughput bufferless precast gels—E-PAGE™ Gel System www.invitrogen.com/epage

High-resolution peptide analysis—Novex® Tricine Gels www.invitrogen.com/novex1d

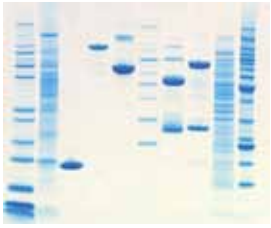
Easy in-gel protease analysis—Novex® Zymogram Gels www.invitrogen.com/novex1d

Simple isoelectric point (IEF) determinations—Novex®Precast Vertical IEF Gels www.invitrogen.com/novex1d

Overview of western blotting

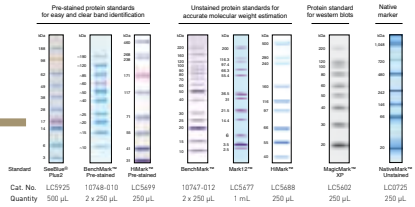
Sample preparation (pages 73–81, 256–257)

Protein separation



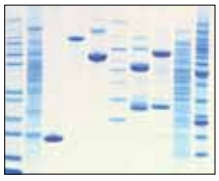
Protein gels
pages 260–267

Electrophoresis
page 270



Protein standards
page 268

Western transfer



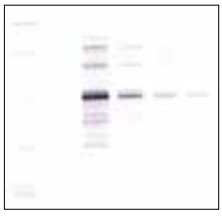
Membrane staining
page 269

Western transfer
page 272

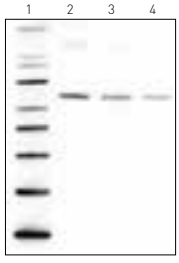
Western detection (manual or automated)

Western detection kits
(iBlot®, WesternBreeze®, Novex® kits)
pages 275–276

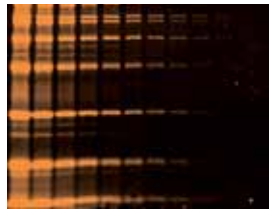
Western processing equipment
page 274



Chromogenic



Chemiluminescent



Fluorescent

iBlot® Dry Blotting System

Revolutionary dry electroblotting system for 7-minute protein transfers—available for both nitrocellulose and PVDF

- Complete transfer typically in 7 minutes or less
- High blotting efficiency and evenness
- Increased blotting reliability and reproducibility

The iBlot® Dry Blotting System efficiently and reliably blots proteins from polyacrylamide gels typically in seven minutes or less without buffers or an external power supply. A self-contained unit, the iBlot® device uses disposable blotting stacks with integrated transfer membranes, offering the convenience of a bufferless, plug-and-play system. In addition to ease and convenience, the iBlot® system offers high-efficiency protein transfer for high downstream detection sensitivities—in many cases, higher than conventional methods. In the end, you'll achieve more accurate detection results using less sample and less time.

When used with the accelerated NuPAGE® method (page 260) and iBlot® Western Detection Kits (page 275), complete protein separation to detection can typically be done in just 1 hour.

Figure 1. iBlot® Dry Blotting System.

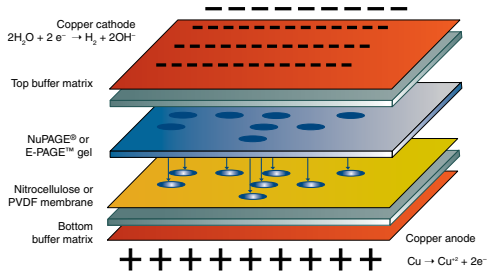


Figure 2. How iBlot® dry blotting works.

Faster protein transfer with the iBlot® Dry Blotting System.

	iBlot® Dry Blotting System	Semi-dry transfer	Wet/semi-wet transfer
Buffer preparation	0 min	30 min	30 min
Soaking gel in transfer buffer	0 min	20 min	0 min
Assembling layers	2 min	10 min	10 min
Transfer	7 min	45–90 min	1–3 hr
Cleanup	0 min	10 min	10 min
Total	9 min	1 hr, 55 min–2 hr, 40 min	1 hr, 50 min–3 hr, 50 min
Time saved with the iBlot® system		1 hr, 45 min–2 hr, 30 min	1 hr, 40 min–3 hr, 40 min

Product	Quantity	Cat. No.
iBlot® Gel Transfer Device	1 each	IB1001EU (EU adaptor) IB1001UK (UK adaptor)
iBlot® Transfer Stack, Regular (Nitrocellulose)	10 sets/box	IB301001
iBlot® Transfer Stack, Mini (Nitrocellulose)	10 sets/box	IB301002
iBlot® Transfer Stack, PVDF Regular	10 sets/box	IB401001
iBlot® Transfer Stack, PVDF Mini	10 sets/box	IB401002
iBlot® Transfer Stack, Regular (Nitrocellulose)	30 sets/box	IB301031
iBlot® Transfer Stack, Mini (Nitrocellulose)	30 sets/box	IB301032
iBlot® Transfer Stack, PVDF Regular	30 sets/box	IB401031
iBlot® Transfer Stack, PVDF Mini	30 sets/box	IB401032
Blotting Roller, 8.6 cm wide	1 each	LC2100

