Product Information	
ZipScript™ One-Step RT-qPCR Mix	
Part Number	P7640L
Storage Temperature	-25°C to -15°C
Reaction Size	1000 Reactions
Volume	0.8 mL
Lot Number	
Reference Number	

Product Description:
The ZipScript One-Step RT-qPCR Mix is a highly sensitive and reproducible RT-qPCR solution optimized for real-time PCR. The 25X enzyme mix is accompanied by a 2X reaction buffer.

Product Specifications	
Assay	**RT-qPCR**
Specification	Amplification of Test Lot within 1Ct of Reference Lot in a one-step RT-qPCR Assay

Quality Control Analysis:

The functionality of the ZipScript is evaluated by amplification of three mRNA transcripts in a one-step RT-qPCR assay. The amplification threshold (Ct) of the test lot is compared to a reference lot.

Notes:
Enzyme components were tested prior to formulation of the master mix and found free of contaminating endonucleases and exonucleases. Enzyme purity was >99% as determined by SDS-PAGE and negligible *E.coli* genomic DNA contamination was confirmed by qPCR. Specific activity was verified for each enzyme pre-formulation.

Supplied with:
ZipScript Reaction Buffer 1 (5X) B7641

Limitations of Use
This product was developed, manufactured, and sold for *in vitro* use only. The product is not suitable for administration to humans or animals. MSDS sheets relevant to this product are available upon request.

Protocol

ZipScript One-Step RT-qPCR Reaction Setup:

1. All components and reaction set up should be kept on ice.
2. Thaw the 2X ZipScript Reaction Buffer I completely and vortex for 3-5 seconds to mix thoroughly. Quick spin to collect contents if necessary.
3. Prepare primer/probe mix. A final concentration of 0.4-0.9 µM for each primer and 0.1-0.5 µM for probe is recommended. However, the optimal concentration for primers/probe needs to be empirically determined for each assay.
4. Determine the number of reactions to prepare, including No Template Controls (NTCs). Add 10% extra volume to compensate for the pipetting loss.
5. Follow the table below to set up the reaction mix. It is recommended to make a master mix to minimize variations and potential errors.

Components	Volume/Rxn	Final Concentration
2X ZipScript Reaction Buffer 1	10 µL	1X
Primer/Probe Mix	X µL	Variable
25X ZipScript Enzyme Mix	0.8 µL	1X
RNA Template	Variable	-
50X ROX (optional)	0.4 µL	1X
Nuclease-Free Water	To a final **20 µL** reaction volume	-

6. Seal PCR plate and spin briefly to bring down reagents.

Thermal Cycling Conditions:

Program the cycling conditions based on the recommendations below.

Standard Cycling Program* Steps	Temperature	Time	Cycles
Reverse Transcription	50°C	15 min	1
Taq Activation/Initial Denaturation	95°C	2 min	1
Denaturation	95°C	15 sec	40
Annealing/Extension*	60°C	30-60 sec	

* Cycling parameters can be modified (especially the annealing/extension condition) to fit specific primer/probe selection.

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