Now includes a lyophilized Luna RT-qPCR Mix

Luna® Universal qPCR & RT-qPCR

LIGHTING THE WAY™

FREE
Next Day
Delivery
(UK mainland)

NEW ENGLAND
Biolabs®

Lighting the Way

Fluorescence-based quantitative polymerase chain reaction (qPCR) is the gold standard for the detection and quantification of nucleic acids due to its sensitivity and specificity. Luna products are optimized for qPCR or RT-qPCR and are available for either intercalating dye or probe-based detection methods.

Each Hot Start *Taq*-based Luna qPCR master mix has been formulated with a unique passive reference dye that is compatible across a wide variety of instrument platforms, including those that require a ROX reference signal. The mixes also contain dUTP, enabling carryover prevention when reactions are treated with Antarctic Thermolabile UDG (NEB #M0372). The Luna Probe One-Step RT-qPCR 4X Mix with UDG (NEB #M3019) includes both components.

LunaScript® RT SuperMix (NEB #E3010/M3010) is an optimized master mix for fast (13 min) and robust first strand cDNA synthesis in amplicon sequencing or a two-step RT-qPCR workflow.

The Luna Cell Ready Lysis Module and kits are designed for direct RNA quantitation from cell lysate, bypassing traditional RNA extraction and purification steps. Coordinated cell lysis, RNA release, and genomic DNA removal is achieved in a 15 min protocol.

Luna products feature a blue tracking dye

Visible tracking dye
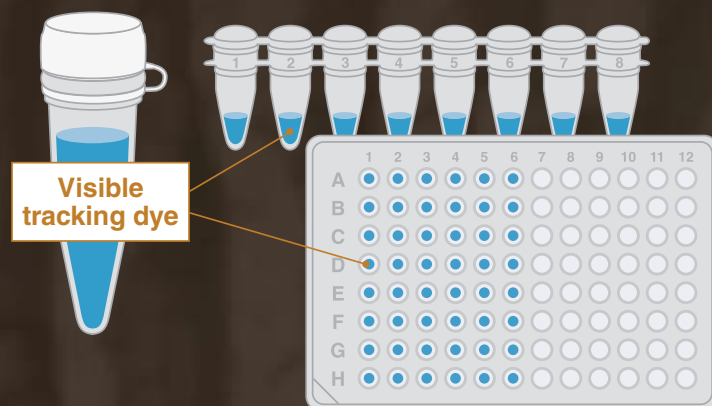
Make a simpler choice

- Non-interfering, visible tracking dye eliminates pipetting errors

- Products are formulated with a unique passive reference dye that is compatible across a wide variety of thermal cyclers, regardless of ROX requirements

Optimize your One-Step RT-qPCR

- Luna WarmStart RT paired with Hot Start *Taq* enables room temperature setup and stability

- 4X option (NEB #M3019) allows for more sample input, increasing sensitiivity

- Available in a lyophilized format: LyoPrime Luna® Probe One-Step RT-qPCR Mix with UDG (NEB #L4001)

Speed up your cDNA Synthesis

- LunaScript RT SuperMix is ideal for first strand cDNA synthesis with a fast 13-minute protocol

- Primer-free option (NEB #E3025) enables flexibility of primers used for optimal cDNA synthesis

Learn more at LUNAqPCR.com

Find the right Luna product for your application.

2 Select your detection method

	Dye-based	Probe-based
Genomic DNA or cDNA	Luna® Universal qPCR Master Mix (NEB #M3003)	Luna Universal Probe qPCR Master Mix (NEB #M3004)*
Purified RNA **One-Step RT-qPCR**	Luna Universal One-Step RT-qPCR Kit (NEB #E3005)	Luna Universal Probe One-Step RT-qPCR: • Kit (NEB #E3006)* • 4X Mix with UDG (NEB #M3019)* • LyoPrime Luna® Probe One-Step RT-qPCR Mix with UDG (NEB #L4001)
Two-Step RT-qPCR	LunaScript® RT SuperMix (NEB #E3010/M3010) **+** Luna Universal qPCR Master Mix (NEB #M3003)	LunaScript RT SuperMix (NEB #E3010/M3010) **+** Luna Universal Probe qPCR Master Mix (NEB #M3004)
RNA from cell lysate	Luna Cell Ready One-Step RT-qPCR Kit (NEB #E3030)	Luna Cell Ready Probe One-Step RT-qPCR Kit (NEB #E3031)

1 Select your target

*No ROX version available (OEM)

For large volume or custom options, contact us at **www.neb.com/CustomContactForm**

One-Step RT-qPCR vs. Two-Step RT-qPCR

Which method should I choose?

Two methods are available for quantifying RNA samples: one-step and two-step RT-qPCR. In both cases, RNA is first reverse transcribed into cDNA, which is then used as the template for qPCR amplification. Reverse transcription can be performed separately from qPCR (i.e., two-step RT-qPCR) or directly in the qPCR mix (i.e., one-step RT-qPCR). One-step workflows are commonly favored in molecular diagnostic assays, where sample inputs may be limiting or numerous samples are examined. Two-step RT-qPCR is preferred when multiple interrogations will be made of the same starting material or where archiving of cDNA may be required.
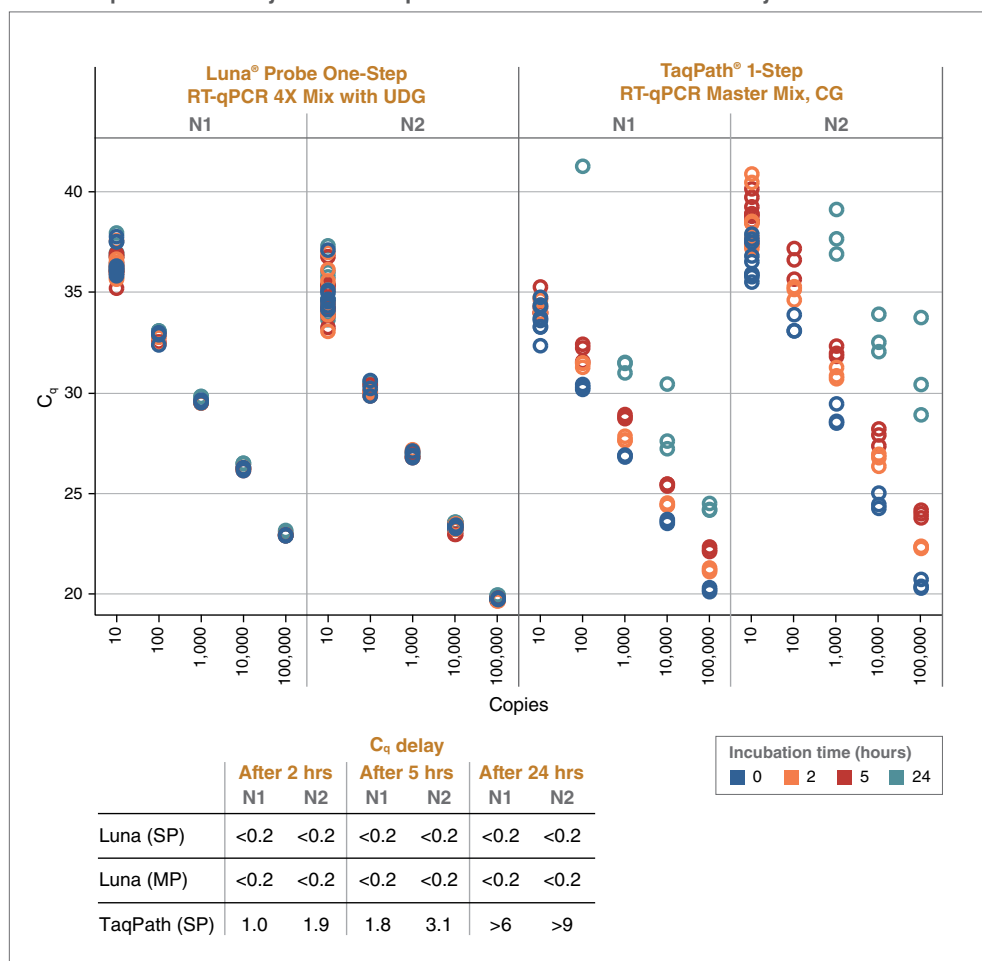
Comparison of One-Step and Two-Step RT-qPCR

Product Name	RT Primers	Advantages	Disadvantages	Ideal Uses
One-Step RT-qPCR	• Gene -specific primers	• Quick setup and limited hands-on time • Single closed-tube reaction, reducing contamination	• Need fresh RNA sample(s) to analyze new targets or repeat experiments	• Assessing many RNA samples • High-throughput applications
Two-Step RT-qPCR	• Oligo(dT) primers • Random hexamer primers • Gene-specific primers • A combination of the above	• Choice of RT primers • Flexible reaction optimization (RNA input, choice of enzyme(s), enzyme amount, & reaction	• More setup and hands-on time • Greater variation and risk of contamination due to extra open-tube step and pipetting	• Assessing multiple targets from few RNA samples • Saving cDNA product for future re-use

What is Luna WarmStart Reverse Transcriptase?