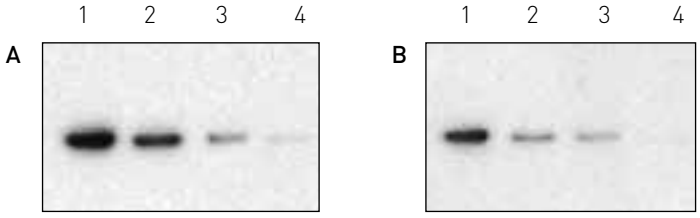
The Regular size fits Novex® Midi Gels (8 cm x 13 cm), E-PAGE™ *48* and *96* gels, or two mini gels (8 cm x 8 cm). The Mini size is suitable for a single mini gel (8 cm x 8 cm). Each iBlot® bottom transfer stack includes an integrated 0.2-µm nitrocellulose or 0.2-µm polyvinylidene difluoride (PVDF) membrane for protein immobilization.

For more details, a video demonstration, and to view other blotting equipment and accessories, go to www.invitrogen.com/iblot.

Precut blotting membranes

Simplify blotting set up with precut membranes/filter-paper sandwiches

Life Technologies makes blotting easier by providing a variety of precut, preassembled membrane/filter-paper sandwiches for mini and midi E-PAGE™ gels. A protein's properties (i.e., charge, hydrophobicity, etc.) affect its ability to bind to membrane surfaces. Finding the right membrane may require experimenting with your specific protein on different membranes.



High signal achieved on Invitrolon™ PVDF. A 53 kDa protein containing a c-Myc epitope was transferred onto Invitrolon™ PVDF (**A**) and another manufacturer's 0.45 µm PVDF (**B**) membrane. Both PVDF membranes were probed with a 1:500 dilution of mouse anti-myc antibody then developed with the WesternBreeze® Chemiluminescent Anti-Mouse Kit. Blots shown here are 2-minute exposures on X-ray film. Lanes 1–4: 2 ng, 1 ng, 0.5 ng, and 0.2 ng of protein, respectively.

Product	Applications	Size	No. of membrane/filter-paper sandwiches	Cat. No.
Nitrocellulose, 0.2 µm pore size	Western transfers, solid-phase assay systems, amino acid analysis	8.3 cm x 7.3 cm (mini)	20	LC2000
		8.5 cm x 13.5 cm (midi)	16	LC2009
Nitrocellulose, 0.45 µm pore size	Western transfers, solid-phase assay systems, amino acid analysis	8.3 cm x 7.3 cm (mini)	20	LC2001
		8.5 cm x 13.5 cm(midi)	16	LC2006
Invitrolon™ PVDF 0.45 µm pore size	Western transfers, solid-phase assay systems, amino acid analysis, reprobing	8.3 cm x 7.3 cm (mini)	20	LC2005
		8.5 cm x 13.5 cm (midi)	16	LC2007
PVDF, 0.2 µm pore size	Western transfers, solid-phase assay systems, amino acid analysis, reprobing	8.3 cm x 7.3 cm (mini)	20	LC2002
Nylon, 0.45 µm pore size	Southern, northern, and western transfers, solid-phase immobilization, dry chemistry test strips, enzyme immobilization, gene probe assays	8.3 cm x 7.3 cm (mini)	20	LC2003

For more blotting membranes, filter papers, pads, and accessories, go to www.invitrogen.com/blotting.

BenchPro® 4100 Western Processing System

Just push "run" and walk away

- Reduces tedious hands-on work and human errors
- Minimizes cross–contamination and no clean up
- Automate without protocol changes
- Compatible with all chemiluminescent, chromogenic, and fluorescent immunodetection reagents and protocols

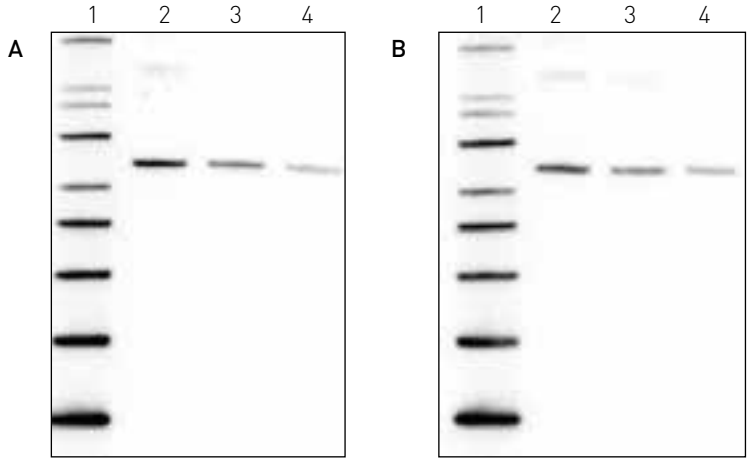
The BenchPro® 4100 Western Processing System helps consistently and accurately deliver the correct volumes of solutions to the membrane at precise times. This automation removes the tedious and repetitive liquid handling steps associated with manual western blot processing. Western blots processed using the BenchPro® 4100 system have minimal human processing errors, so you get greater experimental reproducibility and data consistency, run after run.

