

Tel: Email: Web: +001.301.271.5445 info@expressbiotech.com www.expressbiotech.com

## Certificate of Analysis

## Reovirus PCR Positive Control

**Date:** May 24, 2011

Catalog Number: PCRPC136

Volume: 250µl, about 250 reactions

RNA Concentration: 200 ng/µl in TE buffer. Use 0.5 to 1.0 µl per RT-PCR reaction.

**Storage conditions:** -20° C to -70° C.

**Expiration:** 3 years from date of shipment

**First Strand Reaction:** can be primed with anti-sense REV oligo at 1 μl of a 1 μM solution.

Typical PCR reaction: Total volume 50 µl, 25 µl water, 5 µl 10x PCR Buffer, 2 µl 50

mM MgSO<sub>4</sub>, 1  $\mu$ l 10 mM dNTP mix, 1  $\mu$ l 10  $\mu$ M Forward Primer, 1  $\mu$ l 10  $\mu$ M Reverse Primer, DNA 0.5 to 1  $\mu$ l, add water to 49.5  $\mu$ l, and add last 0.5  $\mu$ l Taq Polymerase. Mix, spin down gently, and put tubes in thermal cycler. Alternatively, a master mix including everything except some water and DNA may be used.

## **Typical Thermal Cycler Program:**

- 1. 94°C; 2 min
- 2. 94°C; 15 sec
- 3. 55°C; 30 sec
- 4. 0.5°/sec; to 68°C
- 5. 68°C; 2 min
- 6. Go to 2; 35 min
- 7. 72°C, 5 min
- 8. 4°C; forever
- 9. End

PCR Primers: FOR 5'-GCT ATT TTT GCC TCT TCC C-3'

**REV 5'-GAT GAA TGG AGC CTG TCC C-3'** 

FIRST STRAND 5'-CTA CTT ACC TCG GAC AGG G-3'

**References:** Leary, TP, J. Clinical Mircobiology **40:4**, April 2002, pages 1368-1375.

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