

# Real-Time PCR

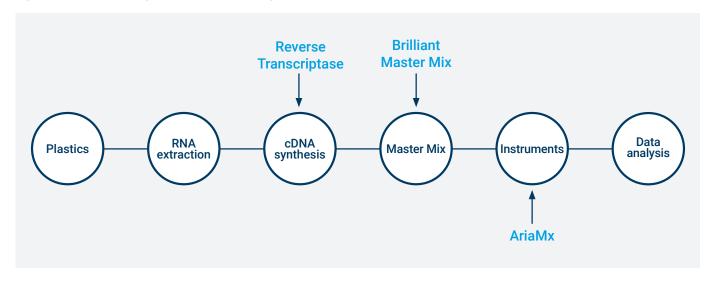
Catalog



### Quantitative PCR & qRT-PCR

Real-time quantitative PCR (qPCR) technology combines DNA, cDNA, or RNA amplification with real-time monitoring of the amplified product in order to calculate the initial quantity of the specific target of interest. We offer a total solutions approach to real-time PCR by simplifying the challenges you face from sample preparation to data analysis and validation. Whether you are new or experienced in qPCR, your individual needs are met with our comprehensive range of products and support. Those getting started in qPCR benefit from web-based training programs, premixed reagent kits, and turnkey instrument installation. More experienced qPCR users appreciate the flexibility of our powerful, user-friendly software as well as reagent kits that support user customization and optimization of even the most demanding assays.

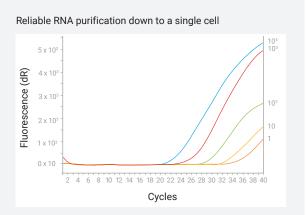
#### Agilent Solutions for qPCR Workflow - Reagents



### Absolutely RNA Purification Kits

- Generate highly pure RNA that is DNA-free
- Maximum RNA yields
- Streamlined 30-minute methods save time
- Application-based testing ensures highest level of quality control

Agilent's Absolutely RNA product portfolio makes purification of DNA-free total RNA from tissue or cell samples easy, even samples from laser microdissection. The proven Absolutely RNA method has no cumbersome steps—no heating, no long centrifugation and no re-purification. The Absolutely RNA kits include all the reagents you need for fast, easy purification of high-quality total RNA including the DNase. DNase I is supplied lyophilized, so the kits can be stored at room temperature, saving valuable freezer space. Each Absolutely RNA kit has been optimized for a specific range of sample sizes and elution volumes. Elution volume is especially critical when purifying RNA from the smallest samples (see Figure 1) in order to provide RNA at a useful working concentration.



**Figure 1.** qRT-PCR results using total RNA template isolated from 10,000, 1,000, 100, 10 and 1 HeLa cells using the Absolutely RNA Nanoprep kit. Duplicate reactions were run for each sample with human GAPDH molecular beacon and primers and 2-µl RNA template. Reactions were performed with an Mx instrument Multiplex Quantitative PCR System.

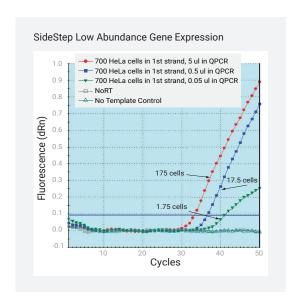
Table 1. Absolutely RNA kits.

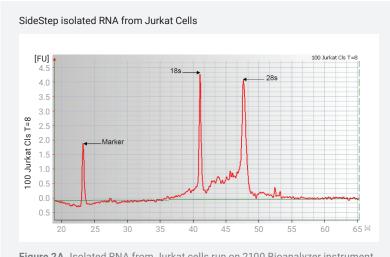
	Contents	Amount	Catalog no.
Absolutely RNA Miniprep kit	Prefilter spin cups, RNA binding spin cups, RNA lysis buffer, ß-mercaptoethanol, DNase (lyophilized), DNase buffers, wash buffers, elution buffer	50 preps	400800
Absolutely RNA Microprep kit	RNA binding spin cups, RNA lysis buffer, ß-mercaptoethanol, DNase (lyophilized), DNase buffers, wash buffers, elution buffer	50 preps	400805
Absolutely RNA 96 Microprep kit	96-well binding plates, 96-well collection plates, RNA binding spin cups, RNA lysis buffer, ß-mercaptoethanol, DNase (lyophilized), DNase buffers, wash buffers, elution buffer, adhesive plate sealer, 96-well storage mat	2 plates	400793
Absolutely RNA Nanoprep kit	RNA binding spin cups, RNA lysis buffer, ß-mercaptoethanol, DNase (lyophilized), DNase buffers, wash buffers, elution buffer	50 preps	400753
Absolutely RNA FFPE kit	Deparaffinization reagents, proteinase K, pre-filter spin cups, RNA binding spin cups, ß-mercaptoethanol, lyophilized DNase, DNase buffers, wash buffers and elution buffer, Real-Time PCR (qPCR) Total RNA (Human)	50 preps	400809
Absolutely RNA FFPE kit w/o deparaffinization	Proteinase K, pre-filter spin cups, RNA binding spin cups, ß-mercaptoethanol, lyophilized DNase, DNase buffers, wash buffers and elution buffer, Real-Time PCR (qPCR) Total RNA (Human)	50 preps	400811

