

Reversease 200 U/μl (M-MuLV, RNase H-)

Description: Reversease is a M–MuLV Reverse Transcription enzyme for fast and specific generation of cDNA. This enzyme is a new genetically engineered variant with improved thermal stability up to 60 °C and is suitable for reverse transcription of RNA templates with complex secondary structures. Due to enhanced affinity to the template, it can be used in assays with a small amount of template and low-copy genes. This Reversease can amplify cDNA up to 10 kb. Due to the improved purity, it is also suitable for pathogen detection assays. Reversease is supplied with optimized Reaction Buffer for best results.

Concentration: 200 units/μl

Storage: -18 °C to – 22 °C

REF	105600	105650	colour
Reversease	10 000 units	50 000 units	blue
5x Complete Reaction Buffer	1x 250 μl	5x 250 μl	yellow

Application: First-strand synthesis of cDNA from RNA or single-stranded DNA. Reversease shows a synthesis capacity up to 10 kb cDNA product, which can be amplified by PCR with a Taq Polymerase afterwards.

For one-step RT-qPCR please use 10 - 15 U of the enzyme for a 20 μl reaction system.

Unit definition: One unit is defined as the amount of enzyme required to incorporate 1 nmol of dTTP into an acid-insoluble form in 10 minutes at 37°C using poly(rA)/ oligo(dT) as template primer.

Please see next page for recommended standard protocol for Reverse Transcription.



- Avoid frequent freeze/thaw of the reagents and final cDNA.
- Do not use more than 10 μl for downstream applications such as qPCR.
- Reversease can be combined with SuperHotTaq REF 129002 or SmartHotTaq REF 102020 for a one-step RT-qPCR assay.

Recommended Standard Protocol:

Component	20 µl Reaction	Final Concentration
Sample	10 µl	total RNA: 10 pg - 5 µg purified mRNA: 10 pg – 500 ng
Primer (100 µM)	1 µl	5 µM *
5x Reaction Buffer	4 µl	1x
dNTP Mix (10 mM)	0.5 µl	500 µM -
Optional: RNase Inhibitor (40 U/µl)	1 µl	1 U/µl
Reversease 200 U/µl	1 µl	200 U
RNase-free Water	adjust to 20 µl final volume	--

* Oligo(dT) Primer or Random Primer or a 1:1 mix of both primers. For gene specific primers we recommend 500 nM final primer concentration.

Thermocycler protocol

Step	Time	Temperature
Reverse Transcription	15 minutes	42 °C *
Optional: Heat Inactivation	2 - 5 minutes	85 °C

* The time for reverse transcription reaction depends on the cDNA length. For difficult templates please increase reverse transcription time to 30-45 min and the temperature to 50 °C. When using Random Primers, a incubation time of 5 min at 25 °C might be necessary beforehand.