

Certificate of Analysis

Cilia-Associated Respiratory Bacillus

PCR Positive Control

Date: May 24, 2011

Catalog Number: PCRPC118

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

Cilia-associated respiratory (CAR) bacillus: Is an unclassified, gram negative, filamentous bacterium that was tentatively designated in 1985 as one of the agents that cause chronic respiratory diseases in laboratory rodents.

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 Cyclor 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'-GAG TTT GAT CCT GGC TCA G-3'

REV 5'-AGA AAG GAG GTG ATC CAG CC-3'

References: Kawano, A., *J. Vet. Med. Sci.* **62:7**, 797-800, 2000.

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