The BenchPro® 4100 Western Processing System generates familiar results the first time and consistent results on subsequent runs. It is compatible with all chemiluminescent, chromogenic, and fluorescent immunodetection reagents and protocols. The BenchPro® 4100 system dramatically reduces the need for protocol optimization and allows you to perform immunoblotting your way, right away.

BenchPro® 4100 Western Processing System.

Product	Quantity	Cat. No.
BenchPro® 4100 Card Processing Station	1 each	WP0001
BenchPro® 4100 Western Card	10 cards	WP1001
BenchPro® 4100 Reagent Vials	50 vials	WP3001

For more information, go to www.invitrogen.com/benchpro4100.



Manual processing vs. BenchPro® 4100 processing of western blots. (A) Manual processing; (B) BenchPro® 4100 system processing. Lane 1: 8 µL of a 1:10 dilution of MagicMark™ XP Standard; lane 2: 50 ng BSA; lane 3: 25 ng BSA; lane 4: 10 ng BSA. Proteins were detected using rabbit anti-BSA antibody and WesternBreeze® Chemiluminescent Kit—Anti-Rabbit. Detection substrate was added to both membranes, and the blots were imaged at the same time.

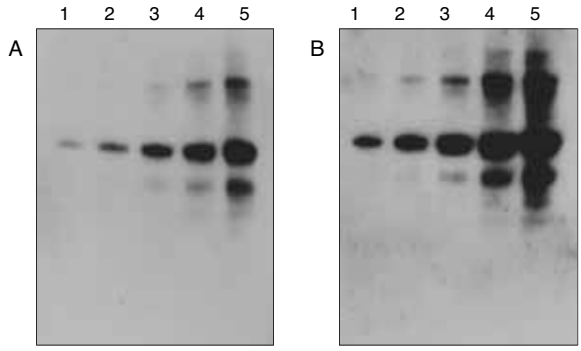
Novex® western blot detection kits

Life Technologies offers a broad range of reagents and kits for western blot detection. Whether you are using chromogenic, fluorescent, or chemiluminescence detection systems, we have the solutions you need.

iBlot® Western Detection Kits

- Fast—complete western detection in less than 25 minutes when used with the iBlot® Dry Blotting System
- Flexible—works with mini and midi gels
- Easy optimization—allows the use of different conditions for different sections of the blot

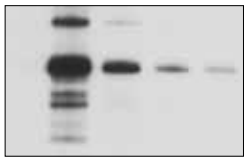
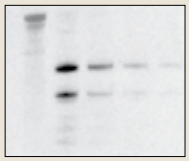
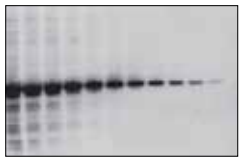
The iBlot® Western Detection Kits consist of iBlot® detection stacks and iBlot® detection reagents. The kits are available with anti-mouse or anti-rabbit secondary antibodies and are compatible with chemiluminescent and chromogenic detection. The iBlot® Western Detection Kits offer comparable or better sensitivity than the conventional protocols for the majority of the antibody-antigen pairs while providing significant time savings.



Comparison of iBlot® Western Detection Kit to WesternBreeze® Kit. Proteins from an SW480 (human colon adenocarcinoma cell line) lysate were transferred using the iBlot® Dry Blotting System. Mouse anti-p53 antibody was used as the primary antibody. Detection was performed with (A) the WesternBreeze® Chemiluminescent Kit—Anti-Mouse or (B) the iBlot® Western Detection, Chemiluminescent Kit (Anti-Mouse). The iBlot® Western Detection Kit detected the proteins with sensitivity comparable to the WesternBreeze® kit.

Product	Quantity	Cat. No.
iBlot® Gel Transfer Device	1 device	IB1001
iBlot® Western Detection Chemiluminescent Kit (Anti-Mouse) – Regular, 10 Pak	10 reactions	IB7110-01
iBlot® Western Detection Chemiluminescent Kit (Anti-Mouse) – Mini, 10 Pak	10 reactions	IB7110-02
iBlot® Western Detection Chemiluminescent Kit (Anti-Rabbit) – Regular, 10 Pak	10 reactions	IB7210-01
iBlot® Western Detection Chemiluminescent Kit (Anti–Rabbit) – Mini, 10 Pak	10 reactions	IB7210-02
iBlot® Western Detection Chromogenic Kit (Anti-Mouse) – Regular, 10 Pak	10 reactions	IB7310-01
iBlot® Western Detection Chromogenic Kit (Anti-Mouse) – Mini, 10 Pak	10 reactions	IB7310-02
iBlot® Western Detection Chromogenic Kit (Anti-Rabbit) – Regular, 10 Pak	10 reactions	IB7410-01
iBlot® Western Detection Chromogenic Kit (Anti-Rabbit) – Mini, 10 Pak	10 reactions	IB7410-02
IBlot® Western Detection Stacks (Regular), 10 Pak	10 reactions	IB7010-01
iBlot® Western Detection Stacks (Mini), 10 Pak	10 reactions	IB6310-02

Additional Novex® western detection kits

Product	WesternBreeze® Chromogenic Kit	WesternBreeze® Chemiluminescent Kit	Novex® ECL Chemiluminescent Kit	WesternDot™ Blotting Kit
				