### SideStep II Lysis Kit

- RNA directly from cells without separate RNA purification step (using probe-based detection)
- Save money by purifying only the most relevant samples after screening
- RNA is stabilized for up to 6 months enabling samples to be safely archived
- High throughput applications may be undertaken as the Master Mix enables faster purification
- A wide range of cell inputs can be accommodated bringing excellent analysis flexibility
- Accurate data is enabled as RNA loss minimized
- Enhance safety by the eliminating the production of hazardous waste disposal and removal

SideStep II products deliver efficient cell lysis for sensitive quantitative gene expression analysis without RNA purification. Utilizing our Brilliant qRT-PCR technology, multiple samples can be screened quickly and easily. The one-step process results in RNA stabilized for up to six months.





**Figure 2A.** Isolated RNA from Jurkat cells run on 2100 Bioanalyzer instrument after 8 hours at room temperature.

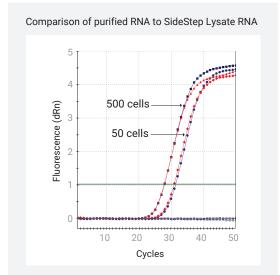


Figure 2B. qRT-PCR of Column Purified RNA from cells stored at -80C and SideStep Lysate Stored at -20C after 6 months. Blue traces, column purification of RNA from cells. Red traces, SideStep lysate. All RNA from HeLa cells. Grey trace, no template control. Green trace, no reverse transcription with column purified RNA. purple trace, no reverse transcriptase with SideStep lysate.

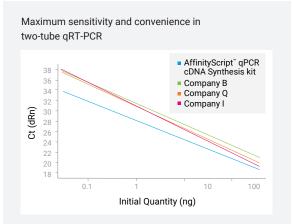
Table 2. SideStep II qRT-PCR Master Mix.

Content	Amount	Catalog no.
SideStep II	400 rxn	400917

### AffinityScript qPCR cDNA Synthesis Kit

- Fast, highly efficient cDNA synthesis for qRT-PCR
- Streamlined protocol produces cDNA in 15 minutes
- Linear detection from 3 pg to 3 μg total RNA
- Master mix format saves time, reduces pipetting variability

Our AffinityScript Multiple Temperature Reverse Transcriptase is engineered to be highly thermostable, allowing you to reverse transcribe at your preferred reaction temperature. The AffinityScript qPCR cDNA Synthesis kit is designed for the highest efficiency conversion of RNA to cDNA and is fully optimized for quantitative PCR applications. Using this kit, you will experience a significant increase in sensitivity over that of competitors' kits. Included in this kit are our qPCR-grade AffinityScript Reverse Transcriptase and a master mix buffer that is optimized for qPCR analysis. This kit employs a fast, easy-to-use 15 minute cDNA synthesis step that allows the generation of cDNA up to 12 kb, with a total reaction time of 25 minutes. This is four times faster than conventional protocols.



**Figure 3.** Our AffinityScript qPCR cDNA Synthesis kit was far more sensitive and linear than competitors' kits, delivering an earlier Ct value across a wider range of RNA input.

Table 3. AffinityScript qPCR cDNA Synthesis kit.

Contents	Amount	Catalog no.
AffinityScript Multiple Temperature Reverse Transcriptase/RNase Block Ribonuclease Inhibitor Enzyme Mix, 2 x cDNA Synthesis Master Mix, oligo(dT) and random primers, RNase free H <sub>2</sub> 0	50 rxn	600559

### AccuScript Hi-Fi cDNA Synthesis Kit

- Proofreading activity reduces errors
- Synthesize cDNA with 3 to 6 -fold fewer errors
- Achieve up to 8-fold better RT-PCR accuracy
- Up to 3 x faster RT-PCR reaction times
- High yields of full length cDNA up to 20 kb

AccuScript High-Fidelity Reverse Transcriptase (RT) delivers the highest reverse transcription accuracy currently available. This MMLV-derived RT generates cDNA with 3 to 6 -fold fewer errors, while promoting full-length cDNA synthesis and superior RT-PCR performance.

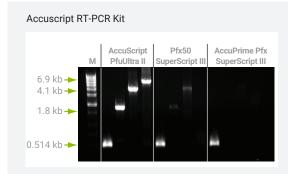


Figure 4. Comparison of Accuscript with High Fidelity PCR RT Combinations from a Leading competitor. Various length cDNAs were amplified from total RNA. PCR reactions were set up using manufacturers' recommended conditions. The different cDNAs amplified over 40 cycles were; .514kb mouse beta-actin, 1.8kb human dystrophin, 4.1kb mouse complement and 6.9kb human apolipoprotein.

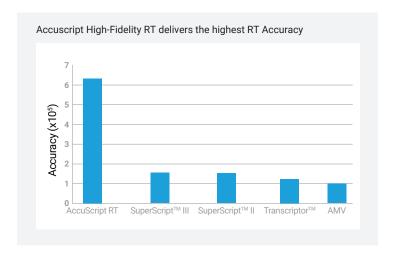


Table 4. AccuScript Hi-Fi cDNA Synthesis kit.

