QUANTITATIVE PCR

Quantitative PCR Systems

+ Our QPCR systems meet your current research needs, offer flexibility for future research goals, and match your budget.

+ HIGH-PERFORMANCE + FULL-FEATURED + FLEXIBLE

STRATAGENE®

Bringing QPCR Systems to Every Lab

Real-time quantitative PCR (QPCR) is today's method of choice for quantifying nucleic acids.

By monitoring PCR amplification in real-time, it is possible to measure the reaction during the exponential phase of growth, a time when none of the reagents are limiting and the reaction is most efficient. This allows real-time QPCR to achieve more sensitive detection, better reproducibility, and a wider linear dynamic range than conventional methods.

Our Mx3000P® and Mx3005P™ QPCR Systems[a] offer the highest performance and flexibility in QPCR instrumentation at an affordable price.

CONTENTS:

The First High-Performance, Low-Cost QPCR System

With the Mx3000P® QPCR System, we were first to offer an affordable system that supported multiple applications.

Features of the Mx3000P® QPCR System

The Mx3000P® system is a high-performance, full-featured instrument system designed to accommodate basic experimental design and also offer the flexibility required for more advanced applications. The Mx3000P QPCR System brings real-time instrumentation to the individual researcher with a limited budget.

- Four optical channels with user-selected filters for greater flexibility
- Broad wavelength range excitation supports most fluorescent dyes
- Defined excitation and emission detection wavelengths ideal for superior multiplex results
- Single photomultiplier tube for detection ensures superior sensitivity and linear dynamic range to ten orders of magnitude (Figures 1 and 2)

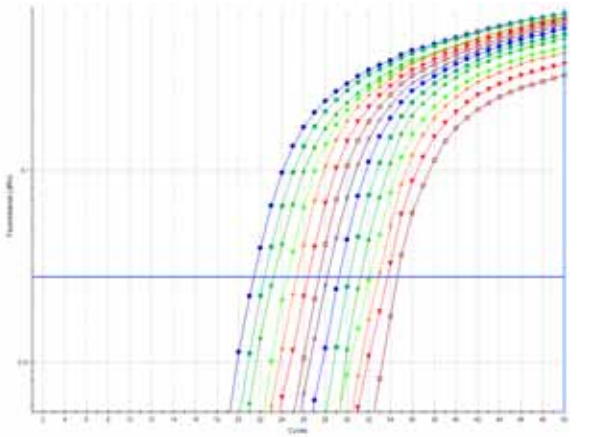

Figure 1
TWO-FOLD DILUTION SERIES
Four replicates of a dilution series from 20,000 to 1.2 copy equivalent of plasmid containing ß-actin target detected with a molecular beacon. Average delta Ct between dilutions is 0.95 cycles.