	Strong, long-lasting signal with ready-to-use BCIP/NBT substrate for alkaline phosphatase	Highly sensitive detection with ready-to-use CDP-Star® chemiluminescent substrate for alkaline phosphatase	Using horseradish peroxidase, low-picogram levels of substrate can be detected with the Novex® ECL Chemiluminescent Kit.	Straightforward, high-sensitivity detection of proteins using fluorescent Qdot® 625 nanocrystals combined with high-affinity streptavidin-biotin binding reaction
Sensitivity	Low-picogram	High-femtogram	High-femtogram	Low-picogram
Emission duration	Months to years	Days	Hours	Days to months
Additional equipment required	•None •Optional: white-light camera or scanner	•Darkroom •Autorad/X-ray film and developer •Scanner •Luminescent imager	•Autorad/X-ray film and developer •Scanner •Luminescent imager	•UV transilluminator or UV illuminator •Digital camera with orange/red filter •Laser scanner

Product	Quantity	Cat. No.
Chromogenic and chemiluminescent immunodetection		
WesternBreeze® Chromogenic Kit—Anti-Mouse	1 kit	WB7103
WesternBreeze® Chromogenic Kit—Anti-Rabbit	1 kit	WB7105
WesternBreeze® Chromogenic Kit—Anti-Goat	1 kit	WB7107
WesternBreeze® Chemiluminescent Kit—Anti-Mouse	1 kit	WB7104
WesternBreeze® Chemiluminescent Kit—Anti-Rabbit	1 kit	WB7106
WesternBreeze® Chemiluminescent Kit—Anti-Goat	1 kit	WB7108
WesternBreeze® Blocker/Diluent (part A and B)	80 mL each	WB7050
WesternBreeze® Wash Solution (16X)	2 x 100 mL	WB7003
Easy molecular weight estimation directly on western blots		
MagicMark™ XP Western Protein Standard	250 µL	LC5602
Novex® ECL Chemiluminescent Kit	1 kit	WP20005
WesternDot™ blotting kits		
WesternDot™ 625 Goat Anti-Mouse Western Blot Kit	1 kit	W10132
WesternDot™ 625 Goat Anti-Rabbit Western Blot Kit	1 kit	W10142

Access the Novex® library of antibodies, recombinant proteins, and immunoassays

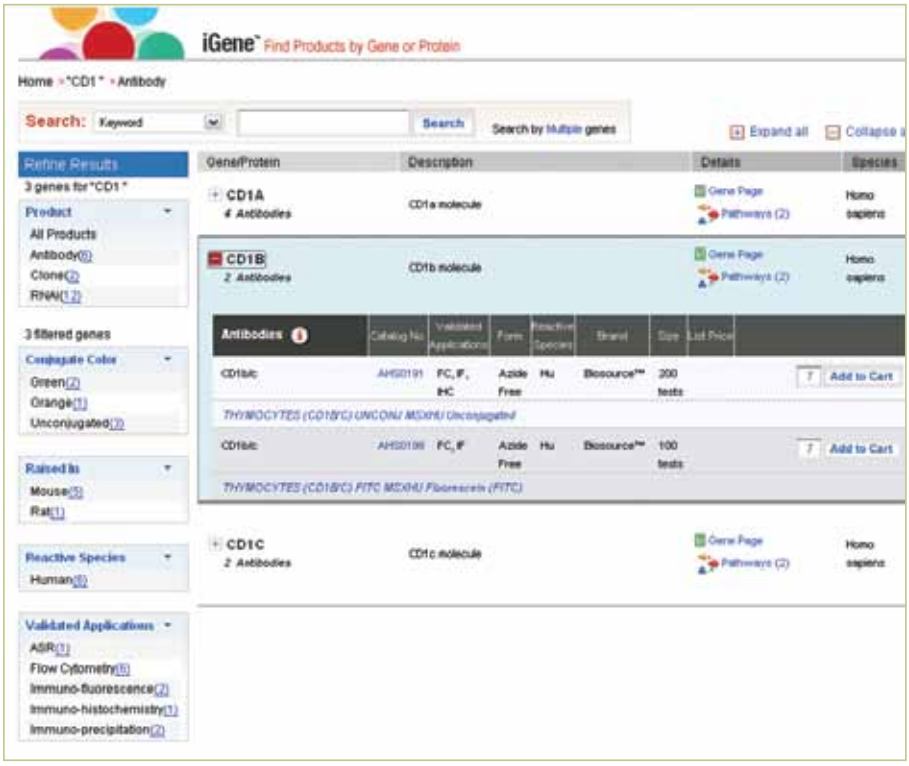
Life Technologies has an extensive menu of Gibco® recombinant proteins and application-validated Novex® antibodies. Our ever-expanding portfolio includes primary antibodies for phosphorylation, site-specific targets, cell junction proteins, neuroscience proteins, and many more targets. We also offer antibodies for dyes and haptens, epitope tags, phosphoamino acids, as well as isotype controls. Our expansive secondary antibody products include Alexa Fluor® and Qdot® fluorescent conjugates as well as conjugates of classic dyes, enzymes, R-PE, and biotin.

Go to www.invitrogen.com/antibodies to find the antibodies and recombinant proteins you need today.

