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Thermo Scientific Verso 1-Step RT-PCR ReddyMix Kit

Description

VersoTM 1-Step RT-PCR ReddyMixTM 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>TM <u>Enzyme Mix</u> includes VersoTM 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>TM (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

VersoTM Reverse Transcriptase

VersoTM is an RNA-dependent DNA polymerase with a significantly attenuated RNase H activity compared to *Reverse*-iTTM. VersoTM can synthesize long cDNA strands, up to 11 kb, at a temperature range of 42°C to 57°C. VersoTM can reverse transcribe total RNA from 1 pg - 1 μ 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° C until ready for use. VersoTM 1-Step RT-PCR ReddyMixTM 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) ^a	25 µl	1X
Reaction	Forward primer (10 μ M) ^b	1 µl	200 nM
Mix	Reverse primer $(10 \mu\text{M})^{b}$	1 µl	200 nM
	RT Enhancer	2.5 μl	
	Water (PCR grade) ^c	variable	
	Template (RNA) ^d	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 ^e	50°C	15 min	1 cycle
Verso inactivation	95°C	2 min	1 cycle
Denaturation	95°C	20 sec	
Annealing ^f	50-60°C	30 sec	35 - 45 cycles
Extension ^g	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 μ 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

VersoTM 1-Step RT-PCR ReddyMixTM Kit is tested functionally for use in RT-PCR.

Ordering Information

AB-1454/LD/A	Verso [™] 1-Step RT-PCR ReddyMix [™] Kit	40 x 50 µl rxns
AB-1454/LD/B	Verso TM 1-Step RT-PCR ReddyMix TM 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 [®] SuperPlate [™] Semi-Skirted 96-Well PCR Plate ABgene [®] SuperPlate [™] Semi-Skirted 96-Well PCR Plate, white ABgene [®] SuperPlate [™] Skirted 96-Well PCR Plate ABgene [®] SuperPlate [™] 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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