"WarmStart" is the term we use to describe a mesophilic enzyme that is inactive at room temperature and becomes active when the reaction is warmed above approximately 40°C. This feature is conferred by the use of aptamers: engineered oligonucleotides that bind to a specific target molecule through non-covalent interactions. Luna WarmStart Reverse Transcriptase activity is inhibited at room temperature, enabling a flexible reaction setup, and the aptamer is released as the temperature is increased in typical RT-qPCR cycling. It is through the use of these aptamers that NEB has been able to identify and prevent non-specific amplification that can occur in certain settings, such as challenging assays or workflows that include a delay between reaction setup and intended initiation.

Room temperature stability of Luna RT-qPCR Mix enables workflow flexibility

|	**After 2 hrs**		**After 5 hrs**		**After 24 hrs**	
C_q delay	N1	N2	N1	N2	N1	N2
Luna (SP)	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2
Luna (MP)	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2
TaqPath (SP)	1.0	1.9	1.8	3.1	>6	>9

Singleplex (SP) and multiplex (MP) RT-qPCR targeting 2019-nCoV_N1 (HEX) and 2019-nCoV_N2 (FAM) was performed using the Luna Probe One-Step RT-qPCR 4X Mix with UDG, according to reaction and cycling conditions provided in the NEB #E3019 product manual. Singleplex RT-qPCR targeting 2019-nCoV_N1 (FAM) and 2019-nCoV_N2 (FAM) was performed using TaqPath 1-Step RT-qPCR Master Mix, CG, according to the CDC 2019-Novel Coronavirus (2019-nCoV) Real-Time RT-PCR Diagnostic Panel guidelines. Performance was evaluated over a 5-log range (100,000-10 copies) of Synthetic SARS-CoV-2 RNA Control 2 diluted in 10 ng of Jurkat total RNA. RT-qPCR reactions were incubated at room temperature for 0, 2, 5 and 24 hours before running on an Applied Biosystems 7500 Fast real-time instrument (96-well, 20 μl reactions). Using synthetic Twist RNA, consistent performance is observed with up to 24 hours of room temperature incubation with the Luna Probe One-Step RT-qPCR Mix, while TaqPath showed a Cq delay ≥1 at 2 hours with declining performance as incubation time increased.

Optimize Your
One-Step RT-qPCR/RT-PCR
# with Unique WarmStart Technology

Luna and LunaScript products contain a novel, *in silico*-designed reverse transcriptase (RT) engineered for improved performance. One-Step RT-qPCR/RT-PCR products contain Luna WarmStart Reverse Transcriptase and Hot Start *Taq* DNA Polymerase, which utilize a temperature-sensitive, reversible aptamer. The aptamer inhibits activity below 45°C and enables room-temperature reaction setup and prevents undesired non-specific activity. Furthermore, the Luna WarmStart RT has increased thermostability, improving performance at higher reaction temperatures.

Luna RT-qPCR products offer exceptional sensitivity, reproducibility and performance
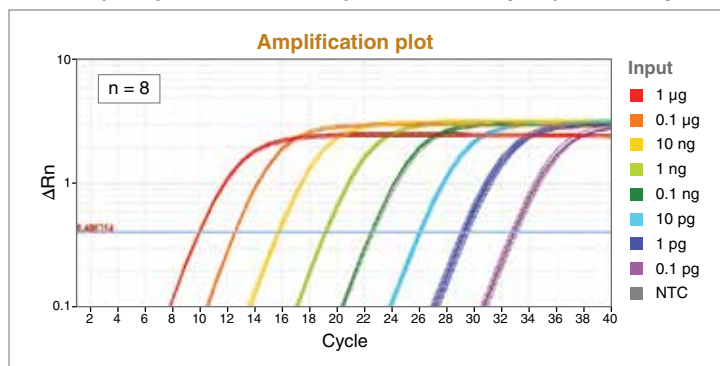
RT-qPCR targeting human GAPDH was performed using the Luna Universal One-Step RT-qPCR Kit over an 8-log range of input template concentrations (1 µg – 0.1 pg Jurkat total RNA) with 8 replicates at each concentration. Reaction setup and cycling conditions followed recommended protocols, including a 10-minute RT step at 55°C for the thermostable Luna WarmStart Reverse Transcriptase. NTC = non-template control.

The increased thermostability of Luna WarmStart Reverse Transcriptase improves performance at higher reaction temperatures
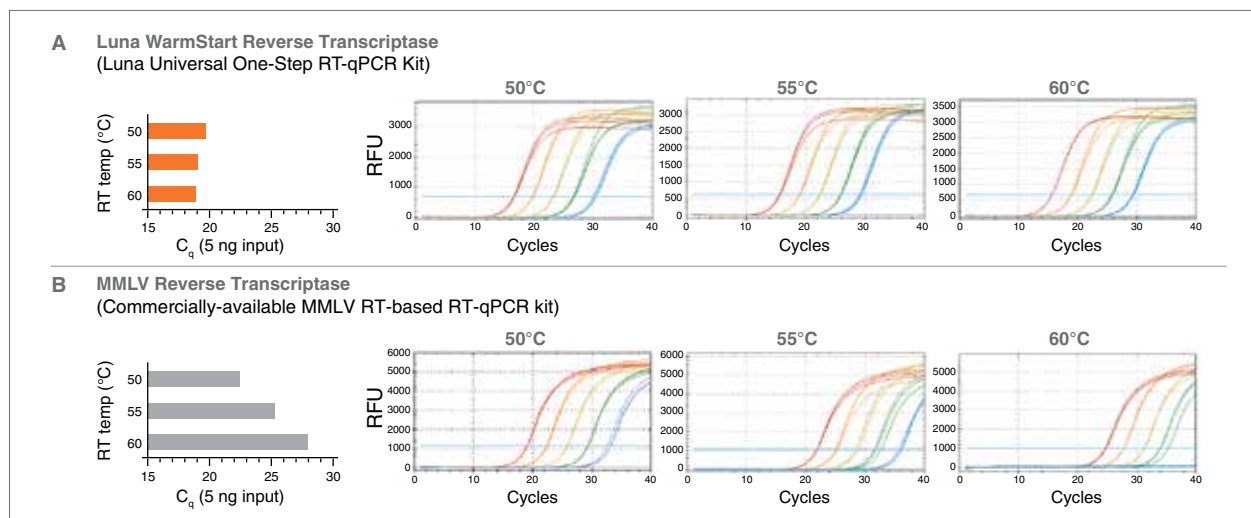
RT-qPCR experiments targeting human ribosomal protein L32 RNA were performed in triplicate over a 5-log range of input human (Jurkat) total RNA (5 pg – 50 ng) using an initial 10 min RT step performed at 50°C – 60°C, as indicated.

A. Luna WarmStart Reverse Transcriptase (recommended incubation temperature: 55°C) exhibited rapid C_q values (bar graph) and robust RT-qPCR performance (amplification plots) at each temperature, indicating that efficient reverse transcription was not perturbed by reaction temperature alterations.

B. In contrast, a commercially available MMLV (recommended incubation temperature: 50°C) exhibited delayed (increased) C_q values, poorer performance, and loss of low-input detection at elevated temperatures, consistent with loss of RT activity.

Luna 4X Mix for High Sensitivity

The Luna Probe One-Step RT-qPCR 4X Mix with UDG (NEB #M3019) is supplied at a 4X concentration and enables higher amounts of sample input. Performance in multiplexing applications has been optimized, with sensitive, linear detection achieved for up to 5 targets across a range of inputs. The mix features Luna WarmStart RT, Hot Start *Taq* DNA Polymerase, dNTPs, a universal passive reference dye, and Murine RNase Inhibitor. The inclusion of dUTP and UDG in the master mix reduces the possibility of carryover contamination between reactions.