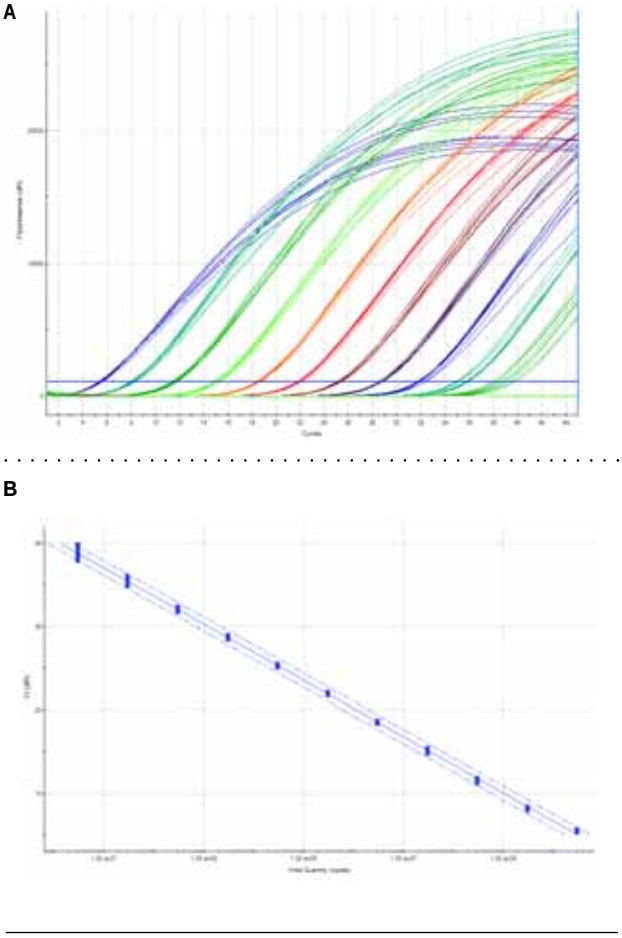

Figure 2
TEN ORDERS OF MAGNITUDE DYNAMIC RANGE
(A) Eight replicates of ten fold dilutions of plasmid DNA for ß-actin target. (B) Standard curve displayed with 99% confidence intervals and efficiency=98% and Rsq=0.999.

Superior Flexibility and High Performance Without the Cost

The Mx3005P™ QPCR System advances the proven Mx3000P® System. Offering unmatched flexibility and capability, the Mx3005P™ System can support even more real-time QPCR applications and chemistries to accommodate your research needs now and in the future.

Advanced Optical System

The Mx3005P™ System features the same optical scanning design as the Mx3000P system. A scanning fiber optic head ensures all wells 1) receive the same amount of excitation light, 2) are detected for the same amount of time, and 3) are the same distance from the detector. The scanning motion system of the fiber optic cable eliminates optical variation based on well position in the 96-well block, thus eliminating the need for signal correction by calibration or reference dyes. The photo-multiplier tube (PMT) has a large dynamic range of detection and a low signal to noise ratio, allowing low- to high-abundance targets to be accurately quantified. Excitation light is generated by a halogen lamp and delivered to the sample through fiber optics. The fiber optic bundle is coaxial so it delivers excitation light and simultaneously detects fluorescence emission from the sample. Detected light is delivered to the PMT via the fiber optic cable (Figure 3).

Figure 3
OPTICAL SYSTEM DESIGN

The halogen lamp in the instrument systems provides a wide-range of excitation allowing more dye flexibility with more intensity than standard light emitting diodes (LEDs). The excitation and emission filters are defined to narrow wave-lengths to minimize fluorescence signal crosstalk. Fiber optic bundles channel the light into the plate and back to the PMT to ensure minimal signal loss.

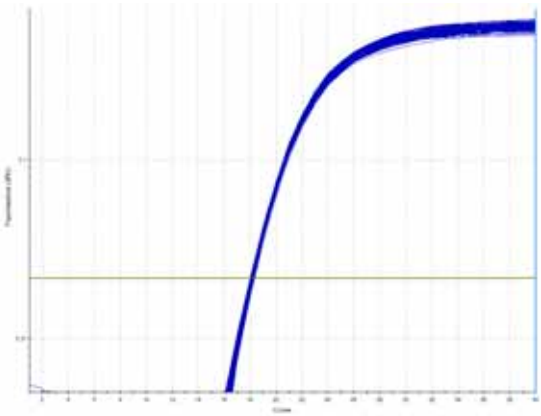

Figure 4
96-WELL UNIFORMITY

SYBR® Green I uniformity assay for ß-actin containing plasmid. Average Ct value at threshold is 18.1 and standard deviation of Ct values is 0.05. Ct range across 96 wells is 0.26 cycles (18.00 to 18.26).

Mx3005P™ Optical Filters

The new five-position customizable filter wheel design offers a multitude of dye choices, multiplex dye combinations enabling up to five targets per well, and the ability to detect fluorescence resonance energy transfer (FRET) signals. You can choose which five filters are installed in the instrument from a list of eight filter sets spanning deep blue dyes to far red dyes thus maximizing the useable wavelength spectrum (Table 3).

Precision Thermal System

The thermal system of the Mx3005P system shares the same Peltier-based design with the Mx3000P system. This system ensures uniform ramping and thermal accuracy for amplification reproducibility from well-to-well and run-to-run (Figure 4).

Powerful Data Analysis Software

The Mx3005P system uses the most advanced version of our graphical user interface and data analysis software. The MxPro™ QPCR Software is easy to use and organized by application so you can easily navigate the software and run assays quickly.

