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Certificate of Analysis

Corynebacterium kutscheri

PCR Positive Control

Catalog Number: PCRPC411

Date: November 2018

Volume: 250 µl, 250 PC Reactions

DNA Concentration: 300 pg/µl of DNA in TE buffer. Use 0.5 to 1.0 µl per PCR 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

PCR Primer sequences (5'-3'): Mouse 16S RNA, For GTGGGTGATCTGCCCCTAAC, Rev CCCCGGAATTTCACAGACGA, Product 477 bases. RpoB, For CGTGAACGTATGACCACC-CA, Rev, ACATACGGCCATAGTGCGAG, Product 250 bases.

Rat 16S RNA, For TCGTCTGTGAAATTCCGGGG, Rev CGGCAGTCTCTCATGAGTCC, Product 568 bases. RpoB, For AAGATCGCTTCGTGGTTGGT, Rev TACCACACCGGACTTC-TTGC, Product 344 bases.

References: Khamis, A. Raoult, D. and La Scola, B. *rpo*B Gene Sequencing for Identification of *Corynebacterium* Species. J. Clinical Microbiology **42:9** 3925-3931, 2004.