Contents	Amount	Catalog no.
AccuScript high fidelity first strand cDNA synthesis system delivers the highest accuracy and offers you greater	1 kit	200820
flexibility to use the cDNA in your choice of downstream applications such as PCR amplification or real-time PCR		
quantification. Sufficient volume for 50 reactions.		

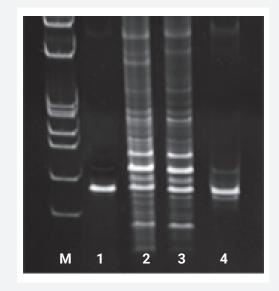
### SureStart Taq DNA Polymerase

- Versatile-can be used for slow or fast hot start activation
- Reliable room-temperature setup
- Use in existing real-time qPCR protocols

#### High specificity HotStart Taq DNA polymerase

SureStart Taq DNA Polymerase is a hotstart version of our Taq2000 DNA Polymerase quality controlled for use in real-time qPCR. This specially modified Taq DNA polymerase allows you to set up PCR reactions at ambient room temperature without the risk of nonspecific primer annealing and extension. The SureStart Taq DNA polymerase can be used in a variety of amplification systems to improve specificity, yield, and detection of low-copy-number targets.

#### SureStart Taq Polymerase Increases Target Specificity



**Figure 5.** A 105 bp fragment of the glucocerebrosidase gene was amplified from human genomic DNA. Lane 1: SureStart Taq DNA Polymerase, Lane 2: unmodified Taq DNA Polymerase, Lane 3: an antibody-based hotstart Taq DNA polymerase, Lane 4: a competitor's modified Taq DNA polymerase.

Table 5. SureStart Taq DNA Polymerase.

Contents	Amount	Catalog no.
SureStart Taq DNA polymerase, 10 x SureStart Taq DNA polymerase buffer	500 U 1000 U	600282 600284

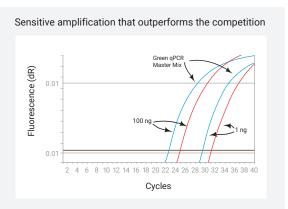
### **Brilliant III SYBR**

# Total reagent solutions for sensitive qPCR of up to four targets

The versatile Brilliant qPCR and qRT-PCR reagents provide a highly sensitive solution for real-time PCR detection and gene quantitation. Agilent's broad Brilliant product portfolio has the perfect kit for you no matter what your experience level, novice to expert, with a choice of convenient Taq-based master mixes or core reagent kits that allow assay optimization.

# Highly sensitive detection of DNA or RNA using SYBR Green Dye

The Brilliant SYBR Green qPCR and qRT-PCR reagents provide a universal solution to real-time qPCR detection and gene quantification and exhibit greater sensitivity compared to other SYBR Green kits. SYBR Green dye binds to any PCR product, and therefore does not require the use of sequence-specific probes. All Brilliant reagent kits contain SureStart Taq DNA Polymerase, a hot-start version of Taq that minimizes amplification of non-specific PCR products.



**Figure 6.** The Brilliant SYBR Green qRT-PCR Master Mix outperforms competitor A in qRT-PCR using 100 ng and 1.0 ng of a TATA Box Binding protein mRNA target. Similar performance advantage is observed using other targets of various sizes. (Blue: Agilent, Brilliant SYBR Green Master Mix; Red: Competitor A SYBR Green Master Mix)

#### Sensitive and specific probe-based detection

Brilliant probe-based qPCR and qRT-PCR reagents are compatible with sequence-specific probes including TaqMan probes, Molecular beacons, and Scorpions. These reagents offer a wide linear dynamic range of amplification. The qRT-PCR kits are available in one-step and two-step formats. A passive reference dye is included separate from the buffer solution for versatility and to maximize performance on different instrument platforms.

Table 6. Brilliant III SYBR MM and Brilliant III SYBR MM with ROX.

	Contents	Amount	Catalog no.
Brilliant III Ultra-Fast	2 x master mix with ROX provided in separate tube. Uses 20 μl/reaction	400 rxn	600882
SYBR Green qPCR Master Mix		(20 µl/rxn) 10 x 400 rxn (20 µl/rxn)	600883
Brilliant III Ultra-Fast SYBR Green qRT-PCR	RT module containing MMLV Reverse Transcriptase and RNase Block, 2 x qPCR master mix with ROX in separate tube Uses 20 µl/reaction	400 rxn (20 μl/rxn)	600886
Master Mix		10 x 400 rxn (20 μl/rxn)	600887
Brilliant III Ultra-Fast	High performance, ultra-sensitive, SYBR Green qPCR master mix reagent	400 rxn	600889
SYBR Green High ROX qPCR Master Mix	with high ROX concentration for reliable quantification across a wide range of targets and templates	10 pack	600904
Brilliant III Ultra-Fast	High performance, ultra-sensitive, SYBR Green qPCR master mix reagent	400 reactions	600892
SYBR Green Low ROX qPCR Master Mix	with low ROX concentration for reliable quantification across a wide range of targets and templates	10 pack	600903

