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# Thermo Scientific Verso 1-Step RT-PCR ReddyMix Kit

## Description

Verso<sup>TM</sup> 1-Step RT-PCR ReddyMix<sup>TM</sup> Kit supplies in only two vials, all the components required to perform a rapid, sensitive and reproducible RT-PCR for the detection and analysis of RNA.

- <u>Verso</u><sup>TM</sup> <u>Enzyme Mix</u> includes Verso<sup>TM</sup> 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.
- <u>1-Step PCR ReddyMix</u><sup>TM</sup> (2X), 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. ReddyMix includes a dye and precipitant to facilitate the visualization and gel loading.

Thermoprime Plus, a DNA Polymerase stable at high temperature.

<u>RT Enhancer</u> is included to remove contaminating DNA, eliminating the need for DNase I treatment.

### **Kit Contents**

Vial	Pack Size (cap color)	
	А	В
Verso Enzyme Mix	40µl (black)	200µl (black)
1-Step PCR ReddyMix (2X)	1 ml (red)	5 x 1ml (red)
RT Enhancer	100µl (yellow)	500µl (yellow)



#### INFORMATION

## **Verso<sup>TM</sup> Reverse Transcriptase**

Verso<sup>TM</sup> is an RNA-dependent DNA polymerase with a significantly attenuated RNase H activity compared to *Reverse*-iT<sup>TM</sup>. Verso<sup>TM</sup> can synthesize long cDNA strands, up to 11 kb, at a temperature range of 42°C to 57°C. Verso<sup>TM</sup> can reverse transcribe total RNA from 1 pg - 1  $\mu$ g. The recommended amount of total RNA template to use in 1-step kits is between 1 pg - 100 ng.

#### **Thermoprime Plus DNA Polymerase**

Thermoprime Plus has 5' to 3' polymerization and exonuclease activity but lacks 3' to 5' exonuclease activity (proofreading).

## **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 after 2 minutes at 95°C.

#### **Storage Conditions**

Store at  $-20^{\circ}$ C until ready for use. Verso<sup>TM</sup> 1-Step RT-PCR ReddyMix<sup>TM</sup> Kit is stable for a minimum of 12 months. Avoid repeated freeze thawing. Shipped on ice within the UK and on dry ice for international and within the US.

#### **Additional Info**

- The use of disposable gloves, RNase and DNase free filter tips and plastics is recommended.
- If DNase I treatment has been performed, RT Enhancer is not required.

#### 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 ReddyMix.

Briefly centrifuge to avoid bubbles within the wells. Always include a no template control (NTC) and a no enzyme control (NEC).





#### AB-1454-LD

## 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	
	1-Step PCR ReddyMix (2X) <sup>a</sup>	25 µl	1X
Reaction	Forward primer (10 $\mu$ M) <sup>b</sup>	1 µl	200 nM
Mix	Reverse primer $(10 \mu\text{M})^{b}$	1 µl	200 nM
	RT Enhancer	2.5 μl	
	Water (PCR grade) <sup>c</sup>	variable	
	Template (RNA) <sup>d</sup>	1 - 5 μl	1 ng
	Total volume	50 µl	

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

	Temp.	Time	Number of cycle
cDNA Synthesis <sup>e</sup>	50°C	15 min	1 cycle
Verso inactivation	95°C	2 min	1 cycle
Denaturation	95°C	20 sec	
Annealing <sup>f</sup>	50-60°C	30 sec	35 - 45 cycles
Extension <sup>g</sup>	72°C	1 min	
Final extension	72°C	5 min	1 cycle

#### Notes

- a The gel precipitant in 1-Step PCR ReddyMix causes a slight increase in the thermal mass of the reaction mix. In a small number of cases this may necessitate some minor re-optimization of the thermal cycler program. If this is the case we suggest increasing the temperature of the denaturation step by 1-2°C and decreasing the temperature of the annealing step by 1-2°C. Alternatively, increase the duration of each step by 5-10 seconds.
- b 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.
- c The volume of the total reaction should be completed up to 50  $\mu$ l with water.
- d The amount of total RNA added as a template should be between 1 pg and 100 ng.
  e 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).
- F Annealing temperature depends on primer sequence.
- g Time of extension depends on the length of the amplicon. If the amplicon exceeds 1 kb amplification time should be adapted (Thermoprime Plus DNA Polymerase extends at approximately 1 kb/min).

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## **Quality control**

Verso<sup>TM</sup> 1-Step RT-PCR ReddyMix<sup>TM</sup> Kit is tested functionally for use in RT-PCR.

## **Ordering Information**

AB-1454/LD/A	Verso <sup>™</sup> 1-Step RT-PCR ReddyMix <sup>™</sup> Kit	40 x 50 µl rxns
AB-1454/LD/B	Verso <sup>TM</sup> 1-Step RT-PCR ReddyMix <sup>TM</sup> Kit	200 x 50 µl rxns

## **Related Products**

Cat. No.	Description	Quantity
AB-2400 AB-2400/W AB-2800 AB-2800/W AB-0745 AB-0626 AB-0558 AB-0386 AB-0783	ABgene <sup>®</sup> SuperPlate <sup>™</sup> Semi-Skirted 96-Well PCR Plate ABgene <sup>®</sup> SuperPlate <sup>™</sup> Semi-Skirted 96-Well PCR Plate, white ABgene <sup>®</sup> SuperPlate <sup>™</sup> Skirted 96-Well PCR Plate ABgene <sup>®</sup> SuperPlate <sup>™</sup> Skirted 96-Well PCR Plate, White Easy Peel (heat seal) Adhesive PCR Foil Adhesive PCR Film Strips of 8 Domed Caps Strips of 8 Flat Caps	25 plates 25 plates 25 plates 25 plates 100 sheets 100 sheets 120 strips 120 strips

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