Immunoassays

Life Technologies also offers a large menu of Novex® assays to measure cytokine and signaling proteins, as well as kinase activity.

To learn more, go to www.invitrogen.com/luminex, www.invitrogen.com/elisa, and www.invitrogen.com/omnia.



Go to www.invitrogen.com/antibodies to connect to the Linnea® Guide to Antibodies.

Enter your protein target in the search box. Then simply refine your search using our filters for application type, conjugate, reactivity, or host.

Expand your proteomics research

Life Technologies offers innovative tools to make your molecular interaction and biomarker discovery research more efficient and successful. From innovative sample fractionation with the ZOOM® IEF Fractionator and mass spectrometry technologies, like SILAC™ Protein Identification and Quantitation Kits (now also available for stem cells), to ProtoArray® Protein Microarrays and the ProQuest™ Two-Hybrid System, Life Technologies has streamlined protein expression workflows, allowing for more accurate and thorough sample profiling.

ZOOM® Benchtop Proteomics System

The ZOOM® System offers an integrated solution to protein profiling and easy 2D electrophoresis.

It consists of the ZOOM® IEF Fractionator, ZOOM® disks, ZOOM® 2D Solubilizers, ZOOM® IPG Runner and Strips, ZOOM® Novex® 2D Gels, buffers, MS-compatible stains, and gel runners.

ZOOM® IEF Fractionator

ZOOM® IPG Runner and 2D Gel System

For more information, go to www.invitrogen.com/zoom

Protein tagging and mass spectrometry reagents

Perform metabolic labeling with SILAC™ Protein Identification and Quantitation Kits. Identify and quantitate crucial proteins involved in disease and altered phenotypes, diagnostic biomarkers, and targets for therapeutic intervention.

Chemically label, track, and profile protein samples with cleavable ICAT® and iTRAQ® reagents.

For more information, go to www.invitrogen.com/piq

ProtoArray® Protein Microarrays

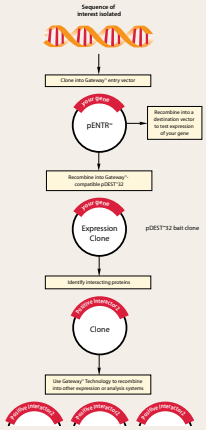
Identify novel protein biomarkers, map protein-protein interactions important to biochemical pathways, discover and identify drug target pathways, profile antibody specificity for research and therapeutic antibody development, and more.

For more information, go to www.invitrogen.com/protoarray

ProQuest™ Two-Hybrid System with Gateway® Technology

A key part of gene functional analysis and potential drug target discovery is an understanding of how proteins interact within the cell. The ProQuest™ Two-Hybrid System with Gateway® technology facilitates the characterization of these interactions in yeast systems.

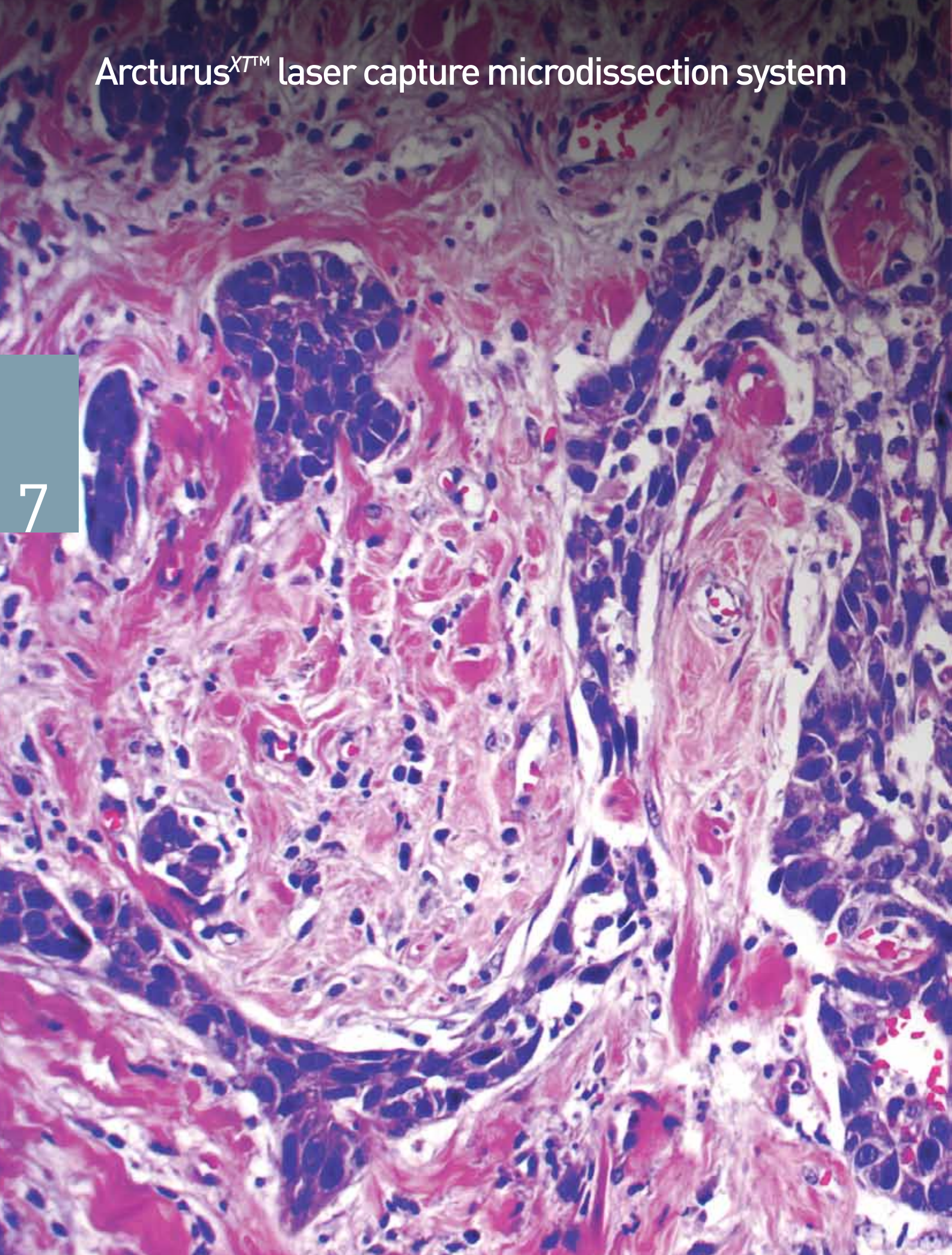
This technology offers reliable protein-protein interaction results, faster and easier, and with fewer false positives than other yeast two-hybrid technologies.



