

Certificate of Analysis for NR-48085

***Staphylococcus aureus* subsp. *aureus*, Strain JE2, Transposon Mutant NE1543 (SAUSA300_0961)**

Catalog No. NR-48085

Product Description: *Staphylococcus aureus* (*S. aureus*) subsp. *aureus*, transposon mutant NE1543 was derived from *S. aureus* subsp. *aureus*, strain JE2. Mutagenesis occurred through the use of the *mariner*-based transposon *bursa aurealis* resulting in an erythromycin-resistant deletion strain of JE2. *S. aureus* subsp. *aureus*, transposon mutant NE1543 was created by disruption of *qoxC*, one of four quinol oxidase genes in the *qoxABCD* operon that encode for the terminal oxidase cytochrome aa₃. Strain JE2 is a plasmid-cured derivative of strain LAC that was isolated in 2002 from a skin and soft tissue infection of an inmate in the Los Angeles County Jail in California, USA.

Lot¹: 63324007

Manufacturing Date: 25FEB2015

| TEST | SPECIFICATIONS | RESULTS | |
|---|---------------------------------------|--|---|
| Phenotypic Analysis² | | | |
| Cellular morphology Colony morphology ³ | Gram-positive cocci Report results | Colony type 1 Gram-positive cocci Circular, convex, entire, smooth and yellow (Figure 1) Non-motile | Colony type 2 Gram-positive cocci Circular, convex, entire, smooth and cream (Figure 1) Not reported |
| Motility (wet mount) | Report results | | |
| Confirmation of Transposon Insertion⁴ | Resistant to erythromycin | Resistant to erythromycin | |
| Purity (post-freeze)⁵ | Consistent with <i>S. aureus</i> | Consistent with <i>S. aureus</i> | |
| Viability (post-freeze)³ | Growth | Growth | |

¹NR-48085 was produced by inoculation of the deposited material into Tryptic Soy broth with 5 µg/mL erythromycin and incubated for 24 hours at 37°C in an aerobic atmosphere. Broth inoculum was added to Tryptic Soy agar with 5 µg/mL erythromycin kolles which were grown 20 hours at 37°C in an aerobic atmosphere to produce this lot.

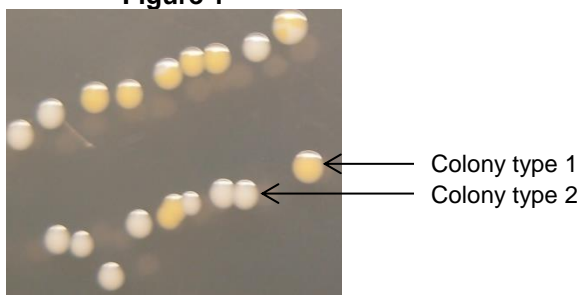
²Two colony types were observed. VITEK[®] MS (MALDI-TOF) analysis identified cells from both colony types as *S. aureus*. Since NR-48085 is a transposon mutant and is expected to produce a single colony type, colony purification of this item is highly recommended.

³24 hours at 37°C in an aerobic atmosphere on Tryptic Soy agar with 5 µg/mL erythromycin

⁴Prior to initiating work, it is recommended that the presence and location of the transposon is confirmed. Gene specific primers should be paired with either the "Upstream" primer (5'-CTCGATTCTATTAACAAGGG-3') for transposons in the "plus" orientation or the "Buster" primer (5'-GCTTTTCTAAATGTTTTTAAGTAAATCAAGTAC-3') for transposons in the "minus" orientation. For additional information, refer to Fey, P. D., et al. "A Genetic Resource for Rapid and Comprehensive Phenotype Screening of Nonessential *Staphylococcus aureus* Genes." MBio 4 (2013): e00537-12. PubMed: 23404398.

⁵Purity of this lot was assessed for 7 days at 37°C in an aerobic atmosphere on Tryptic Soy agar with 5% defibrinated sheep blood.

Figure 1



Certificate of Analysis for NR-48085

Date: 20 APR 2015

Signature:



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