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Certificate of Analysis Simian Virus 5 **PCR Positive Control**

Date: May 24, 2011

Catalog Number: PCRPC124

Volume: 250µl, about 250 reactions

RNA Concentration: 200 ng/µl in TE buffer. Use 0.5 to 1.0 µl per first strand reaction.

Storage conditions: -20° C to -70° C

Expiration: 3 years from date of shipment

First Strand Reaction: primed with oligo dT or 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 MgSO₄, 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 min
- 7. 72°C, 5 min
- 8. 4°C: forever
- 9. End

PCR Primers: FOR 5'-GAC ACA AAA ATG TCA TCC GTG-3'

REV 5'-CGG CTA CCA TCC GTC CTT GG-3'

References: Tsurudome, M. Journal General Virology **72:9** 1991, pages 2289-2292.

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