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Certificate of Analysis

Mouse Hepatitis Virus

PCR Positive Control

Catalog Number: PCRPC105

Date: August 2012

Volume: 100 µl, 250-500 PCR reactions.

RNA Concentration: 200 ng/µl in low TE buffer or 20 µg RNA, enough for 20 first strand cDNA reactions.

Specific Activity: At least 500 copies per µl.

Storage conditions: -70º C

First Strand Reaction: Prime with random hexameres, oligo dT, or anti-sense MHV reverse primer oligo at 1 µl of a 2 µM solution in a standard first strand cDNA synthesis reaction. Use 0.5 to 1.0 µl of the first strand reaction cDNA per RT-PCR reaction. Caution: too much first strand cDNA inhibits the PC reaction.

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

mM $MgSO_4$, 1 µl 10 mM dNTP mix, 1 µl 10 µM Forward Primer, 1 µl 10 µM Reverse Primer, DNA 0.5 to 1 µl, add water to 49.5 µl, and add last 0.5 µl *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 times
7.	72°C	5 min
8.	4°C	forever
9.	End	

PCR Primers: FOR 5'-GTC ATG AGG CTA TTC CTA CTA-3'

REV 5'-ATA CAC ATC TTT GGT GGG-3'

Anti-sense REV 5'-CCC ACC AAA GAT GTG TAT-3'

References: Comparative Medicine **54:4**, August 2004, page 384.

This product is distributed for laboratory research use only. The safety and efficacy of this product in diagnostic or other clinical uses has not been established.