For more information, go to www.invitrogen.com/y2h

Arcturus^{XT}™ laser capture microdissection system

7



Applied Biosystems® Arcturus^{XT}™ Laser Capture Microdissection System 281

Arcturus$^{XT™}$ Laser Capture Microdissection System

Laser capture microdissection and UV laser cutting in a single system

- Two lasers in a single system
- Open, modular platform to suit your research
- Simple, intuitive operation
- Superior image quality
- Sample custody maintained at all times

The Applied Biosystems® Arcturus$^{XT™}$ Laser Capture Microdissection System is a unique microdissection instrument that combines infrared (IR) laser capture microdissection (LCM) and ultraviolet (UV) laser cutting in one platform. The open, modular design enables unparalleled research flexibility and versatility. The system allows you to maintain custody of the sample throughout the experiment, helping to ensure that only the desired material has been collected.

Two lasers in a single platform

The solid-state IR laser, exclusive to the Arcturus$^{XT™}$ system, delivers a gentle capture technique that preserves biomolecular integrity and is ideal for single cells and small numbers of cells. The solid-state UV laser permits unprecedented speed and precision and is well-suited for microdissecting dense tissue structures and for capturing large numbers of cells. This unique combination allows you to easily collect individual cells and large regions from the same sample, confidently microdissect adjacent cells in a single sample, and rapidly microdissect challenging samples.

Modular configuration for maximum flexibility

The Applied Biosystems® Arcturus$^{XT™}$ Laser Capture Microdissection System allows you to choose from a variety of modules to fit your research requirements. The system utilizes a Nikon Eclipse® Ti-E inverted research microscope with top-of-the-line options. Each instrument has IR-enabled LCM, an interactive pen-display monitor, and a trackball-actuated stage for easy and ergonomic navigation. The system may be configured with LED bright-field illumination or high-intensity halogen illumination.

The available microscope port allows you to modify the system for alternate applications, such as adding a second camera for high-resolution imaging. The open system design also makes possible the easy exchange of stage inserts to accommodate alternate sample formats such as larger slides and petri dishes. The system may be purchased with or without UV laser cutting and attenuable fluorescence. In addition, a wide range of objectives are available, including 2x–100x dry and 100x oil.

The Applied Biosystems® Arcturus$^{XT™}$ Laser Capture Microdissection System.

Flexibility in sample source and preparation

The unique combination of IR laser capture and UV laser cutting permits the use of any slide type, including glass membrane or framed membrane slides, for contact or non-contact microdissection, as well as low-cost plain glass slides. In addition, a wide variety of specimen preparations may be used with the Applied Biosystems® Arcturus$^{XT™}$ Laser Capture Microdissection System.