Learn more and request a sample at **www.neb.com/M3019**

Robust amplification and detection of different viral RNA with Luna Probe One-Step RT-qPCR 4X Mix with UDG
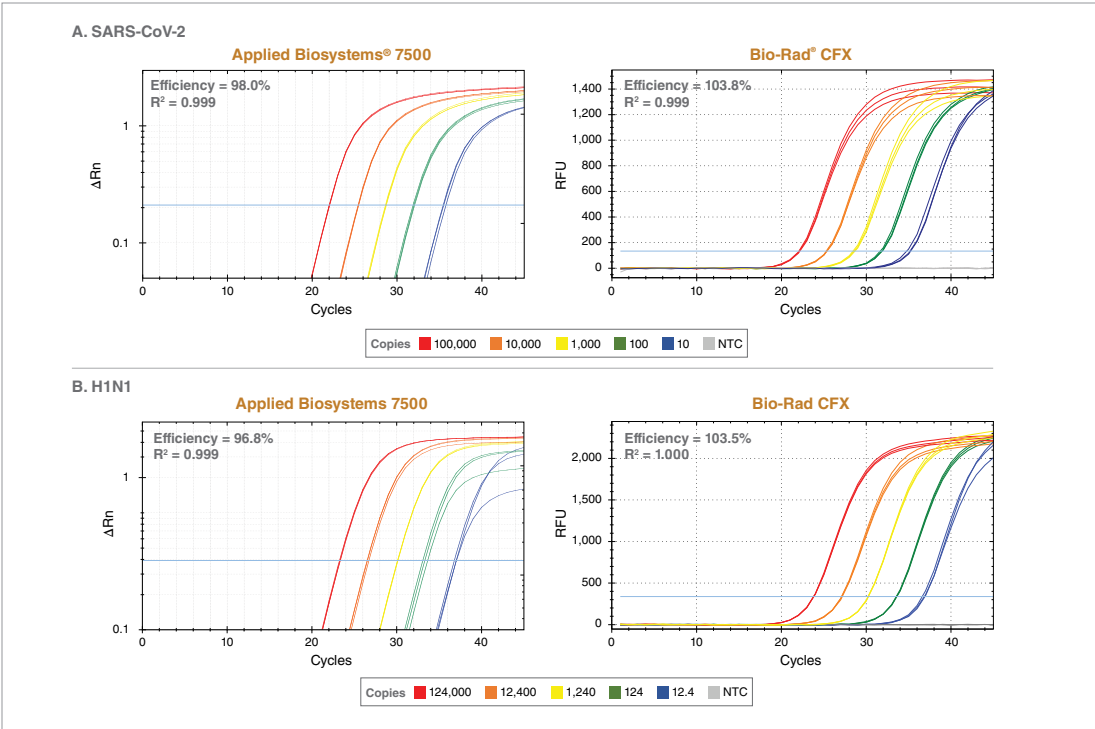
RT-qPCR targeting SARS-CoV-2 (N1 target) and H. influenza H1N1 (HA target) was performed using the Luna Probe One-Step RT-qPCR Mix with UDG. Performance in 20 μl reactions was evaluated in two real-time instruments over a 5-log range of (A) 10–100,000 copies Synthetic SARS-CoV-2 RNA Control 2 (Twist Bioscience®, SKU: 102024) diluted in 10 ng of Jurkat total RNA (BioChain, #R1255815-50) and (B) 12.4–124,000 copies Influenza A (H1N1) RNA (ATCC® VR-95-DQ™) diluted in 10 ng Jurkat total RNA. Sensitive, linear performance can be observed in the amplification of both viral targets.

The Luna Probe One-Step RT-qPCR 4X Mix with UDG outperforms comparable products
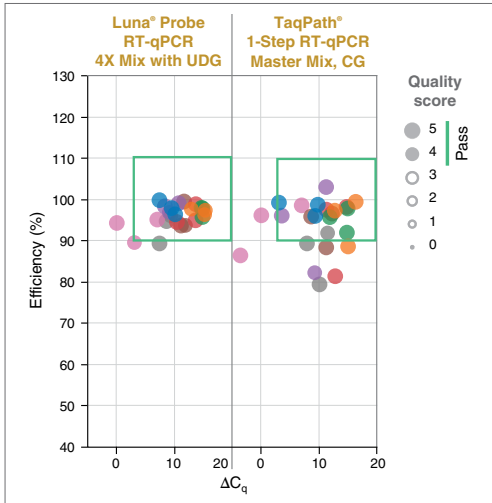
One-step RT-qPCR was tested on 8 RT-qPCR targets (indicated by color) varying in abundance, length, and %GC. Data was collected on multiple days by two users according to manufacturer's recommendations using the Applied Biosystems® QuantStudio® 6 real-time PCR system. Results were evaluated for efficiency, low input detection and lack of non-template amplification (where ΔC_q = average C_q of non-template control – average C_q of lowest input). In addition, consistency, reproducibility and overall curve quality were assessed based on metrics described previously (Quality Score). Although both products performed reasonably well, the Luna Probe RT-qPCR 4X Mix with UDG outperformed the TaqPath® 1-Step RT-qPCR Master Mix, CG, as evidenced by the higher number of experiments whose results fell in the green box.

Luna Probe RT-qPCR 4X Mix with UDG (NEB #M3019)

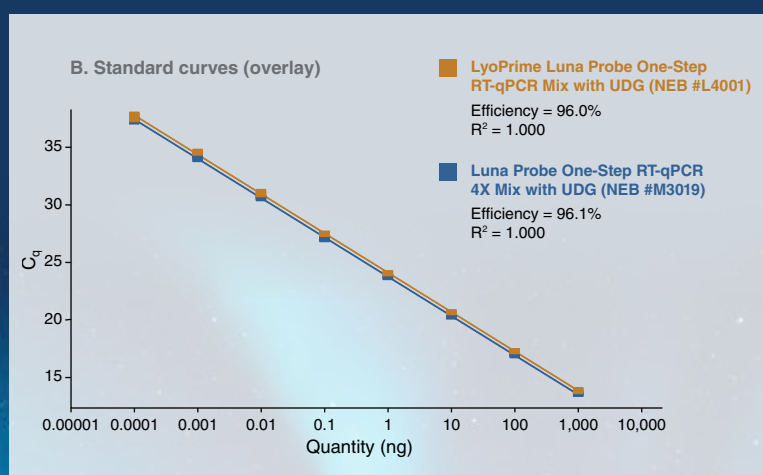|	
Number of tubes	1
Concentration (sample-limiting)	4X
dUTP included	✓
UDG included	✓
Universal ROX included	✓

See p.11 for an explanation of Dots in Boxes qPCR data assessment tool

LyoPrime Luna Probe One-Step RT-qPCR Mix with UDG

Bringing together expertise in enzyme development, manufacturing and lyophilization, NEB Lyophilization Sciences™ has created shelf-stable, lyophilized products that do not sacrifice the high-performance qualities of their liquid counterparts. The first of these products includes a mixture of enzymes and inhibitors to enable robust detection of RNA via hydrolysis-probe-based RT-qPCR. The LyoPrime Luna Probe One-Step RT-qPCR Mix with UDG enables sensitive detection of target RNA sequences in a lyophilized format. This product contains the same versatile features and strong performance as the liquid version: Luna Probe One-Step RT-qPCR 4X Mix with UDG (NEB #M3019).

Lyophilized and liquid Luna RT-qPCR mixes demonstrate equivalent strong performance

B. Standard curves (overlay)

LyoPrime Luna Probe One-Step RT-qPCR Mix with UDG (NEB #L4001)
Efficiency = 96.0%
R^2 = 1.000

Luna Probe One-Step RT-qPCR 4X Mix with UDG (NEB #M3019)
Efficiency = 96.1%
R^2 = 1.000

Axis: C_q vs Quantity (ng)

Standard Curve results were substantially equivalent for the lyophilized (gold) and liquid-format (blue) mixes, with strong linearity and reproducibility observed, even at the lowest concentrations tested.

LyoPrime Luna rehydration process: pre-hydration, hydration, post-hydration

Pre-hydration (before)   Hydration   Post-hydration (20 sec. after)

Learn more and access our on-demand webinar discussing considerations for lyophilizing reagents at
**www.neb.com/LyoPrime**

Optimized SARS-CoV-2 RNA Detection

The Luna SARS-CoV-2 RT-qPCR Multiplex Assay Kit (NEB #E3019) is a real-time RT-PCR assay for the qualitative detection of SARS-CoV-2 nucleic acid. The primer/probe mix is specific to two regions of the SARS-CoV-2 virus N gene [based on sequences provided by the Centers for Disease Control and Prevention (CDC)]. The probes have been modified to contain different fluorophores (N1: HEX; N2: FAM) to enable multiplexing. To ensure the integrity of the input material and absence of inhibition, an internal control primer and probe set, designed to amplify the human RNase P gene, is included in the primer mix. The kit also includes the Luna Probe One-Step RT-qPCR Mix with UDG (NEB #M3019) and a positive control containing the full SARS-CoV-2 nucleocapsid protein (N gene).

