Thermo SCIENTIFIC

PRODUCT INFORMATION

Thermo Scientific Verso 1-Step RT-PCR Hot-Start Kit

#AB-1455/A 40 x 50 µL

Lot _ **Expiry Date _**

Ordering Information

Component	#AB-1455/A 40 rxns of 50 µL	#AB-1455/B 200 rxns of 50 µL
Verso Enzyme Mix	40 µL	200 µL
2X 1-Step PCR Hot-Start Master Mix	1 mL	5 × 1 mL
RT Enhancer	100 µL	500 µL

Store at -20°C

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Description

Thermo Scientific Verso 1-Step RT-PCR Hot-Start Kit supplies all the components required to perform a rapid, sensitive and reproducible RT-PCR for the detection and analysis of RNA.

Verso™ Enzyme Mix includes Verso Reverse Transcriptase, which is active at high temperatures, is highly sensitive and can generate long cDNA strands. This mix also contains RNase inhibitor to protect RNA templates from degradation.

2X 1-Step PCR Hot-Start Master Mix, a proprietary reaction buffer which has been optimized to allow both reverse transcription and PCR amplification to occur in the same reaction across a wide range of templates. It contains Thermo Scientific Thermo-Start DNA Polymerase, a chemically modified hot-start version of Thermo Scientific ThermoPrime DNA Polymerase, which prevents non-specific amplification during cDNA synthesis. Thermo-Start™ has 5' to 3' polymerization and exonuclease activity but lacks 3' to 5' exonuclease activity (proofreading)**.**Thermo-Start requires an **activation step at 95°C for 15 minutes**.

RT Enhancer

RT Enhancer is included to remove contaminating DNA, eliminating the need for DNase I treatment. It degrades double stranded DNA during the transcription of RNA and is inactivated during the activation step of the Thermo-Start DNA Polymerase.

Verso Reverse Transcriptase

Verso is an RNA-dependent DNA polymerase with a significantly attenuated RNase H activity. Verso can synthesize long cDNA strands, up to 11 kb, at a temperature range of 42°C to 57°C. Verso can reverse transcribe total RNA from 1 pg - 1 µg. The recommended amount of total RNA template to use in 1-step kits is between 1 pg - 100 ng.

Storage Conditions

Store at -20°C until ready for use. Avoid repeated freeze thawing.

Additional Info

The use of disposable gloves, RNase and DNase free filter tips and plastics is recommended.
RT Enhancer is not required if DNase I treatment is performed prior to qRT-PCR.

Tips before use

Thaw the reagents on ice. Mix and spin down the solutions before use to recover the maximum amount. **Do not vortex the Verso Enzyme Mix or the 1-Step PCR Hot-Start Master Mix.**
Briefly centrifuge to avoid bubbles within the wells. Always include a no template control (NTC) and a no enzyme control (NEC).

Protocol

Example of reaction mix preparation.
The volume of each component is for a 50 µL final reaction.

	Volume	Final Concentration
Verso Enzyme Mix	1 µL	
2X 1-Step PCR Hot-Start Master Mix	25 µL	1X
RT Enhancer	2.5 µL	
Forward primer (10 µM)*	1 µL	200 nM
Reverse primer (10 µM)*	1 µL	200 nM
Template (RNA)**	1-5 µL	1 ng
Water, nuclease-free (#R0581)	To 50 µL	
Total volume	50 µL	

* For optimization, a primer titration should be performed from 50 nM to 500 nM final concentration. Scale up or down the volume and concentration as appropriate.
** The amount of total RNA added as a template should be between 1pg and 100 ng.

Example of a 1-Step RT-PCR thermal cycling program:

	Temp.	Time	Number of cycles
cDNA synthesis*	50°C	15 min	1 cycle
Verso inactivation	95°C	15 min	1 cycle
Denaturation	95°C	20 s	35-45 cycles
Annealing**	50-60°C	30 s	
Extension***	72°C	1 min	
Final extension	72°C	5 min	1 cycle

* Depending on the length of template and degree of secondary structure, the efficiency of the first strand synthesis maybe improved by optimizing temperature and time (42-57°C for 5-30 minutes).
** Annealing temperature depends on primer sequence.
*** Time of extension depends on the length of the amplicon. If the amplicon exceeds 1 kb amplification time should be adapted. Thermo-Start *Taq* DNA Polymerase extends at approximately 1 kb/min.

CERTIFICATE OF ANALYSIS

Verso 1-Step RT-PCR Hot-Start Kit is tested functionally for use in RT-PCR.

Quality authorized by:  Jurgita Zilinskiene

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- Use of this product is covered by one or more of the following US patents and corresponding patent claims outside the US: 6,127,155, 5,677,152 (claims 1 to 23 only) and 5,773,258 (claims 1 and 6 only). The purchase of this product includes a limited, non-transferable immunity from suit under the foregoing patent claims for using only this amount of product for the purchaser's own internal research. No right under any other patent claim and no right to perform commercial services of any kind, including without limitation reporting the results of purchaser's activities for a fee or other commercial consideration, is conveyed expressly, by implication, or by estoppel. This product is for research use only. Diagnostic uses under Roche patents require a separate license from Roche. Further information on purchasing licenses may be obtained by contacting the Director of Licensing, Applied Biosystems, 850 Lincoln Centre Drive, Foster City, California 94404, USA.
- Use of Verso 1-Step RT-PCR Hot-Start Kit is licensed from bioMerieux, is covered by US Patent 5,654,143, US RE39031 and equivalents, and is for Research Use Only.

PRODUCT USE LIMITATION

This product is developed, designed and sold exclusively *for research purposes and in vitro use only*. The product was not tested for use in diagnostics or for drug development, nor is it suitable for administration to humans or animals.
Please refer to www.thermoscientific.com/onebio for Material Safety Data Sheet of the product.