Simple software automates your workflow

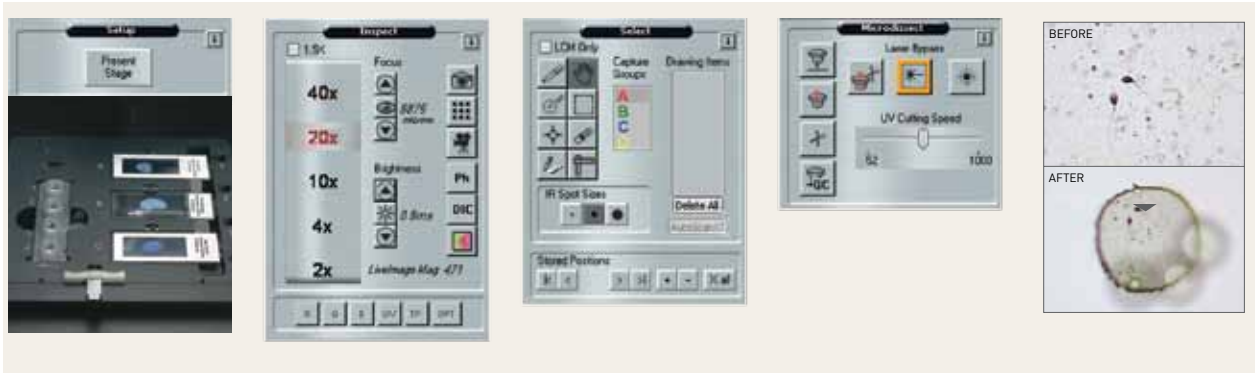
The Arcturus$^{XT™}$ System Software simplifies your laser capture microdissection workflow (see figure below). With the click of a mouse, you can control all system operations, including stage translation, slide and objective selection, focus and light intensity, laser parameters, cap transfers (including QC confirmation), and camera settings. Automatic electronic documentation may be employed to record each step of the process. Static images and live video can be taken at any point during the process, providing a record of the entire experiment. The optional AutoScanXT Image Analysis Software Module automatically identifies cells and regions based on user-defined criteria, greatly reducing the overall time required to perform a microdissection experiment.

The complete solution for microgenomics

Applied Biosystems® Arcturus® products offer everything you need for microgenomics. Kits are available for tissue staining, extraction, amplification, and labeling.

- HistoGene® Kits are available for frozen tissue and immunofluorescence staining, providing excellent contrast while preserving nucleic acid integrity and quality.
- PicoPure® Kits are available for reliable and reproducible DNA and RNA extraction and isolation from small numbers of cells.
- RiboAmp® PLUS Kits are available for RNA linear amplification from frozen samples with as little as 1 ng of total RNA.
- Paradise® PLUS Reagents are available for staining, RNA isolation and amplification, and whole-transcript reverse transcription from formalin-fixed, paraffin-embedded (FFPE) samples.
- Turbo Labeling™ Kits are available for nonenzymatic labeling of unmodified amplified RNA (aRNA) for microarray gene expression profiling. Choose from Cy®3, Cy®5, or biotin labeling to fit with your downstream analysis.

Step 1: Set up	Step 2: Inspect	Step 3: Select	Step 4: Microdissect	Step 5: QC
Load materials onto stage, and input important study information.	Identify cells of interest using the fully automated microscope tools, including autofocus and digital zoom.	Use simple drawing tools to designate cells for microdissection by drawing freehand or using defined-area circles.	Save time and increase productivity with easy-to-use tools for laser cutting and laser capture.	Inspect the microdissected material on the CapSure® cap for positive identification of captured material and assurance of proceeding with the exact cells of interest.



Arcturus$^{XT™}$ System workflow. The streamlined user interface simplifies your workflow—go from sample loading to extraction of biomolecules in just 5 steps.

Product	Quantity	Cat. No.
Applied Biosystems® Arcturus$^{XT™}$ Laser Capture Microdissection Systems		
Arcturus$^{XT™}$ Microdissection Instrument (LCM only)	1 instrument	ArcturusXT
Arcturus$^{XT™}$ Enhanced (EUV) Laser Cutting	1 instrument	0310-5950
Arcturus$^{XT™}$ Basic UV Laser Cutting	1 instrument	0310-5538
Applied Biosystems® Arcturus® CapSure® LCM Caps and Accessories		
CapSure® Macro LCM Caps	48 caps	LCM0211
CapSure® Macro LCM Caps, Bulk Pack	240 caps	LCM0212
CapSure® HS LCM Caps Starter Pack with Alignment Tray and Incubation Block	24 caps	LCM0213
CapSure® HS LCM Caps	32 caps	LCM0214
CapSure® HS LCM Caps, Bulk Pack	160 caps	LCM0215
PEN Membrane Frame Slides	50 slides	LCM0521
PEN Membrane Glass Slides	50 slides	LCM0522
PEN Membrane Frame Slides for Live Cell Microdissection	5 slides	LCM0530
PEN Membrane Frame Slides for Live Cell Microdissection, Bulk Pack	25 slides	LCM0531
Arcturus$^{XT™}$ Live Cell Growth Chambers	6 chambers (sterile)	5000300
Arcturus$^{XT™}$ Microdissection Petri Dishes	6 petri dishes (sterile)	5000301
Applied Biosystems® Arcturus® Microgenomics Reagents		
Arcturus® HistoGene® LCM Frozen Section Staining Kit	72 samples	KIT0401
Arcturus® HistoGene® LCM Immunofluorescence Staining Kit	32 samples	KIT0420
Arcturus® PicoPure® DNA Extraction Kit	150 extractions (HS Cap) 30 extractions (Macro Cap)	KIT0103
Arcturus® PicoPure® RNA Isolation Kit	40 isolations	KIT0204
Arcturus® RiboAmp® PLUS RNA Amplification Kit	(12) 1-round or (6) 2-round amplifications	KIT0521
Arcturus® RiboAmp® PLUS HS RNA Amplification Kit (High Sensitivity)	(6) 2-round amplifications	KIT0525
Arcturus® RiboAmp® PLUS Kit with Amino Allyl Labeling	6 amplifications	KIT0521AA
Arcturus® RiboAmp® PLUS Kit with Biotin Labeling	12 amplifications	KIT0511B
Arcturus® RiboAmp® PLUS Kit with Cy3 Labeling	12 amplifications	KIT0511C
Arcturus® RiboAmp® PLUS Kit with Cy5 Labeling	12 amplifications	KIT0511D
Arcturus® RiboAmp® HS PLUS Kit with Amino Allyl Labeling	6 amplifications	KIT0525AA
Arcturus® RiboAmp® HS PLUS Kit with Biotin Labeling	12 amplifications	KIT0515B
Arcturus® RiboAmp® HS PLUS Kit with Cy3 Labeling	12 amplifications	KIT0515C
Arcturus® RiboAmp® HS PLUS Kit with Cy5 Labeling	12 amplifications	KIT0515D
Arcturus® Paradise® PLUS Reagent System	12 samples	KIT0312
Arcturus® Paradise® PLUS Whole-Transcript RT Reagent System	12 samples	KIT0315
Arcturus® Paradise® PLUS qRT-PCR Kit	12 samples	KIT0310
Arcturus® Paradise® PLUS QC Kit	12 samples	KIT0313
Arcturus® Turbo Labeling™ Kit–Biotin	12 samples	KIT0608
Arcturus® Turbo Labeling™ Kit–Cy3	12 samples	KIT0609
Arcturus® Turbo Labeling™ Kit–Cy5	12 samples	KIT0610