Learn more about
the Luna SARS-CoV-2
RT-qPCR Multiplex
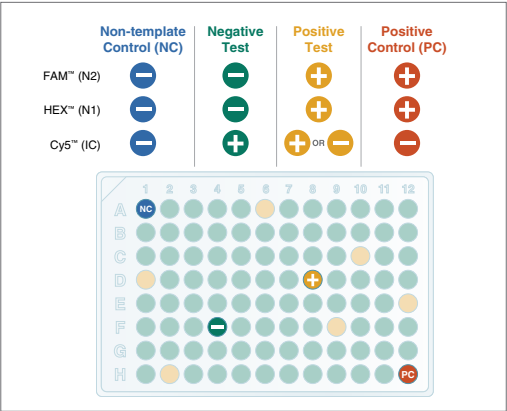Assay Kit at
www.neb.com/E3019

The Luna SARS-CoV-2 RT-qPCR Multiplex Assay Kit demonstrates a lower limit of detection than TaqPath® 1-Step RT-qPCR Master Mix, CG
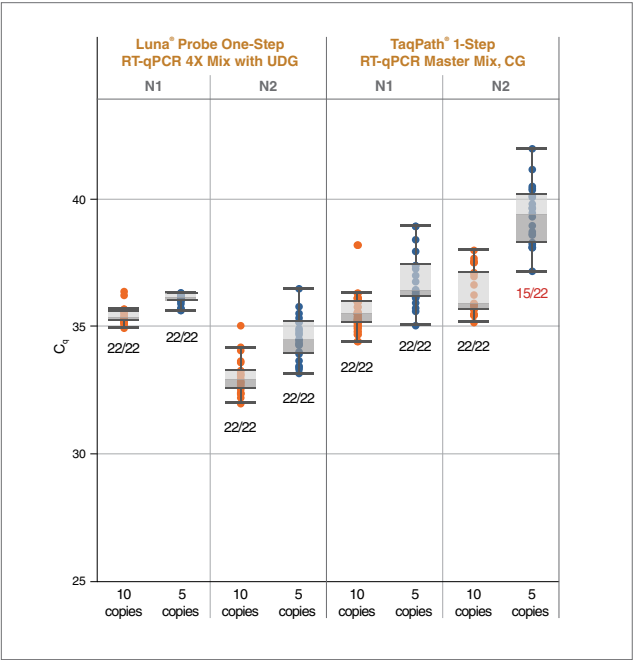
LOD comparison using: Luna SARS-CoV-2 RT-qPCR Multiplex Assay Kit for multiplex RT-qPCR targeting 2019-nCoV_N1 target (HEX) and 2019-nCoV_N2 target (FAM), according to reaction and cycling conditions provided in the E3019 product manual, and TaqPath 1-Step RT-qPCR Master Mix, CG for singleplex RT-qPCR targeting 2019-nCoV_N1 (FAM) and 2019-nCoV_N2 (FAM), according to the CDC 2019-Novel Coronavirus (2019-nCoV) Real-Time RT-PCR Diagnostic Panel guidelines. Performance was evaluated using Synthetic Twist SARS-CoV-2 RNA Control 2 diluted in 10 ng of Jurkat total RNA. Data was collected on an Applied Biosystems 7500 Fast real-time instrument (96-well, 20 μl reactions). Under these conditions, the Luna Kit has an LOD of 5 copies/reaction for both targets while the LOD using TaqPath is 10 copies/reaction for these targets.

Example Assay Setup and Anticipated Results

Using the Luna SARS-CoV-2 RT-qPCR Multiplex Assay Kit, up to 94 different samples can be assessed in a single 96-well plate. Anticipated results for each sample type are shown (in each fluorophore channel).

Go Direct to RNA Quantitation: Luna Cell Ready Module and Kits

The Luna Cell Ready One-Step RT-qPCR Kit provides all the necessary components for direct RNA detection and quantitation from cultured mammalian and insect cell lines. Removing the need for traditional RNA extraction and purification, it offers a robust, sensitive, and convenient workflow for evaluating RNA expression levels in a 15-minute sample preparation protocol (prior to RT-qPCR).
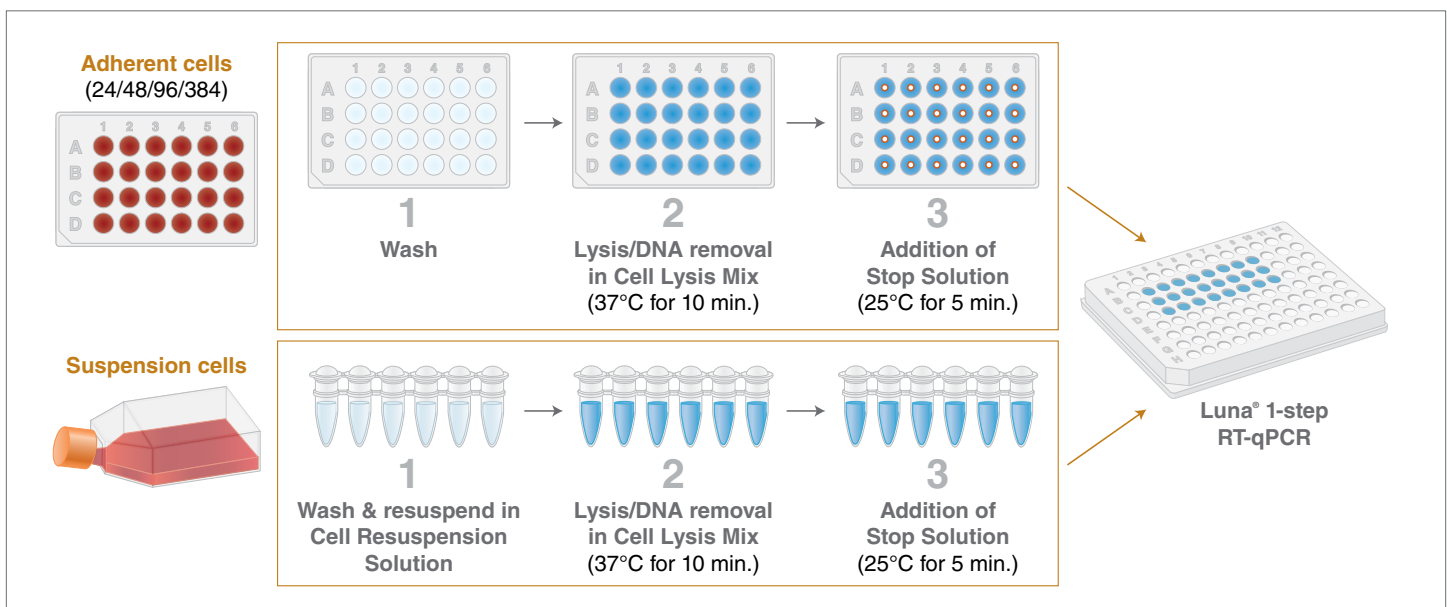
The Luna Cell Ready Lysis One-Step RT-qPCR Kit is available for both dye (NEB #E3030) and probe (NEB #E3031) detection methods. In addition, the lysis module can be purchased separately (NEB #E3032).

- Sensitive qPCR quantitation: linear RNA detection across a 5-log range of cell input dilutions

- Coordinated cell lysis, RNA release, and genomic DNA removal in a fast 15-minute protocol

- Increased convenience and minimal sample loss compared to alternative RNA purification methods

- Efficient cell lysate preparation from 10 to 100,000 cells across numerous cell lines

- Obtain reliable and precise results comparable to purified RNA

- Non-interfering, visible tracking dye eliminates pipetting errors

- Features Luna Universal One-Step RT-qPCR Kits (NEB #E3005/#E3006) for robust performance

Luna Cell Ready Workflow

Adherent cells (24/48/96/384)

1 — Wash
2 — Lysis/DNA removal in Cell Lysis Mix (37°C for 10 min.)
3 — Addition of Stop Solution (25°C for 5 min.)

Suspension cells

1 — Wash & resuspend in Cell Resuspension Solution
2 — Lysis/DNA removal in Cell Lysis Mix (37°C for 10 min.)
3 — Addition of Stop Solution (25°C for 5 min.)

