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Certificate of Analysis Kilham Rat Virus PCR Positive Control

Date: May 24, 2011

Catalog Number: PCRPC113

Volume: 250µl, about 250 reactions

DNA Concentration: 150 ng/µl in TE buffer. Use 0.5 to 1.0 µl per PCR reaction.

Storage conditions: -20° C to -70° C

Expiration: 3 years from date of shipment

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

mM MgSO₄, 1 μ l 10 mM dNTP mix, 1 μ l 10 μ M Forward Primer, 1 μ l 10 μ M Reverse Primer, DNA 0.5 to 1 μ l, add water to 49.5 μ l, and add last 0.5 μ 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'-GCA CAG ACA ACC AAA CAG GAA CTC TCC-3'

REV 5'-AGT CTC ACT TTG AGC GGC TG-3'

References: Besselsen, DG, et.al. J. Clinical Microbiology 33:7 July 1995, pages 1699-1703.

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