

RT-qPCR Extraction Control Red

Catalogue No.: abx461007

RT-qPCR Extraction Control Red (emission wavelength 670 nm). This product consists of artificial cells containing an internal control RNA sequence with no known homology to any organism, and a specific primer/probe control mix for PCR detection. It can be used to monitor the RNA extraction process in RT-qPCR assays.

Target: RT-qPCR Extraction Control Red

Tested PCR

Applications:

Form: Liquid

Storage: Store at -80°C. Avoid repeated freeze/thaw cycles.

Shelf Life: Up to 12 months.

Buffer: The exact formulation is proprietary.

Kit Components:

Component	Volume
Internal Control RNA	5 × 200 µl
25X Control Mix	5 × 100 µl
50 mM MgCl ₂	1.2 ml

Note: THIS PRODUCT IS FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC, THERAPEUTIC OR COSMETIC PROCEDURES. NOT FOR HUMAN OR ANIMAL CONSUMPTION.

For Reference Only

Directions for use: All reactions should be carried out at room temperature unless stated otherwise.

Extraction Steps:

1. Spin down all tubes before opening.
2. Add 2 µl Internal Control RNA into each sample.
3. Follow the manufacturer's protocol for total RNA extraction.
4. Elute total RNA to a volume of 100 µl.
5. Use 5 µl of the elution volume for a 20 µl reaction volume.

Recommended reagent volumes per 20 µl qPCR mix:

Component	Volume
RT-qPCR Mix with 3 mM MgCl ₂ (not provided)	10 µl
Target Probe/Primer Mix (not provided)	Variable
Extracted RNA Template (not provided)	Variable
25X Control Mix	0.8 µl
<i>Vortex the Control Mix tube before use</i>	
50 mM MgCl ₂	1.2 µl
Reverse Transcriptase (not provided)	0.2 µl
RNase Inhibitor (not provided)	0.4 µl
Total Volume	20 µl

Assay Setup:

Step	Number of Cycles	Temperature	Time per Cycle
Reverse Transcription	1 cycle	42 °C	10-20 min
Activation	1 cycle	95 °C	3 min
Denaturation	30-40 cycles	95 °C	10 seconds
Annealing/Extension/Acquisition		60 °C	30-45 seconds

Notes:

- The fluorescence signal for the RNA Internal Control can be observed at 670 nm.
- It is recommended that the end user carries out a validation step to ensure that the primers used do not cross-react with the Internal Control RNA.
- Ct values of the Internal Control may vary due to elution volumes of nucleic acid and reagents used.
- The optimal conditions will vary from reaction to reaction and are dependent on the system used.
- Each parameter needs to be adjusted by the end user and some optimization may be required. For example, the standard annealing temperature is 60 °C, though this may need to be optimized by the user depending on the RT-qPCR mix used.