- Five optical channels with user-selected filters for greater flexibility
- Defined excitation and emission detection wavelengths are ideal for superior multiplex results, up to five targets simultaneously (Figure 5)
- Custom filter path selection to support FRET signal detection
- Open platform design supports all fluorescent chemistries and numerous applications (Figure 6)

Mx3005P™ QPCR System

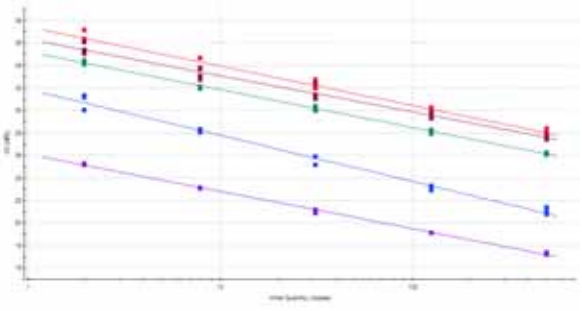

Figure 5
5-PLEX STANDARD CURVES

Three replicates of 4-fold dilutions of QPCR Human Universal Reference cDNA detecting five gene targets simultaneously. Detection from the highest abundance to the lowest abundance gene target (CYCLO to ENOS gene targets) spans a Ct range of 17-37 (delta Ct = 20).

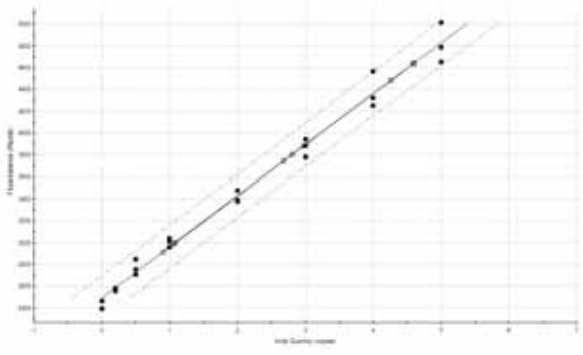

Figure 6
RNA QUANTIFICATION BY RIBOGREEN® PLATE READ EXPERIMENT

Three replicates of RNA standards from 500 pg to 5 ng. Standard curve generated from plate read experiment automatically quantitates RNA concentration in unknown samples. Ideal for quick and accurate quantification of RNA samples before performing gene expression QPCR experiments.

Novel Data Analysis in the Easiest-to-Operate QPCR Software Available

The MxPro™ QPCR Software for the Mx3000P® and Mx3005P™ Systems combines cutting-edge data analysis algorithms with intuitive organization designed for ultimate ease-of-use.

Setting Up and Running the Experiment

The experimental setup features well definition by target assay name (e.g., FAM = p53), automated standard curve setup, customized well naming, importing plates, and a flexible thermal profile setup to accommodate any thermal cycling programming (Figure 7). During the run, you can view your QPCR data in 96 individual wells or as a consolidated view on a single screen. In addition, the internal storage memory of the Mx3000P and Mx3005P systems allows you to analyze the data during a run in a standalone application while the instrument-connected application continues to collect data.

Data Analysis and Reporting the Results

The data analysis module includes two automated methods for baseline subtraction and threshold setting, both of which can be customized for any particular application. The MxPro QPCR software "Adaptive Baseline" algorithm dynamically assigns baseline start and end cycles for all amplification plots independently. This ensures optimal baseline correction and increases the reliability of accurately detecting high and low template concentrations in the same assay and analyze multiple assays simultaneously (Figure 8).

The Comparative Quantification module automatically determines gene expression fold change, calculates statistical error, and generates a publication quality chart (Figure 9). After analysis, the MxPro QPCR Software is capable of creating custom reports and exporting all plots, charts, and tables (Figure 10). Images can be directly exported to Microsoft PowerPoint®, and high-resolution bitmap images, Chart and plot data can be exported to Microsoft Excel®, .xml, and .txt formats to easily re-create the data in different formats (Figure 11). All text data can also be exported in any of these formats.

