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Certificate of Analysis

Pneumocystis carinii

PCR Positive Control

Catalog Number: PCRPC400

Date: August 2012

Volume: 250 µl, 250 PCR Reactions

DNA Concentration: 25 ng/µl in TE buffer. Use 0.5 to 1.0 µl per PCR reaction. CAUTION: use 10-50 ng of purified DNA per PC Reaction, too much DNA inhibits the reaction.

Specific Activity: At least 500 copies per µl.

Storage conditions: -20° C to -70º C

Typical PCR 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-1 5'-AGT TAC GGC CAT ACC TCA GA-3'

REV-1 5'-AAA GCT ACA GCA CGT CGT AT-3'

FOR-2 5'-ATT TAT GGG TTT CAA TGG-3'

REV-2 5'-GTT CCC TTT AAT ATT GCA-3'

References: 1. Saito, K. et al. Rheumatology **43:4,** pages 479-485, 2004. and 2. Weisbroth, S.H., et al,. J. Clinical Microbiology **37:5,** pages 1441-1446, 1999.

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