### **Brilliant III Probe**

- Extremely fast while maintaining sensitivity
- Greater resistance to common qPCR inhibitors (i.e. whole blood or NaCl)
- Optimized fast cycling formulation ensures reliable and reproducible data with shorter run times
- Convenient pre-blended formulations compatible with any sequence-specific probe detection chemistry

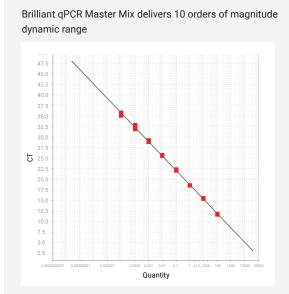


Figure 7. Brilliant III Ultra-Fast RT-qPCR probe on StepOnePlus instrument; GAPD; Human cDNA ranging from 100 ng - 0.01 pg/rxn. Brilliant III displays high efficiency and RSq values (BIII, 96.9 % and 0.999).

Table 7. Brilliant III Probe MM and Brilliant III Probe MM with ROX.

	Contents	Amount	Catalog no.
Brilliant III Ultra-Fast qPCR Master Mix	Designed for researchers who want access to their data faster without compromising data quality	400 rxn (20 μl/rxn)	600880
		10 x 400 rxn (20 μl/rxn)	600881
Brilliant III Ultra-Fast qRT-PCR Master Mix	Designed for researchers who want access to their data faster without compromising data quality	400 rxn (20 μl/rxn)	600884
		10 x 400 rxn (20 μl/rxn)	600885
Brilliant III Ultra-Fast	High performance, ultra-sensitive, probe qPCR master mix reagent with High	400 rxn	600888
Probe High ROX qPCR Master Mix	ROX concentration for reliable quantification across a wide range of targets and templates	10 x 400 rxn	600899
Brilliant III Ultra-Fast	High performance, ultra-sensitive, probe qPCR master mix reagent with low	400 rxn	600890
Probe Low ROX qPCR Master Mix	ROX concentration for reliable quantification across a wide range of targets and templates	10 x 400 rxn	600898

## Brilliant HRM Ultra-Fast Loci Master Mix Reagent

#### User applications-

- DNA methylation
- Heterozygosity screening
- Genotyping
- Viral/bacterial population diversity
- HLA compatibility testing
- Species identification

#### High resolution melt application

For the scientist seeking to "mix and go" faster and with greater confidence, we offer the Brilliant HRM Ultra-Fast Loci Master Mix. The Master Mix combines a mutant Fast-Start Taq polymerase, optimized MgCl2, dNTPs, and an EvaGreen, release-on-demand dye to provide faster HRM with total confidence — even for difficult genotypes. It is validated for use on the AriaMx Real-Time PCR instrument and third-party HRM-capable thermal cyclers.



**Figure 8.** Agilent developed an HRM assay to resolve a Class IV SNP (A/T), Rs9939609 FTO (142 bp fragments). Performed on AriaMx instrument.

Table 8. Brilliant HRM Ultra-Fast Loci Master Mix.

	Description	Amount	Catalog no.
Brilliant HRM Ultra-Fast Loci Master Mix	Brilliant HRM ultra-fast Loci master mix is for high-resolution melt (HRM) analysis	200 rxn (2 ML)	5190-7827

### Brilliant Multiplex qPCR Master Mix

- Maximizes analysis of limited or rare samples
- Allows detection of multiple targets plus an internal control
- More economical per sample than singleplex, while saving time and increasing throughput

# Simultaneous amplification of targets and control genes

The Brilliant Multiplex qPCR Master Mix allows you to amplify up to four targets in a single real-time PCR reaction (see Figure 9). The Brilliant multiplex qPCR master mix provides sufficient reaction components to accurately quantify both low and high abundance targets in the same tube. This allows you to more successfully multiplex without concern for bias due to abundance level. Importantly, the sensitivity remains equivalent to that seen in singleplex reactions.

### Equivalent performance with four target multiplex using Brilliant Multiplex qPCR Master Mix

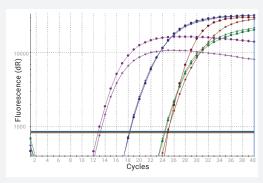


Figure 9. We amplified four targets—eNOS (FAM), HFE (HEX), CFTR (ROX), and Cyclophilin (Cy5) — in singleplex and multiplex using the Brilliant Multiplex qPCR Master Mix on our qPCR System. Ct values for each target were virtually identical in the two reactions, indicating full sensitivity and performance in multiplex. Like colored lines correspond to single and multiplex reactions for the same target.

Table 9. Brilliant probe-based quantitative PCR reagents.

	Contents	Amount	Catalog no.
Brilliant Multiplex qPCR Master Mix	2 x Brilliant Multiplex Master Mix, passive reference dye	200 rxn (25 μl/rxn)	600553

# qPCR NGS Library Quantification Kit for Illumina Systems

- Provides researchers with an accurate and sensitive method for quantifying NGS libraries
- Validated in the SureSelect Target Enrichment Protocol for barcoding and indexing applications
- 1fM sensitivity

#### Quantify qPCR NGS libraries with great accuracy

The Agilent qPCR NGS Library Quantification Kit provides researchers an accurate and sensitive method for quantifying NGS libraries. Accurate library quantification leads to optimal cluster densities for improved sequence efficiency and data quality. Consistent quantification across a broad range of samples, varying library insert sizes, and GC content. Quantify 84 libraries with each kit.