- Intuitive organization and easy-to-use
- View and analyze data in real-time
- Multiple customizable data analysis algorithms for baseline correction and threshold setting
- Export images directly from the software, export the raw data to re-create the image, and export the text data in multiple formats

Figure 7
FLEXIBLE, EASY TO USE PLATE SETUP
Import Plate Setup from previous experiments or templates. Assign assay/gene target names to dyes. Setup standard curve and replicates using auto-increment feature. Add well information to identify sample name, number, quantity, or any other sample specific information.

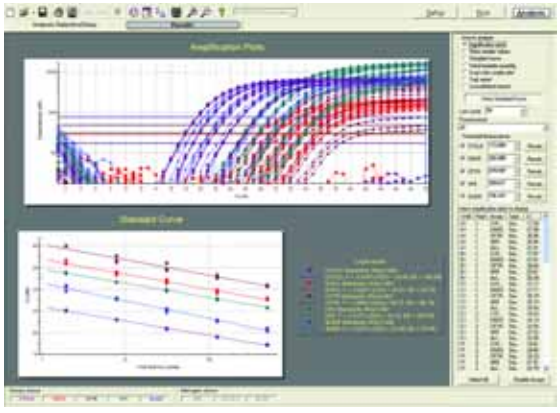

Figure 8
MULTIPLE DATA ANALYSIS VIEWS
View data in multiple formats. Amplification plot and standard curve data can be displayed simultaneously and changes in threshold are instantly displayed on the standard curve. The Adaptive Baseline algorithm ensures accurate quantification across all five gene targets.

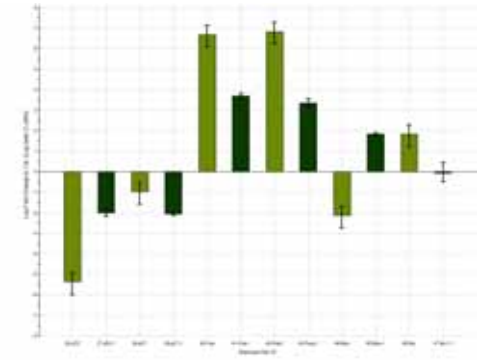

Figure 9
AUTOMATED ANALYSIS OF GENE EXPRESSION DATA

The Comparative Quantitation module in MxPro™ Software automatically calculates relative quantity for gene expression experiments. Data is displayed as normalized fold gene expression to a reference control on a log(2) scale with upper and lower error limits based on variation in the replicates. In the figure above, fold expression change for six genes across two treatments is displayed.

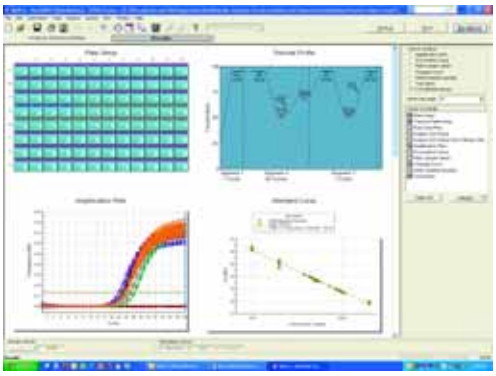

Figure 10
CREATE CUSTOM DATA REPORTS

Create a custom data report by determining report format and which data sets to display. In the figure above, Plate Setup, Thermal Profile, Amplification Plots, Standard Curve, and Text Report are selected for the report.