Luna® 1-step RT-qPCR

The Luna Cell Ready One-Step RT-qPCR Kit provides all the necessary components for direct RNA detection and quantitation from cultured cells (up to 100,000 cells per 50 µl lysis reaction). Coordinating the actions of DNase I and the Luna Cell Ready Protease, the Luna Cell Ready Lysis Module offers a simple workflow resulting in effective cell lysis, RNA release, and genomic DNA removal simultaneously in a 15-minute protocol. Up to 2 µl lysate (equivalent to RNA from 0.2 - 4,000 cells) can be transferred into 20 µl downstream RT-qPCR reactions.

Speed up Your Two-Step RT-qPCR: LunaScript RT SuperMix

Two-step RT-qPCR uncouples cDNA synthesis and qPCR analysis, allowing greater freedom in selecting reverse transcriptases and qPCR reagents separately. This flexibility can be useful for controlling sequence representation, qPCR efficiency, and optimization of reaction conditions when working with difficult RT-qPCR reactions or low RNA inputs.

LunaScript RT SuperMix is an optimized master mix for first strand cDNA synthesis and can be used in amplicon sequencing or a two-step RT-qPCR workflow. It employs the Luna Reverse Transcriptase in a convenient supermix format containing random hexamer and oligo-dT primers, dNTPs, and Murine RNase Inhibitor. This kit delivers best-in-class performance and requires the shortest reaction time (< 15 min) and tolerates elevated temperatures (55°–65°C) for working with templates with complex secondary structures.

The cDNA products generated by LunaScript have been extensively evaluated in qPCR using the Luna qPCR Master Mixes (NEB #M3003/M3004). In combination, these products provide a two-step RT-qPCR workflow with excellent sensitivity and accurate, linear quantitations.

At just 13 minutes, the LunaScript RT SuperMix Kit offers the shortest available first-strand cDNA synthesis protocol
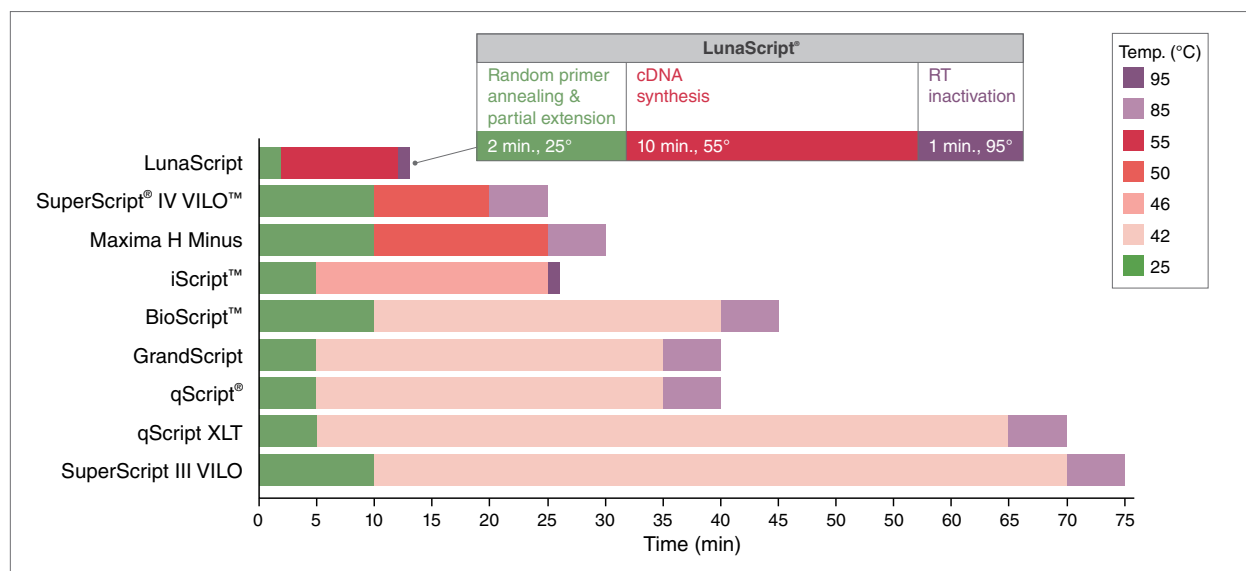
Comparison of recommended protocols for cDNA synthesis. The LunaScript RT SuperMix Kit requires the shortest reaction time and tolerates elevated temperatures, reducing complications from RNA secondary structure.
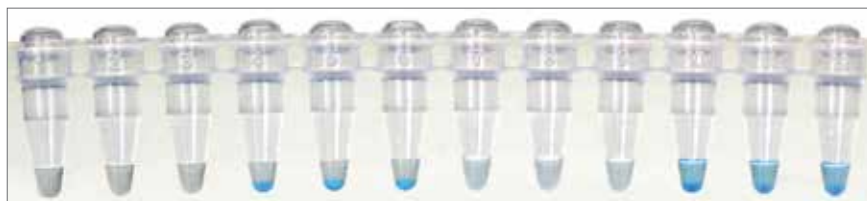
LunaScript forms a dark blue pellet at the bottom of the reaction vessel, easily discernible in clear or blue color mixes.
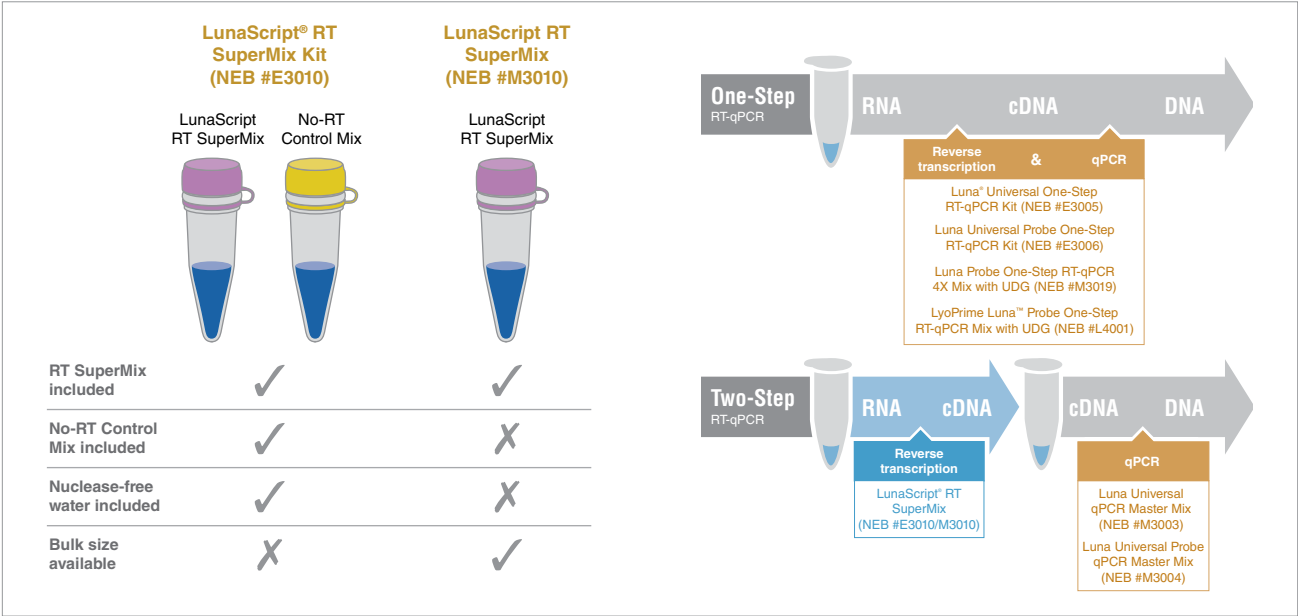
LunaScript RT SuperMix: Product Formats

LunaScript RT SuperMix is offered in multiple formats. The full kit version (NEB #E3010) includes the supermix, a No-RT control mix, and nuclease-free water.

LunaScript RT SuperMix (NEB #M3010) is a streamlined, supermix-only offering and does not include the No-RT control mix or nuclease-free water. It is also available in large, bulk-size formats for high throughput users.