### High-specificity miRNA qRT-PCR Detection kit

- Detects mature miRNA
- Differentiate between miRNA that differ by a single nucleotide
- Sensitive detection down to 10 copies
- Highly accurate results in 3 hours
- miRNA Specific Forward Primers

#### A novel PCR enzyme formulation and qPCR detection reagents for the utmost specificity

The Agilent High-Specificity miRNA QRT-PCR Detection kits provide qualified reagents to polyadenylate microRNAs (miRNAs) followed by synthesis of first-strand cDNA from these tailed miRNAs. A novel PCR enzyme formulation and qPCR detection reagents give utmost specificity.

The kits detect mature miRNA from as little as 15 ng of total RNA input on various sample types.

### Mycosensor Detection Kit

- Specific detection of the eight most common Mycoplasma species
- Rapid results in under two hours
- Detect as few as 50 copies of Mycoplasma genomic DNA

Agilent's MycoSensor qPCR assay kit detects *Mycoplasma* contamination by real-time quantitative PCR utilizing SYBR Green dye detection. The convenient master mix format provides all the reagents needed for amplification and fluorescence detection. The kit includes two positive control templates to validate the detection of polymerase-mediated amplification of *Mycoplasma* DNA and confirm the SYBR Green dissociation profile for your test samples. To minimize false positives, the closed tube real-time detection format minimizes the potential for cross-contamination with PCR amplicon. Agilent includes a passive reference dye in a separate tube with the kit to allow you to run the assay on most real-time qPCR platforms. The kit also contains the DNA purification reagents for removal of potential PCR inhibitors in cell culture supernatants and cell pellets.

#### 

**Figure 10.** The MycoSensor qPCR assay kit detects a range from 100,000 copies to 10 copies of M.orale genomic DNA in the presence of HeLa cell culture supernatant. The reactions were done in duplicate on a Mx qPCR System and the amplification plots are shown here. Blue, 100,000 copies. Dark Orange, 10,000 copies. Green, 1,000 copies. Grey, 100 copies. Light Orange, 10 copies.

Table 10. MycoSensor qPCR assay kit.

Contents	Amount	Catalog no.
2 x MycoSensor qPCR Master Mix, MycoSensor Primer Mix, amplification control, M. orale positive control,	50 rxn	302107
A. laidlawii positive control, reference dye, DNA purification kit	100 rxn	302106

#### Porcine Detection Kit

- Multiplex detection of porcine specific DNA and Alien DNA control
- DNA Isolation Module, with spin cup protocol
- qPCR assay to detect nucleic acids of Porcine origin
- No interference from inhibitor

#### Detect two distinct targets in a single reaction

The Porcine Detection Kit contains reagents and materials for the extraction and isolation of DNA from food samples and other materials, including gel caps, as well as subsequent qPCR amplification and detection of porcine DNA down to 300 fg. Isolated DNA may contain contaminants that inhibit PCR, therefore, the qPCR kit amplifies and detects two distinct targets in a single reaction: a porcine-specific DNA sequence and an external DNA control that enables detection of PCR inhibition.

### qPCR Inhibitor Detection kit with Alien RNA

- External control for detecting inhibitors in RNA samples
- Highly sensitive to various inhibitors
- Known copy number provided
- Ideally suited for assay standardization applications

Agilent's Alien qRT-PCR Inhibitor Alert is a useful tool in determining the quality of different RNA samples when studying gene expression levels with samples obtained from various sources. There are a variety of inhibitors that can affect the efficiency of qRT-PCR reactions and may be co-purified with RNA samples, depending on the source of starting material, the methods of extraction, etc. The amplification of Alien RNA is highly sensitive to a number of common qRT-PCR inhibitors such as phenol, ethanol, guanidine, and EDTA. A known amount of Alien RNA is amplified in the presence of an RNA sample of interest using the Alien primer mix. An increase in the threshold cycle (Ct value) for amplification of the Alien RNA in the sample compared with the Alien RNA alone will be an indicator of the presence of inhibitory substances in the sample (see Figure 11).

#### Assay standardization

The Alien qRT-PCR inhibitor alert is ideally suited for assay standardization applications, and displays a dynamic range of eight orders of magnitude. Using the Alien qRT-PCR inhibitor alert as a reference control to generate standard curves allows data comparisons from multiple experiments, across

Amplifications of Alien RNA and GAPDH are inhibited in the presence of 10 µM Guanidine Fluorescence (dR) 0.7 0.5 0.4 0.2 0.2 4 6 8 10 12 14 16 18 20 22 24 26 28 30 32 34 36 38 40 Cycles В 0.6 Fluorescence (dR) No Guanidine 0.4 10 µM Guanidine 0.3 NTC 4 6 8 10 12 14 16 18 20 22 24 26 28 30 32 34 36 38 40 Cycles

Figure 11. Amplifications were performed in the presence or absence of 10  $\mu$ M guanidine. Reactions contained 100,000 copies of Alien RNA, 50 ng Universal Human Reference RNA, 100 nM Alien primer mix (A), or GAPDH primers (B) using our Brilliant SYBR Green qRT-PCR Master Mix, 1-Step.

platforms, and between laboratories. The Alien RNA is produced in large lots and subject to stringent quality-control measures to ensure the availability of consistent reference RNA material over long-term experimental studies. Since Alien RNA has no significant homology to known sequences, it is a valuable tool as external standardization for real-time PCR experiments.