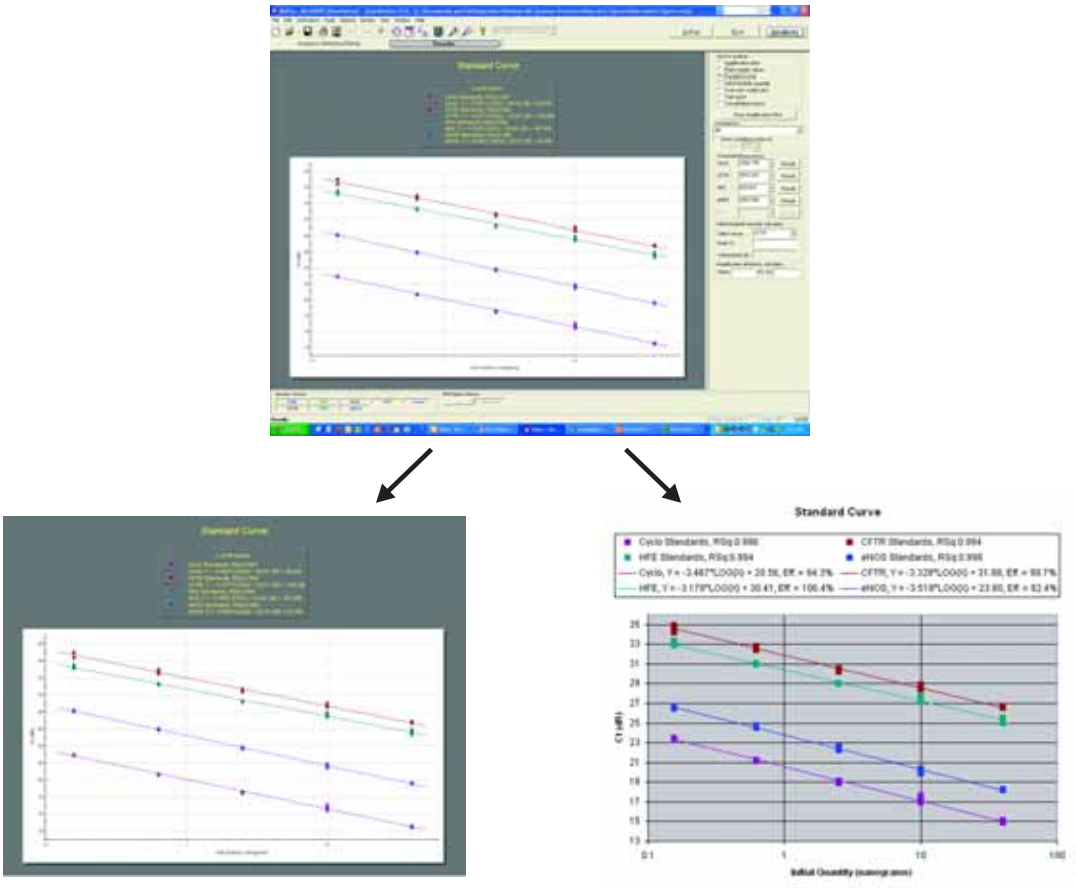

Figure 11
EXPORT DATA IN MULTIPLE FORMATS

Export any data set directly from the MxPro™ Software in .xls, .ppt, .bmp, .txt, or .xml format. Charts and graphs can be exported directly to Microsoft® PowerPoint® and Excel®. Exporting to Excel® automatically generates a data table and the chart.

Brilliant® and FullVelocity™ QPCR and QRT-PCR Reagents

We offer two families of QPCR and QRT-PCR reagents, Brilliant® and FullVelocity™ master mixes. Each uses a different PCR enzyme formulation to provide efficient, reproducible quantification using probe-based or SYBR® Green detection chemistries.

Brilliant® QPCR and QRT-PCR Master Mixes[b,c]

Brilliant® QPCR Reagents provide high sensitivity and broad dynamic range required for real-time PCR analysis. Brilliant QPCR Master Mixes utilize SureStart *Taq*® DNA polymerase[b] for high specificity, and StrataScript Reverse Transcriptase®[d] provides superior real-time QPCR sensitivity compared to RNase H(+) reverse transcriptases.

Brilliant® SideStep™ QPCR and QRT-PCR Master Mixes

Combining our SideStep™ Lysis and Stabilization Buffer[e] with our Brilliant® QPCR and QRT-PCR master mixes allows you to use cell lysates directly in a real-time QPCR amplification – skipping RNA purification steps. This method prevents sample loss and degradation, ensuring accurate gene quantification in downstream QRT-PCR. SideStep cell lysates can be stored for up to 6 months before QRT-PCR analysis.

FullVelocity™ QPCR and QRT-PCR Reagents[b]

FullVelocity™ QPCR Master Mixes provide a fast and economical system for real-time QPCR applications. FullVelocity master mix results are produced 30 to 50% faster than traditional *Taq*-based methods. Key to the FullVelocity technology is the unique high-speed archaeal DNA polymerase engineered to excel in rapid, two-step cycling conditions. FullVelocity SYBR® Green master mixes are economical and formulated with two specificity enhancers for high specificity during every cycle.