Comparison of LunaScript products

	LunaScript® RT SuperMix Kit (NEB #E3010)		LunaScript RT SuperMix (NEB #M3010)
	LunaScript RT SuperMix	No-RT Control Mix	LunaScript RT SuperMix
RT SuperMix included	✓		✓
No-RT Control Mix included	✓		✗
Nuclease-free water included	✓		✗
Bulk size available	✗		✓

One-Step RT-qPCR — RNA → cDNA → DNA

Reverse transcription & qPCR

Luna® Universal One-Step RT-qPCR Kit (NEB #E3005)
Luna Universal Probe One-Step RT-qPCR Kit (NEB #E3006)
Luna Probe One-Step RT-qPCR 4X Mix with UDG (NEB #M3019)
LyoPrime Luna™ Probe One-Step RT-qPCR Mix with UDG (NEB #L4001)

Two-Step RT-qPCR — RNA → cDNA → cDNA → DNA

Reverse transcription
LunaScript® RT SuperMix (NEB #E3010/M3010)

qPCR
Luna Universal qPCR Master Mix (NEB #M3003)
Luna Universal Probe qPCR Master Mix (NEB #M3004)

LunaScript RT SuperMix demonstrates superior linear detection of RNA targets

Dye-based detection | Probe-based detection

LunaScript™ | iScript™ | SuperScript™ IV VILO | LunaScript

Efficiency (%) vs ΔCq

| Targets passing performance criteria | 95% | 73% | 60% | 95% |

Commercially available cDNA supermixes were used according to manufacturer's recommendations to generate cDNA from 1 µg – 100 pg human (Jurkat) total RNA. cDNA products were then evaluated by qPCR using eight targets varying in abundance, length and %GC. qPCR detection was performed using the Luna Universal qPCR Master Mix (NEB #M3003) or Luna Universal Probe qPCR Master Mix (NEB #M3004). Results were evaluated for efficiency and ΔC_q, where ΔC_q measures low input detection and lack of non-template control (NTC) amplification (ΔC_q = average C_q of NTC - average C_q of lowest input). Green box indicates target performance criteria (Efficiency = 90-110%, $\Delta C_q \geq 3$).
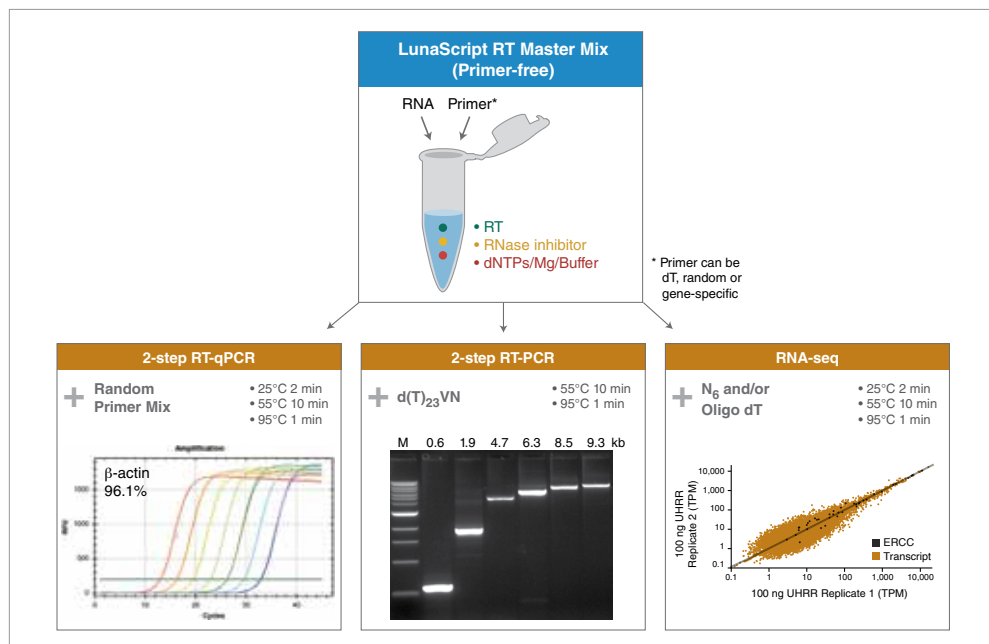
Flexible cDNA Synthesis

The LunaScript RT Master Mix Kit (Primer-free) (NEB #E3025) features an optimized 5X master mix containing all the necessary components for first-strand cDNA synthesis, except primers. The mix includes the thermostable Luna Reverse Transcriptase, which supports cDNA synthesis at elevated temperatures, dNTPs, and Murine RNase Inhibitor to protect template RNA from degradation. The mix is compatible with random primers, oligo dT primers, and gene-specific primers, enabling maximum cDNA synthesis flexibility.

LunaScript RT Master Mix (Primer-free) supports numerous applications

By adding different primers including a Random Primer Mix, $d(T)_{23}VN$ oligos, or random hexamers, the LunaScript RT Master Mix can produce cDNA that is ideally suited for downstream applications such as RT-qPCR, RT-PCR, and RNA-seq studies.

Superior Multiplexing with Luna and Q5

The LunaScript Multiplex One-Step RT-PCR Kit (NEB #E1555) offers a streamlined protocol for cDNA synthesis and PCR amplification in a single reaction. It features Luna WarmStart RT and Q5 Hot Start High-Fidelity DNA Polymerase. The kit has robust multiplex target amplification capacity and enables various applications such as diagnostics, pathogen detection, and viral genome sequencing (including the ~50 amplicons per reaction used in ARTIC SARS-CoV-2 sequencing protocols).
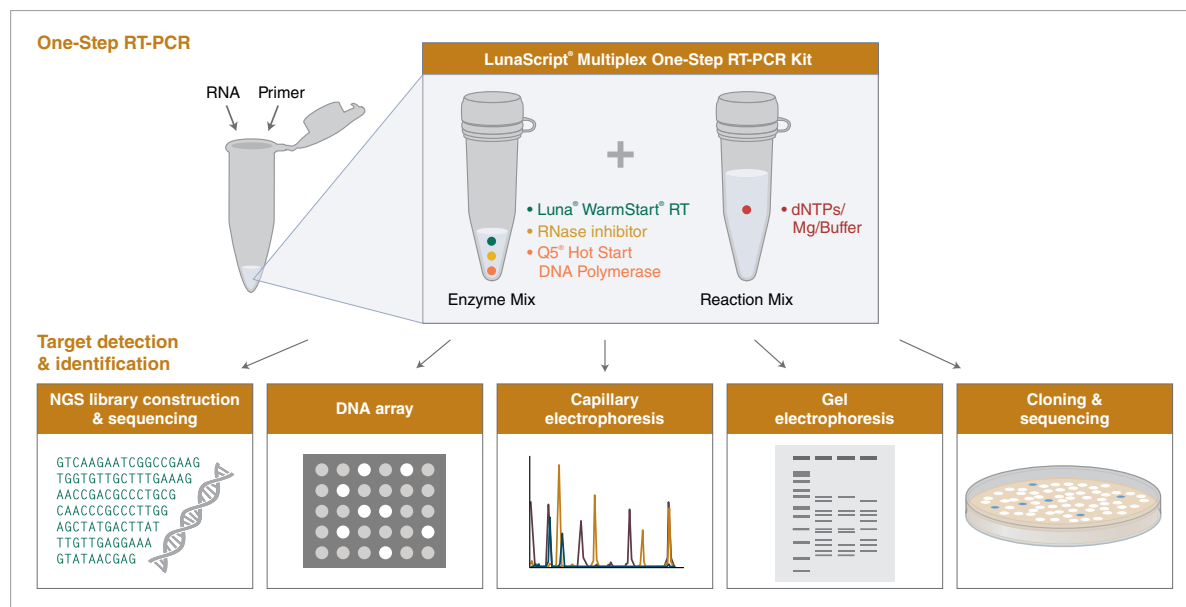
We tested plates and plates of reactions so you don't have to.

Evaluating qPCR results: capturing performance as "dots in boxes"

NEB has developed a method to better evaluate the large amount of qPCR data generated in an experiment. The output of this analysis is known as a dot plot, and captures the key features of a successful, high-quality qPCR experiment as a single point. This method of analysis allows many targets and conditions to be compared in a single graph.

For each experiment, triplicate reactions are set up across a five-log range of input template concentrations (Amplification plot, bottom-left). Three non-template control (NTC) reactions are also included, for a total of 18 reactions per condition/target. Efficiency (%) is calculated (Standard plot, top-left) and is plotted against ΔC_q (dot plot, top-center), which is the difference between the average C_q of the NTC and the lowest input. This parameter captures both detection of the lowest input and non-template amplification.

Acceptable performance criteria are defined as an Efficiency of 90–110% and a ΔC_q of ≥ 3 (green box – pass).