#### Use Alien Control with Brilliant SYBR Green qRT-PCR Master Mix, 1-Step & 2-Step

The Alien qRT-PCR inhibitor alert can be used to detect inhibitors in both one-step (single-tube) and two-step (two-tube) qRT-PCR assays that employ SYBR Green dye for detection.

Table 11. Alien qRT-PCR Inhibitor Alert.

	Description	Amount	Catalog no.
Alien qRT-PCR Inhibitor Alert	External control for detecting inhibitors in RNA samples for qRT-PCR analysis	400 rxn	300600
Alien control for use with Brilliant SYBR Green qRT-PCR Master Mix, 2-Step	Delivers sensitive detection of RNA, with high quality Alien RNA control	400 rxn	300602

### qPCR Instrument & Software

- Easy touchscreen set-up
- Plate maps at your fingertips
- Onboard diagnostics and remote monitoring

# Confidence in a system that meets your needs today and tomorrow

The AriaMx Real-Time PCR System is a fully integrated qPCR amplification, detection, and data analysis system. The system's modular design combines a state-of-the-art thermal cycler, an advanced optical system with spectra-optimized LED cartridges, and data analysis software.

The instrument leverages a comprehensive software suite of on-board instrument diagnostics, giving you confidence that instrument failpoints are identified prior to running your assay. Experience total confidence with AriaMx's blend of speed, agility, and precision.



Figure 12. Easy-to-access plate and well maps.

### AriaMx Optical Cartridges

The AriaMx Real-Time PCR System is a fully integrated quantitative PCR instrument that can hold up to six optics modules. This future-proof design of the instrument allows the accommodation of as many or as few optic channels as required.

Table 12. AriaMx optical cartridges.

Product	Amount	Catalog no.
SYBR/FAM optical cartridge	1 pack	G8830-67001 (Option 101)
ROX optical cartridge	1 pack	G8830-67002 (Option 102)
HEX optical cartridge	1 pack	G8830-67003 (Option 103)
CY3 optical cartridge	1 pack	G8830-67004 (Option 104)
CY5 optical cartridge	1 pack	G8830-67005 (Option 105)
Atto425 optical cartridge	1 pack	G8830-67006 (Option 106)

#### **Explore Agilent cannabis applications and solutions**

Cannabis microbial testing is challenging, due to the variety of administration methods. While culture-based methods have long been used, there are a substantial number of microbial species that cannot be cultured. Molecular methods, such as qPCR, detect unculturable organisms as well as organisms that clump and distort during plating. These organisms include heterogeneous microcolonies that can occur with various aspergillus species. Learn more how Agilent has paired our qPCR instrument with partner assays to address the unique challenges in cannabis microbial testing on https://www.agilent.com/en/promotions/cannabis

Agilent products and solutions are intended to be used for cannabis quality control and safety testing in laboratories where such use is permitted under state/country law.



### qPCR Accessories

- The perfect fit frame, tubes and caps are validated for best experimental results on Agilent's platform can be found using plastics reccomended below.
- 96-Well Semi-skirted Polypropylene PCR plates validated for optimal performance and compatible with the optical Strip Caps.
- StrataCoolers LP Benchtop cooler provides the most protection for your enzymes whether in your freezer or on your benchtop: it maintains - 15 °C for at least 2 hours, includes adapters for use with 0.5 ml tubes and eliminates ice build-up and potential contamination.

The qPCR Plastics and accessories have been validated for optimal performance with the AriaMx Real-Time PCR Systems.

Table 13. qPCR plastics & accessories.

Product	Amount	Catalog no.
AriaMx SYBR Green Starter Pack	1 pack	600906
AriaMx qRT-PCR Starter Pack	1 pack	600907
Brilliant HRM Ultra fast Starter Pack	1 pack	5190-9370
AriaMx 96 well plates, skirted and low profile	1 x 25/pack	401490
AriaMx 96 well plates, skirted and rigid	1 x 25/pack	401491
Agilent 96 well plates, non skirted and low profile	1 x 25/pack	401494
AriaMx adhesive plate seals	1 x 50/pack	401492
AriaMx low profile strip tubes for PCR and qPCR applications, without caps	8/strip x 120/box	401493
Polypropylene 96-Well Tube Plates	1 pack	410088
qPCR 96-Well Plates, Non-Skirted	1 pack	401333
qPCR 96-Well Plates, Semi-Skirted	1 pack	401334
StrataCooler LP Benchtop Cooler, Blue	1 item	401349
Tube-Strip Capping Tool	2 tools	410099
Strip caps for PCR and qPCR applications	8/strip x 120/box	401425
Benchtop Rack for 200 μl Tubes/V Bottom Plates	1 rack	410094