For a complete list of our QPCR and QRT-PCR reagents, see Table 1.

REAL-TIME QPCR REAGENTS GUIDE

PROBE-BASED DETECTION				SYBR® GREEN DETECTION			
Format	DNA (cDNA) Quantification	RNA Quantification		Format	DNA (cDNA) Quantification	RNA Quantification	
		1-Step	2-Step			1-Step	2-Step
Master Mix	Brilliant® QPCR Master Mix (up to 2 targets) or Brilliant® Multiplex QPCR Master Mix (up to 4 targets)	Brilliant® QRT-PCR Master Mix, 1-Step	Brilliant® QRT-PCR Master Mix, 2-Step	Master Mix	Brilliant® SYBR® Green QPCR Master Mix	Brilliant® SYBR® Green QRT-PCR Master Mix, 1-Step	Brilliant® SYBR® Green QRT-PCR Master Mix, 2-Step
Core Reagent Kit (Standard dNTPs)	Brilliant® QPCR Core Reagent Kit	Brilliant® QRT-PCR Core Reagent Kit, 1-Step	Stratascript® QPCR cDNA Synthesis Kit plus Brilliant® QPCR Core Reagent Kit	Master Mix	FullVelocity™ SYBR® Green QPCR Master Mix	FullVelocity™ SYBR® Green QRT-PCR Master Mix, 1-Step	FullVelocity™ SYBR® Green QRT-PCR Master Mix, 2-Step
Core Reagent Kit (dUTP and/or UNG)	Brilliant® QPCR *Plus* Core Reagent Kit	Brilliant® QRT-PCR *Plus* Core Reagent Kit, 1-Step	Brilliant® QRT-PCR *Plus* Core Reagent Kit, 2-Step	Core Reagent Kit (Standard dNTPs)	Brilliant® SYBR® Green Core Reagent Kit		StrataScript® QPCR cDNA Synthesis Kit plus Brilliant® SYBR® Green Core Reagent Kit

Table 1
CHOOSE THE RIGHT MASTER MIX OR CORE REAGENT DEPENDING ON YOUR APPLICATION.

Rapid and Effective QPCR Education On-Demand

The Fast Track QPCR Education Program is a comprehensive program of training and advanced education for users of our QPCR instruments.

Getting Started with QPCR

Our Introduction to QPCR Guide provides you with a review of the technology and in-depth details on experimental design, assay setup, assay optimization, and data analysis. This guide walks you through a QPCR experiment from start to finish and includes some advanced methods for optimization, troubleshooting, and alternative approaches to quantification.

The Fast Track QPCR Education Program provides a wide range of tools to get you up and running with QPCR quickly and effectively. We offer a wide selection of web seminars, updated Technical and Application Notes, a comprehensive Introduction to QPCR Guide, and regularly scheduled Regional QPCR User Group Meetings. The program also offers you access to expert Field Application Scientists providing advanced education.

To download our Introduction to Quantitative PCR: Methods and Application Guide, review our updated web seminars, or check for the next Regional QPCR User Group Meeting in your area, visit www.stratagene.com/fasttrack.

The Fast Track QPCR web seminars can be found at www.stratagene.com/fasttrackseminars. The web seminars can be viewed in real-time or you can download the recorded version and pdf version of the PowerPoint slides. The list of current Fast Track QPCR Seminars includes:

- Principles of Quantification in QPCR
- QPCR Assay Design Analysis
- QPCR Primer and Probe Design
- QPCR Assay Controls
- QPCR Assay Validation and Optimization
- QPCR Data Analysis
- MxPro™ QPCR Software: Basic Functionality
- MxPro™ QPCR Software: Advanced Functionality