Other performance criteria are captured using a 5-point quality score (Quality score metrics, top-right). Included are:

1. Linearity of amplification, as indicated by the R^2 standard curve
2. Reproducibility, as indicated by the consistency of triplicate C_q values for each input concentration
3. Fluorescence consistency, as indicated by similar endpoint fluorescence (RFU_{max})
4. Curve steepness
5. Sigmoid curve shape

Breaking it down: how we translate qPCR data into "dots in boxes"
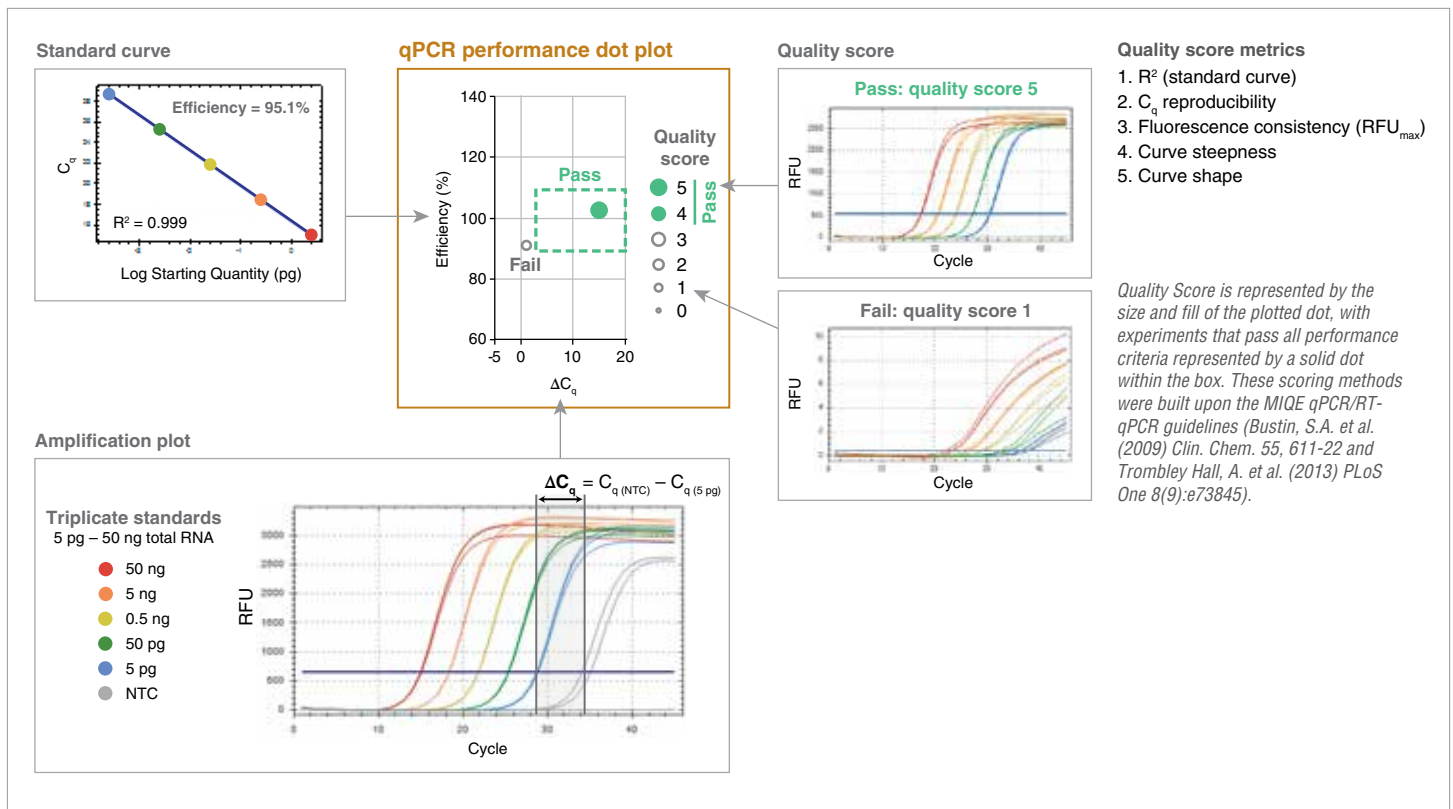
Experience Best-in-class Performance

All NEB products undergo rigorous testing to ensure optimal performance, and Luna is no exception. We took into consideration numerous important traits when evaluating qPCR, including specificity, sensitivity, accuracy and reproducibility, to develop best-in-class qPCR reagents. Furthermore, we did a comprehensive evaluation of commercially-available qPCR and RT-qPCR reagents and developed a method of analysis that allows you to quickly compare and evaluate the performance of these products. We wanted to be sure that Luna products will perform to your expectations for all your targets.

Luna products offer exceptional sensitivity, reproducibility and qPCR performance

qPCR targeting human GAPDH was performed using the Luna Universal Probe qPCR Master Mix over a 6-log range of input template concentrations (20 ng – 0.2 pg Jurkat-derived cDNA) with 8 replicates at each concentration. cDNA was generated from Jurkat total RNA using the Protoscript® II First Strand cDNA Synthesis Kit (NEB #E6560). NTC = non-template control
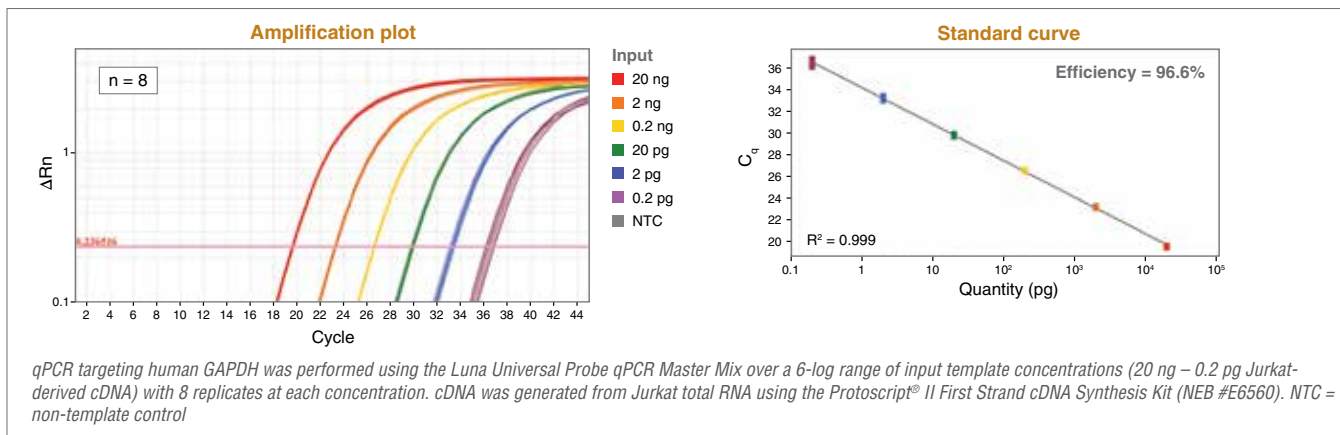
Evaluation of commercially-available dye-based qPCR reagents demonstrates the robustness and specificity of Luna
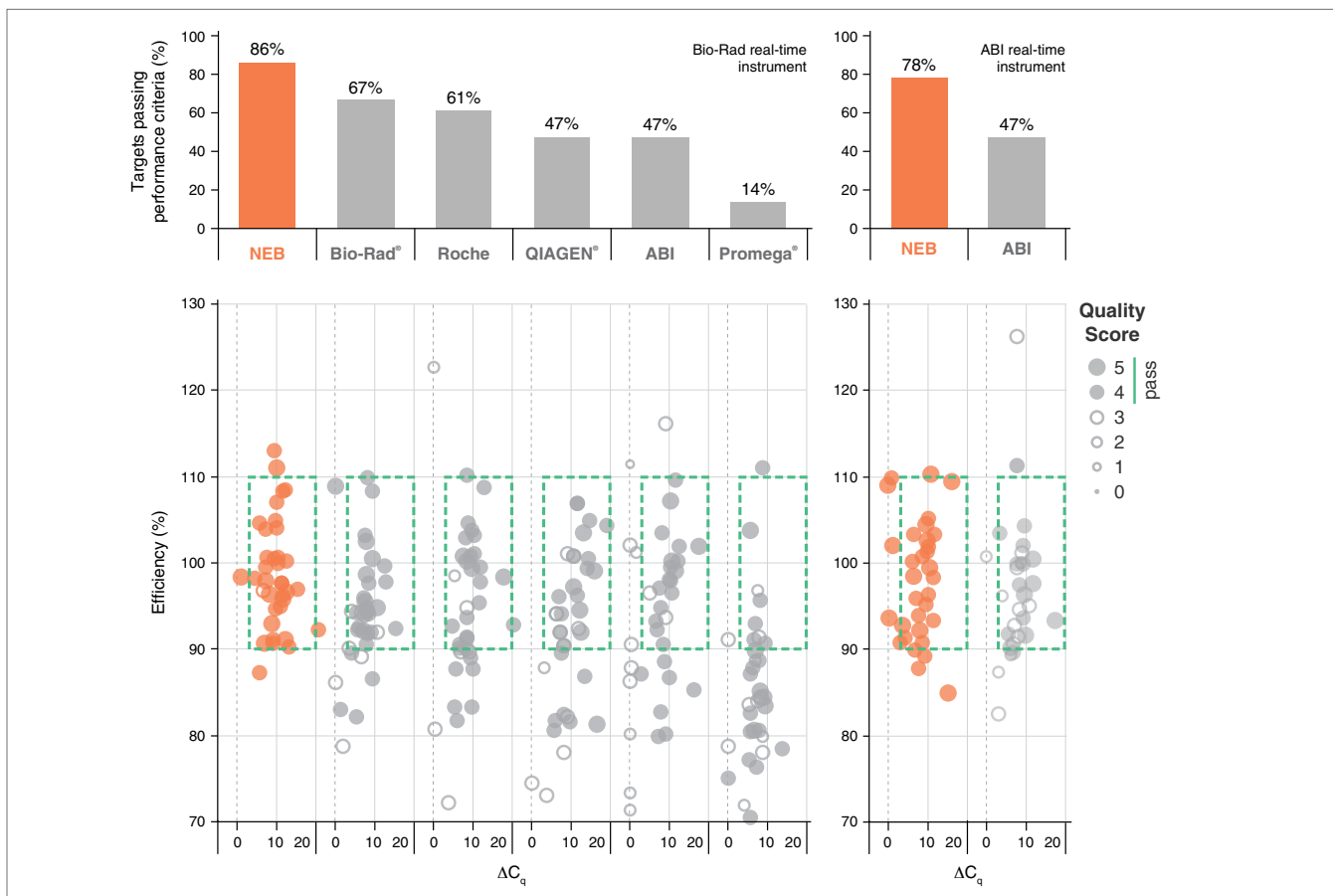
qPCR reagents from NEB and other manufacturers were tested across 16–18 qPCR targets varying in abundance, length and %GC, using either Jurkat genomic DNA or Jurkat-derived cDNA as input (10 genomic DNA targets and 8 cDNA targets on a Bio-Rad real-time instrument, 9 genomic and 7 cDNA targets on an ABI instrument). For each testing condition, data was collected by 2 users and according to manufacturer's specifications. Results were evaluated for efficiency, low input detection and lack of non-template amplification (where ΔC_q = average C_q of lowest input – average C_q of non-template control). In addition, consistency, reproducibility and overall curve quality were assessed (Quality Score). Bar graph indicates % of targets that met acceptable performance criteria (indicated by green box on dot plot and Quality Score > 3). Results for NEB and other major manufacturers are shown: Bio-Rad, SsoAdvanced™ Universal SYBR® Green Supermix; Roche®, FastStart™ SYBR Green Master; QIAGEN, QuantiTect® SYBR® Green PCR Kit; ABI, PowerUP® SYBR Green Master Mix; Promega, GoTaq® qPCR Master Mix. The Luna Universal qPCR Master Mix outperformed all other reagents tested.

