

Certificate of Analysis for NR-19819

Leptospira interrogans, Strain M874, LA0615 Mutant (Serovar Manilae)

Catalog No. NR-19819

Product Description: Leptospira interrogans (L. interrogans), strain M874 (serovar Manilae) is a transposon mutant of wild-type strain L495 created by disruption of the gene encoding conserved hypothetical protein LA0615, which is located downstream of the gene encoding LipL41 in strain L495

Lot¹: 59581726 Manufacturing Date: 06JAN2011

| TEST | SPECIFICATIONS | RESULTS |
|--|--|---|
| Phenotypic Analysis Colony morphology | Growth below the soft agar surface (Dinger's disk) | Growth below the soft agar surface (Dinger's disk) ² |
| Genotypic Analysis Sequencing of 16S ribosomal RNA gene (~ 1340 base pairs) | Consistent with <i>L. interrogans</i> | Consistent with <i>L. interrogans</i> ³ |
| Viability (post-vialing) Visual observation LIVE/DEAD [®] BacLight [™] Bacterial Viability | Growth Green fluorescence visible | Growth ² Green fluorescence visible ⁴ |

¹L. interrogans, strain M874 (serovar Manilae) was deposited by Ben Adler, Professor of Microbiology, Monash University, Clayton, Victoria, Australia. The deposited material was inoculated into EMJH semisolid agar (0.15%) and incubated for 10 days at 30°C in an aerobic atmosphere. The material from the initial growth was passaged twice in EMJH semisolid agar (0.15%) for 18 days and 17 days, respectively, at 30°C in an aerobic atmosphere to produce this lot.

Date: 31 JAN 2013 Signature:

Title: Technical Manager, BEI Authentication or designee

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

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² Disk of dense growth below the soft agar surface (Dinger's disk) (Czekalowski, J. W., J. W. McLeod and J. Rodican. "The Growth and Respiration of *Leptospira* in Solid or Semi-Solid Media with Special Reference to Dinger's Phenomenon." <u>Br. J. Exp. Pathol.</u> 34 (1953): 588-595.) was evident after 15 days at 30°C in EMJH semisolid agar (0.15%).

³Also consistent with other *Leptospira* species

⁴Determined after 15 days incubation under cultivation conditions with LIVE/DEAD[®] BacLight[™] Bacterial Viability Kit, 100x magnification (Invitrogen[™] L34856). Cells with a compromised membrane that are dead or dying will stain red, while cells with an intact membrane will stain green.