Table 2
Mx3000P® AND Mx3005P™ FEATURES AND SPECIFICATIONS

SAMPLE CAPACITY	Standard 96-well plates, 0.2 ml 8-strip tubes or individual tubes
DIMENSIONS AND WEIGHT	33 cm W x 46 cm D x 43 cm H and 20 kg
SAMPLE VOLUME	10-60 µl
FLUORESCENCE EXCITATION	Quartz tungsten-halogen lamp, 350-750 nm range
FLUORESCENCE DETECTION	Single photomultiplier tube (PMT), 350-700 nm range
FILTERS AVAILABLE	Alexa Fluor® 350, FAM™/SYBR® Green I, TET™, HEX™/JOE™/VIC™, Cy™3, TAMRA™, ROX™/Texas Red®, Cy™5
NUMBER OF OPTICAL CHANNELS	Four (Mx3000P® System) or five (Mx3005P™ System) user-selected excitation/emission filter sets
OPTICAL FILTER MOVEMENT	Simultaneous matched movement of excitation and emission filters; Mx3005P™ System capable of custom filter alignment (e.g., mismatch excitation and emission filters: FAM excitation / ROX emission)
OPTICAL MEASUREMENTS	Fiberoptic scanning head takes measurements at any single plateau or multiple plateaus
THERMAL UNIFORMITY	+/- 0.25°C within 12 seconds at 72°C
THERMAL BLOCK RAMP RATE	Up to 2.5°C/second
TYPICAL RUN TIME	Standard 40 cycle 2-step QPCR reaction completed in 90 minutes; the same reaction is completed in 60 minutes using FullVelocity™ reagents (plus dissociation curve)
MULTI-INSTRUMENT CONTROL	Up to 6 instruments simultaneously from one computer
MULTIPLEX	Up to 4 targets (Mx3000P® System) or five targets (Mx3005P™ System) simultaneously
CHEMISTRIES SUPPORTED	SYBR® Green I, Taqman, molecular beacons, Eclipse probes, Scorpion primers, Lux primers, and Plexor system

Table 3
Mx3000P® and Mx3005P™ SYSTEM FILTER CHOICES

FILTER SETS	EXCITATION WAVELENGTHS / EMISSION WAVELENGTHS
ALEXA Fluor® 350	350 nm / 440 nm
FAM™/SYBR® Green I	492 nm / 516 nm
TET™	517 nm / 538 nm
HEX™/JOE™/VIC™	535 nm / 555 nm
Cy™3	545 nm / 568 nm
TAMRA™	556 nm / 580 nm
ROX™/Texas Red®	585 nm / 610 nm
Cy™5	635 nm / 665 nm

Table 4
QPCR SYSTEMS AND REAGENTS SELECTION GUIDE

APPLICATION	PRODUCT	ADVANTAGES
Single-color to five-color multiplex, 96-well format with powerful data analysis software	Mx3000P® or Mx3005P™ QPCR System	+ Four or five optical channels with user-selected filters for greater flexibility + Broad wavelength range excitation supports most fluorescent dyes + Defined excitation and emission detection wavelengths are ideal for superior multiplex results + Open platform design supports all fluorescent chemistries
High-throughput single-color to five-color multiplex, 96-well x 2 format (192 wells) with powerful data analysis software	Mx3000P® Duet QPCR System or Mx3005P™ Duet QPCR System	+ Run two Mx3000P® or Mx3005P™ QPCR Systems from a single computer + Ideal for higher throughput labs on a budget + Capable of running up to 6 systems from a single computer simultaneously
Multiple instruments with single-color to five-color multiplex on a budget with powerful data analysis software	Mx3000P® / Mx3005P™ Combo QPCR System	+ Run one Mx3000P® system and one Mx3005P™ system from a single computer simultaneously + Ideal for larger labs and core labs requiring a flexible platform to support research applications in the future
Real-time QPCR with significantly shorter run times (DNA, cDNA, and RNA targets)	FullVelocity™ QPCR and QRT-PCR Master Mixes	+ Sensitive one-step QRT-PCR in less time + High speed enzyme supports rapid cycling conditions + Increase throughput
Pre-optimized for sensitive and reproducible real-time quantification (DNA, cDNA, and RNA targets)	Brilliant® QPCR and QRT-PCR Master Mixes	+ Made with optimized buffers and performance tested for reproducible results up to 24 months + Master mix format reduces pipetting steps and minimizes user error + dUTP in nucleotide mixes so UNG can be added for carryover contamination control

