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SuperScript IV One-Step RT-PCR System

For superior RT-PCR performance

Even with challenging RNA samples, you can get better results faster and easier than with any other one-step RT-PCR product. The Invitrogen™ SuperScript™ IV One-Step RT-PCR System combines high-processivity SuperScript IV Reverse Transcriptase and high-fidelity Invitrogen™ Platinum™ SuperFi™ DNA Polymerase to provide superior one-step RT-PCR performance.

- Two-phase hot-start activation mechanism for high specificity, yield, and room-temperature setup
- Superior performance with sensitivity down to 0.01 pg of RNA, target length up to 13.8 kb, and the fastest one-step RT-PCR protocol
- Reliable target detection even with RNA samples of suboptimal purity
- Fast and easy gDNA removal for superior accuracy and confidence in your results

Two-phase hot-start activation mechanism explained

The innovative two-phase hot-start mechanism allows temporal separation of the activities of the reverse transcriptase and DNA polymerase, in order to deliver high specificity and yield in one-step RT-PCR.

Reaction setup

18-23°C



SuperScript IV Reverse Transcriptase and Platinum SuperFi DNA Polymerase remain inactive because of the hot-start mechanisms preventing nonspecific activity.

First hot-start activation phase

45-60°C



Reverse transcriptase is activated and cDNA synthesis is initiated. DNA polymerase remains inactive to prevent any residual activity.

Second hot-start activation phase

98°C



DNA polymerase is activated and reverse transcriptase is simultaneously inactivated to allow highly efficient and specific DNA amplification following this step.

To find out more, watch the one-step RT-PCR video at **thermofisher.com/ssiv-onestep**



High specificity and yield with the shortest protocol

With the innovative two-phase hot-start activation mechanism, the SuperScript IV One-Step RT-PCR System generates specific products at high yields in half of the reaction time of other commercial kits.

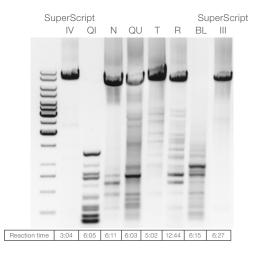


Figure 1. Amplification of long targets with high specificity in significantly shorter times. Detection of 7.8 kb target from total HeLa RNA using SuperScript IV One-Step RT-PCR System, SuperScript III One-Step RT-PCR System with Platinum *Taq* High Fidelity DNA Polymerase, and supplier QI, N, QU, T, R, and BL one-step RT-PCR products. Reactions were performed according to the suppliers' recommendations. Total reaction times for one-step RT-PCR are indicated in hours:minutes. The RNA target failed to amplify with products from suppliers QI and BL.

T_m calculator

Annealing temperature rules for Platinum SuperFi DNA Polymerase are different from many common DNA polymerases (such as Taq DNA polymerase). For optimal one-step RT-PCR results with the SuperScript IV One-Step RT-PCR System, use the T_m calculator on our website.

Go to thermofisher.com/tmcalculator

Superior sensitivity-down to 0.01 pg RNA

The high sensitivity of the SuperScript IV One-Step RT-PCR System enables detection of low-abundance targets and allows for one-step RT-PCR experiments, even when RNA input is limited.

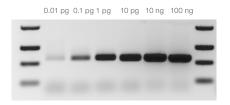


Figure 2. Detection from low amounts of input RNA. A 0.35 kb RNA target was detected from a broad range of HeLa total RNA inputs with the SuperScript IV One-Step RT-PCR System.

Broad range of RNA target lengths—up to 13.8 kb

Due to the high processivity of SuperScript IV Reverse Transcriptase and Platinum SuperFi DNA Polymerase, the SuperScript IV One-Step RT-PCR System enables detection of a broad range of target lengths.

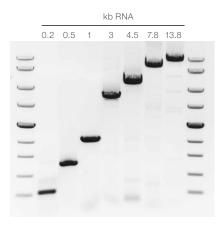


Figure 3. Versatility across a broad range of target lengths. Detection of human RNA fragments ranging from 0.2 to 13.8 kb with the SuperScript IV One-Step RT-PCR System.