Notes

appendix

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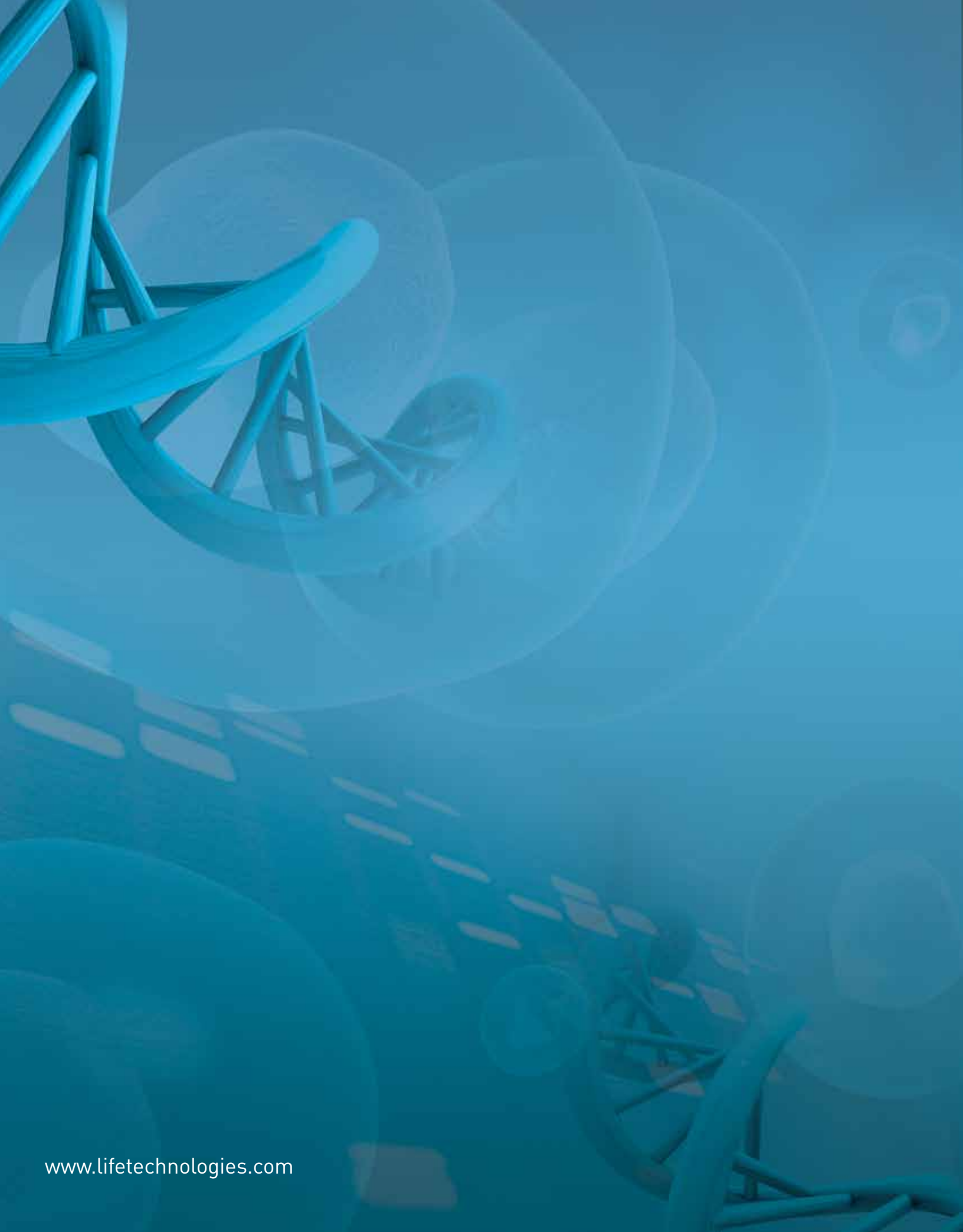

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