Ordering Information

PRODUCT	QUANTITY / FORMAT	CATALOG NO.
QPCR Systems		
Mx3000P™ QPCR System	4-color system (110v) with notebook computer	401403
	4-color system (110v) with desktop computer	401405
	4-color system (230v) with notebook computer	401406
	4-color system (230v) with desktop computer	401407
Mx3005P™ QPCR System	5-color system (110v) with desktop computer	401456
	5-color system (110v) with notebook computer	401449
	5-color system (230v) with desktop computer	401458
	5-color system (230v) with notebook computer	401457
QPCR and QRT-PCR Reagents		
Brilliant® QPCR Master Mix Kits	SYBR® Green-based detection, 400 rxn x 25 µl	600548
	Probe-based detection, 400 rxn x 25 µl	600549
Brilliant® QRT-PCR Master Mix Kits, 1-step	SYBR® Green-based detection, 400 rxn x 25 µl	600552
	Probe-based detection, 400 rxn x 25 µl	600551
Brilliant® QRT-PCR Master Mix, 2-Step	Probe-based detection, 400 rxn x 25 µl	600556
Brilliant® SYBR® Green QRT-PCR Master Mix, 2-Step	SYBR® Green-based detection, 400 rxn x 25 µl	600555
Brilliant® SYBR® Green SideStep™ QPCR Master Mix	SYBR® Green-based detection, 400 rxn x 25 µl	400904
Brilliant® SideStep™ QRT-PCR Master Mix, 1-step	Probe-based detection, 400 rxn x 25 µl	400907
Brilliant® SYBR® Green SideStep™ QRT-PCR Master Mix, 2-Step	SYBR® Green-based detection, 400 rxn x 25 µl	400906
FullVelocity™ SYBR® Green QPCR Master Mix Kits	SYBR® Green-based detection, 400 rxn x 25 µl	600581
FullVelocity™ SYBR® Green QRT-PCR Master Mix Kits, 1-step	SYBR® Green-based detection, 400 rxn x 25 µl	600582
FullVelocity™ SYBR® Green QRT-PCR Master Mix, 2-Step	SYBR® Green-based detection, 400 rxn x 25 µl	600558
StrataScript® QPCR cDNA Synthesis Kit	2-step QRT-PCR, 50 rxn	600554

LEGAL LANGUAGE

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Technical Services: 800-894-1304 x2

Stratagene Europe

Order: 00800-7000-7000
Technical Services: 00800-7400-7400

Stratagene Japan K.K.

Order: 03-5159-2060
Technical Services: 03-5159-2070

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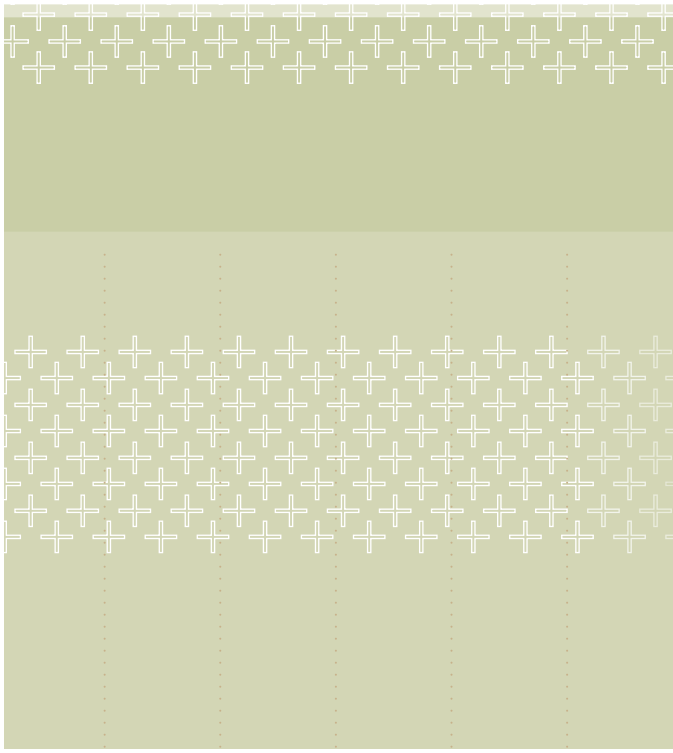

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