#### Quantitative PCR & qRT-PCR

#### **MVP Human Total RNA**

- Purified Total and Poly(A)+ RNA available from a variety of species and tissues
- Extensive quality control ensures high quality Poly(A) + RNA
- Eliminates tedious, time consuming RNA isolation procedures
- Application ready for real-time RT-PCR, miRNA detection, and Northern blot analysis
- Small, economically priced in convenient 25-µg pack sizes

Our MVP (Maximum Value and Purity) RNA product line, offers you high quality, pure, application-ready total RNA in small economical pack sizes. The extensive and rigorous quality control, provides you with valuable assurance that our total RNA is intact, full-length and DNA-free. This makes it ideal for cDNA synthesis and for sensitive qRT-PCR assays. Moreover, we have extensively validated that our isolation method efficiently co-purifies mRNA and small miRNA.

Table 14.

Contents	Amount	Catalog no.
Total RNA, Placenta, Human	25 μg	540025

### Quantitative PCR Mouse Reference Total RNA

- High-quality total RNA control for quantitative PCR gene expression analysis
- No detectable levels of genomic DNA
- Maximum representation of low, medium, and high abundant gene transcripts
- Ideal for use as a template during assay optimizations

The Quantitative PCR Mouse Reference Total RNA is a collection of RNA pooled from 11 mouse cell lines which are also derived from different tissues. We choose cell lines, rather than tissues as starting material since this is the most consistent and highest quality source of RNA. Our mouse reference total RNA provides you with a single, common control and enables you to compare data sets from multiple experiments and between laboratories. The cell lines are grown at industrial scales to produce extremely large lots, which undergo stringent quality-control procedures to address lot-to-lot variability. This guarantees that you can use our reference RNA in multiple experiments carried out over long periods of time. Human reference total RNA also available (Figure 13).

Table 15. qPCR Reference Total RNA, Human and Mouse.

	Description	Amount	Catalog no.
Quantitative PCR Human Reference Total RNA	<ul> <li>Pool of 10 human cell lines providing broad gene coverage, qualified for use in qPCR</li> </ul>	25 μg	750500
Quantitative PCR Mouse Reference Total RNA	<ul> <li>Pool of 11 mouse cell lines providing broad gene coverage, qualified for use in qPCR</li> </ul>	25 μg	750600

### Quantitative PCR Human Reference Total RNA

- High-quality total RNA control for quantitative PCR gene expression analysis
- No detectable levels of genomic DNA
- A consistent reference for cross-platform data set comparisons
- Ideal for use as a template during assay optimization

Ideal for detecting high, medium, and low abundant targets in qRT-PCR

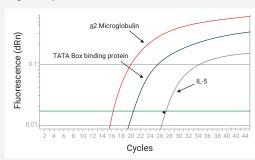


Figure 13. We amplified and detected high, medium, and low abundant targets: beta2 microglobulin (red curve), TATA Box binding protein (blue curve), and IL-5 (black curve). 1  $\mu g$  of Quantitative PCR Human Reference Total RNA was added as template to all reactions. qRT-PCR reactions were prepared using the Brilliant qRT-PCR Master Mix, 1-Step and TaqMan probes. The real-time fluorescence data were analyzed on a Mx Multiplex qPCR System.

Table 16. Product overview.

