Product Description

AcuGenix™ LongR One-Step RT-PCR Kit is a premium closed-tube amplification solution optimized for long RNA templates. It integrates reverse transcription (RT) and high-fidelity PCR into a single workflow via a synergistically engineered long-range enzyme cocktail (derived from directed evolution) and a pre-optimized buffer system tailored for long-amplicon synthesis, eliminating intermediate purification or tube-opening steps to minimize aerosol contamination risks.

Rigorously batch-validated, the kit delivers industry-leading performance for long-template amplification: stable synthesis and amplification of targets >10 kb, an ultra-fast (20 sec/kb) extension rate, robust detection from as low as 10 pg of high-abundance total RNA input, and RT workflow completion (from reaction assembly to amplification) within 30 minutes-representing a >2-fold efficiency improvement over conventional long-range one-step RT-PCR kits.

The kit features a thermostable long-range reverse transcriptase with exceptional processivity and RNA secondary structure unwinding activity, enabling efficient full-length cDNA synthesis across GC-rich or structurally complex regions. It is paired with an antibody-blocked hot-start high-fidelity DNA polymerase retaining 3′→5′ exonuclease proofreading activity, delivering a fidelity approximately 50-fold higher than that of wild-type Taq DNA polymerase to minimize mismatch risks in long-amplicon synthesis. The pre-optimized buffer is supplemented with long-range amplification enhancers and background suppressors to effectively inhibit primer-dimer formation and non-specific amplification, enabling uniform amplification of long targets without laborious primer titration.

Each batch is strictly quality-controlled to be free of RNase, DNase, and extraneous nucleic acid contamination, with an inter-lot coefficient of variation (CV) <5% for amplification efficiency and specificity. The kit is suitable for applications including full-genome sequencing of long RNA viruses, full-length transcript analysis, lncRNA/circRNA amplification, and full-length cDNA synthesis for single-cell sequencing. ***For research use only.***

Components

Components	BR3M321-03 (24 T)	BR3M321-06 (96 T)
2× One Step Buffer	300 µL	1,200 µL
One Step Enzyme Mix	72 µL	288 µL

Storage

Store at -20±5℃.

Notes

1. For Research Use Only.
2. Use RNase-free consumables and regularly decontaminate the experimental area to prevent RNase contamination.
3. Other reagents required for amplification are not included and must be prepared by the user or purchased separately.

Protocol

1. Reaction Setup

1.1 Thaw **2× One Step Buffer** on ice, mix thoroughly, and briefly centrifuge before use. Gently mix the **One Step Enzyme** Mix to avoid foaming, centrifuge, and keep on ice. Prepare the reaction mixture in an RNase-free microcentrifuge tube according to **Table 1**.

Table 1. RT-PCR Reaction Setup

Components	Volume (µL)	Final Concentration
2× One Step Buffer	12.5	1×
One Step Enzyme Mix	3	
Gene Specific Primer Forward (10 µM)	1	0.4 µM
Gene Specific Primer Reverse (10 µM)	1	0.4 µM
RNA Template	X	
RNase-free ddH_2O	Up to 25	

Note: The total reaction volume can be adjusted based on experimental requirements; adjust all components

proportionally.

2. Amplification Program

2.1 Gently mix by pipetting, briefly centrifuge to collect the reaction mixture at the bottom of the tube, and perform the following reaction on a PCR instrument:

Table 2. Amplification Program

Step	Temperature	Time	Cycles
Heated Lid	105℃	—	—
Reverse Transcription	50℃	30 min	1
Initial Denaturation	95℃	3 min	1
Denaturation	95℃	20 sec	30-35
Annealing	60℃ [a]	15 sec	
Extension	72℃	20 sec-1 min/kb [b]	
Hold	4℃	∞	1

Note a: Adjust the annealing temperature according to the primer's Tm values (typically set 1-2℃ lower than the primer annealing temperature).

Note b: Adjust the extension time as needed. Generally, increasing the extension time helps to improve amplification yield.

2.2 Analyze the amplification products by agarose gel electrophoresis.