Ordering Information

Product Name	NEB #	Size
Luna Universal qPCR Master Mix	M3003S/L/X/E	200/500/1,000/2,500 rxns
Luna Universal Probe qPCR Master Mix	M3004S/L/X/E	200/500/1,000/2,500 rxns
Luna Universal One-Step RT-qPCR Kit	E3005S/L/X/E	200/500/1,000/2,500 rxns
Luna Universal Probe One-Step RT-qPCR Kit	E3006S/L/X/E	200/500/1,000/2,500 rxns
Luna Probe One-Step RT-qPCR Kit (No ROX)	E3007E	2,500 rxns
Luna Probe One-Step RT-qPCR 4X Mix with UDG	M3019S/L/X/E	200/500/1,000/2,000 rxns
Luna Probe One-Step RT-qPCR 4X Mix with UDG (No ROX)	M3029S/L/E	200/500/2,000 rxns
LyoPrime Luna Probe One-Step RT-qPCR Mix with UDG	L4001S	120 rxns
Luna SARS-CoV-2 RT-qPCR Multiplex Assay Kit	E3019S	96 rxns
LunaScript RT Master Mix Kit (Primer-free)	E3025S/L	25/100 rxns
LunaScript RT SuperMix Kit	E3010S/L	25/100 rxns
LunaScript RT SuperMix	M3010L/X/E	100/500/2,500 rxns
LunaScript Multiplex One-Step RT-PCR Kit	E1555S/L	50/250 rxns
Luna Cell Ready One-Step RT-qPCR Kit	E3030S	100 rxns
Luna Cell Ready Probe One-Step RT-qPCR Kit	E3031S	100 rxns
Luna Cell Ready Lysis Module	E3032S	100 rxns

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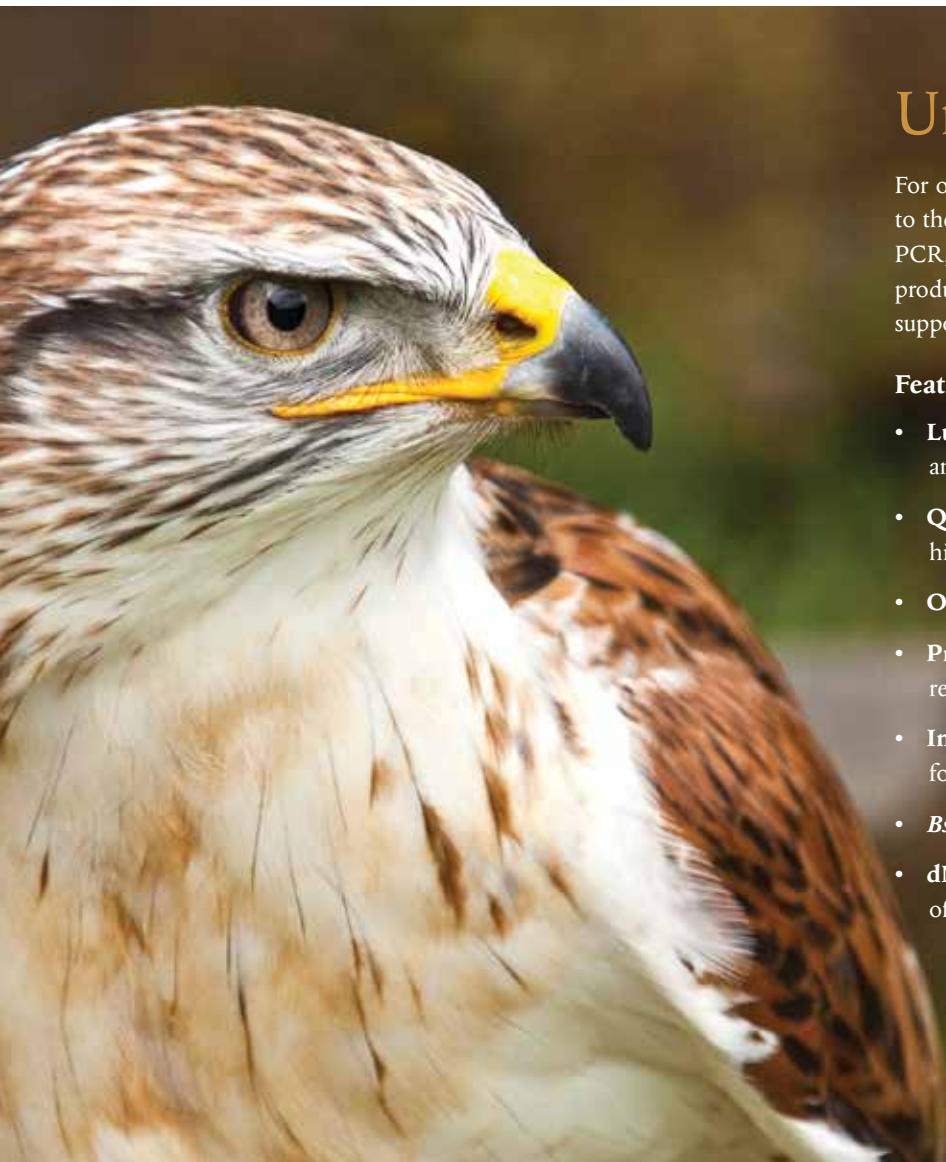
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e-mail: info@neb.com

New England Biolabs (UK) Ltd
75-77 Knowl Piece
Wilbury Way
Hitchin
Herts
SG4 0TY

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