Nucleic Acid Isolation		
Application	Product	Advantages
Purification of high-quality RNA from cells or tissues	Absolutely RNA Purification kits	<ul> <li>RNA is DNA-free for use in qRT-PCR</li> <li>Three versions: Miniprep, Microprep, and Nanoprep</li> <li>96-well format available</li> <li>Rapid and streamlined protocol</li> </ul>
Reverse Transcription (cDNA synthesi	s) & RT and RT-PCR	
Application	Product	Advantages
cDNA synthesis for qRT-PCR	AffinityScript qPCR cDNA Synthesis kit	<ul> <li>Fast, highly efficient cDNA synthesis for qRT-PCR</li> <li>Streamlined protocol produces cDNA in 15 minutes</li> <li>Linear detection from 3 pg to 3 µg total RNA</li> <li>Master mix format saves time, reduces pipetting variability</li> </ul>
	AccuScript Hi-Fi cDNA Synthesis kit	<ul> <li>Proofreading activity reduces errors</li> <li>Synthesize cDNA with 3 to 6-fold fewer errors</li> <li>Achieve up to 8-fold better RT-PCR accuracy</li> <li>Up to 3 x faster RT-PCR reaction times</li> <li>High yields of full length cDNA up to 20 kb</li> </ul>
qPCR with improved specificity	SureStart Taq DNA Polymerase	<ul> <li>Hotstart formulation of Taq DNA polymerase, qPCR-grade</li> <li>Reduce nonspecific background</li> </ul>
qPCR - Master Mix		
Application	Product	Advantages
Sensitive qPCR and qRT-PCR using SYBR Green detection	Brilliant SYBR Green qPCR and qRT-PCR reagents	<ul> <li>Excellent sensitivity and reproducibility</li> <li>Master mix and core reagent formats available</li> <li>Both one-step and two-step qRT-PCR formats available</li> </ul>
Sensitive qPCR and qRT-PCR using probe-based detection	Brilliant qPCR and qRT-PCR Master Mixes	<ul> <li>Made with optimized buffers and performance tested for reproducible results up to 24 months</li> <li>Reduces pipetting step and increases throughput</li> <li>dUTP in nucleotide mixes so that UNG can be added for carry-over contamination control</li> </ul>
Multiplex qPCR	Brilliant HRM Ultra-Fast Loci Master Mix Reagent	<ul> <li>Stable after multiple freeze thaws, reducing wastage and increasing reliability</li> </ul>
	Brilliant Multiplex qPCR Master Mix	- Multiplex up to four reactions in a single tube
qPCR Instrument & Software		
Application	Product	Advantages
Quantitative PCR amplification, detection, and data analysis	AriaMx	<ul> <li>Unique modular and flexible design</li> <li>Intuitive touch-screen interface</li> <li>Advanced, easy-to-use reporting</li> </ul>
Hight Resolution Melt (HRM) Analysis	AriaMx HRM qPCR Software	<ul> <li>Intuitive software operation and system calibration brings HRM capabilities to every laboratory</li> </ul>
21 CFR Part 11 enabled software	AriaMx ET Software	<ul> <li>Secure application login, database file management, electronic audit tra and report generation</li> </ul>
qPCR accessories		
Application	Product	Advantages
Validated for optimal performance with the AriaMx real time PCR Systems	AriaMx qPCR Plastics	<ul> <li>The perfect fit frame, tubes and caps are validated for best experimenta results on Agilent's platform can be found using plastics recommended below</li> </ul>
Quantitative and qualitative gene expression analysis, miRNA analysis, genetic mapping, genetic fingerprinting, NGS library quantification, 2-6 channel multiplex ability, pathogen quantification	AriaMx Optical Modules	<ul> <li>AriaMx can hold up to six optical modules to accommodate a variety of qPCR applications</li> </ul>

#### Quantitative PCR & qRT-PCR

Table 16. Product overview continued...

Specialty kits & other reagents			
Application	Product	Advantages	
qPCR detection of Mycoplasma contamination in cell cultures	MycoSensor qPCR assay kit	<ul> <li>Specific detection of the eight most common Mycoplasma species</li> <li>Results in less than 2 hours</li> <li>Detect as few as 50 copies</li> </ul>	
Assay optimization and data comparison across qPCR experiments, platforms, and laboratories	Real-Time PCR (qPCR) Total RNA, Human and Mouse	<ul> <li>High-quality pool of DNA-free total RNA to use as template for qRT-PCR experiments</li> <li>Produced from 10 cell lines (human) or 11 cell lines (mouse) for maximum representation</li> <li>Manufactured in large lot sizes to ensure lot-to-lot consistency demanded of this sensitive assay</li> </ul>	
Detection of qRT-PCR inhibitors in RNA samples, and can also act as external RNA control	Alien qRT-PCR Inhibitor Alert	<ul> <li>More reliable qRT-PCR data</li> <li>Sensitive to most common inhibitors</li> <li>Ideal reference tool for assay standardization</li> </ul>	
Gene expression analysis via qRT- PCR, microarray, Northern blotting, and RT-PCR	MVP Total and Poly(A)+ RNA from Human, Mouse, and Rat	<ul> <li>High-quality, pure RNA, mRNA, and cDNA</li> <li>Eliminates tedious, time-consuming RNA isolation and cDNA synthesis procedures</li> <li>Highest level of quality control</li> <li>Well-documented human donor and tissue pathology information</li> </ul>	
Efficient cell lysis, RNA stabilization, and sensitive quantitative gene expression analysis without RNA purification	SideStep II qRT-PCR products	<ul> <li>Single-tube format</li> <li>RNA stabilization for at least 6 months @ -20°C</li> <li>Samples are ready for qRT-PCR in 10 minutes</li> <li>Prepare lysates from a few cells or up to 1 million cells</li> </ul>	

**Table 17.** Real-Time qPCR and qRT-PCR reagents guide.

SYBR Green detection				
Format	DNA (cDNA)	RNA Quantification		
	Quantification	1-Step	2-Step	
Master Mix	Brilliant SYBR Green qPCR Master Mix	Brilliant SYBR Green qRT-PCR Master Mix, 1-Step	Brilliant SYBR Green qRT-PCR, AffinityScript Two-Step Master Mix	
Core Reagent kit (Standard dNTPs)	Brilliant SYBR Green Core Reagent kit		AffinityScript qPCR cDNA Synthesis kit plus Brilliant SYBR Green Core Reagent kit	

Probe-Based detection				
Format	DNA (cDNA) Quantification	RNA Quantification		
romat		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 II qRT-PCR, AffinityScript Two-Step Master Mix	
Core Reagent kit (Standard dNTPs)	Brilliant qPCR Core Reagent kit	Brilliant qRT-PCR Core Reagent kit, 1-Step	AffinityScript qPCR cDNA Synthesis kit plus Brilliant qPCR Core Reagent kit	

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