Certificate of Analysis for NR-47166

Staphylococcus aureus subsp. *aureus*, Strain JE2, Transposon Mutant NE623 (SAUSA300_2570)

Catalog No. NR-47166

Product Description: *Staphylococcus aureus* (*S. aureus*) subsp. *aureus,* transposon mutant NE623 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 NE623 was created by disruption of *arcA,* which encodes for an arginine deiminase and is a member of the arginine deiminase (ADI) pathway genes *arcABDC* that are important for the utilization of arginine as a source of energy for growth under anaerobic conditions. 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.

Lot[1]: 63529651 **Manufacturing Date: 29MAY2015**

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis		
Cellular morphology	Gram-positive cocci	Gram-positive cocci
Colony morphology[2]	Report results	Circular, convex, entire, smooth and yellow (Figure 1)
Motility (wet mount)	Report results	Non-motile
Confirmation of Transposon Insertion[3]	Resistant to erythromycin	Resistant to erythromycin
Purity (post-freeze)[4]	Growth consistent with *S. aureus*	Growth consistent with *S. aureus*
Viability (post-freeze)[2]	Growth	Growth

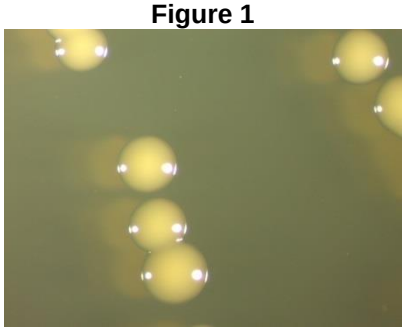
[1]NR-47166 was produced by inoculation of the deposited material into Tryptic Soy broth with 5 µg/mL erythromycin and incubated for 1 day at 37°C in an aerobic atmosphere. Broth inoculum was added to Tryptic Soy agar with 5 µg/mL erythromycin kolles which were grown for 1 day at 37°C in an aerobic atmosphere to produce this lot.

[2]1 day hours at 37°C in an aerobic atmosphere on Tryptic Soy agar with 5 µg/mL erythromycin

[3]Prior 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'-GCTTTTTCTAAATGTTTTTTAAGTAAATCAAGTAC-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.

[4]Purity 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



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Date: 24 JUN 2015 **Signature:**

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