High performance even with suboptimally pure RNA

The SuperScript IV One-Step RT-PCR System is able to withstand common inhibitors of reverse transcriptase and PCR, such as copurified compounds from biological samples or reagents used for RNA purification. This exceptional robustness makes the system less dependent on RNA sample purity to achieve reliable results.

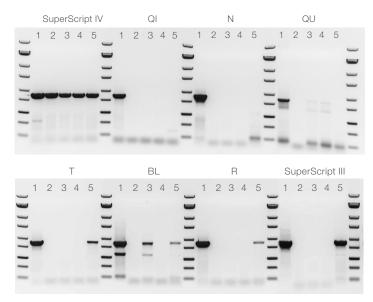


Figure 4. Resistance to inhibitors. Detection of a 1 kb RNA target from total HeLa RNA using the SuperScript IV One-Step RT-PCR System or other one-step kits in reaction mixtures containing: 1—no inhibitor, 2—heparin (0.18 µg/µL), 3—xylan (2.5 µg/µL), 4—humic acid (0.02 µg/µL), or 5—LiCl (2 µg/µL). The enzymes in all kits except the SuperScript IV One-Step RT-PCR System were inhibited by the indicated amounts of inhibitors.

High enzyme processivity for better inhibitor tolerance

Numerous compounds that have inhibitory effects on reverse transcriptases and PCR enzymes are commonly present in RNA samples, even after employing thorough RNA purification methods.

SuperScript IV Reverse Transcriptase and Platinum SuperFi DNA Polymerase are engineered for high processivity and increased affinity to templates, which enhances their ability to work in the presence of common reaction inhibitors. This enables RT and PCR even with impure RNA samples.

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Benchtop stability of preassembled reactions

The stability of SuperScript IV One-Step RT-PCR reagents at room temperature for an extended time enables high-throughput applications.

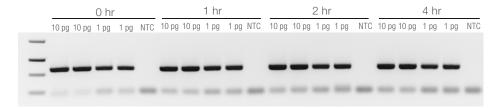


Figure 5. Extended stability at room temperature. Duplicate one-step RT-PCR reactions on a 0.35 kb RNA target from 1 and 10 pg of total HeLa RNA were assembled using SuperScript IV One-Step RT-PCR reagents and left at room temperature for up to 4 hours before cycling. Even after 4 hours at room temperature, highly efficient and specific target amplification was achieved. NTC: no-template control.

Complete system with integrated gDNA removal

RNA purification methods, including on-column DNase digestion protocols, often fail to remove gDNA completely. Amplification of contaminating gDNA can cause nonspecific or misleading one-step RT-PCR results. Traditional gDNA decontamination protocols with DNase I include time-consuming DNase inactivation or removal

steps under conditions that can damage RNA and affect results. The SuperScript IV One-Step RT-PCR System with Invitrogen™ ezDNase™ Enzyme allows efficient, fast, and gentle (5 min at 37°C) gDNA elimination from RNA samples and ensures the highest accuracy and confidence in one-step RT-PCR results.

Ordering information

Product	Quantity	Cat. No.
SuperScript IV One-Step RT-PCR System		
Includes SuperScript IV RT Enzyme Mix (SuperScript IV RT, RNase inhibitor, RT blocker, storage buffer), 2X Platinum SuperFi RT-PCR Master Mix (Platinum SuperFi DNA Polymerase, reaction buffer, dNTPs), nuclease-free water	25 reactions	12594025
	100 reactions	12594100
SuperScript IV One-Step RT-PCR System with ezDNase Enzyme		
Includes SuperScript IV RT Enzyme Mix (SuperScript IV RT, RNase inhibitor, RT blocker, storage buffer), 2X Platinum SuperFi RT-PCR Master Mix (Platinum SuperFi DNA Polymerase, reaction buffer, dNTPs), ezDNase Enzyme, ezDNase buffer, nuclease-free water	25 reactions	12595025
	100 reactions	12595100

Explore the complete portfolio of Applied Biosystems[™] and Invitrogen[™] PCR products at **thermofisher.com/pcrworkflow**

