

Certificate of Analysis for NR-55219

Staphylococcus aureus, Strain AJUL5

Catalog No. NR-55219

Product Description:

Staphylococcus aureus (S. aureus), strain AJUL5 is deposited as a kanamycin-, neomycin- and tobramycin-resistant strain derived from *S. aureus*, strain SH1000 through introduction of plasmid pSK5487M containing the gene *aph*(3')-Illa (encoding aminoglycoside O-phosphotransferase) and a chloramphenicol resistance gene *(cat)* for selection. NR-55219 was produced by resuspension of a lyophilized vial of deposited material in Tryptic Soy broth. Broth inoculum was added to Tryptic Soy broth containing 25 µg per mL chloramphenicol and grown for 1 day at 37°C in an aerobic atmosphere. The material from the initial growth was added to Tryptic Soy agar containing 25 µg per mL chloramphenicol kolles, which was grown for 1 day at 37°C in an aerobic atmosphere to produce this lot. Quality control testing was completed under propagation conditions unless otherwise noted.

Lot: 70052530 Manufacturing Date: 05MAY2022

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| TEST | SPECIFICATIONS | RESULTS |
|---|----------------------------------|---|
| Phenotypic Analysis | | |
| Cellular morphology | Gram-positive cocci | Gram-positive cocci |
| Colony morphology | Report results | Circular, convex, entire, smooth and yellow |
| Motility (wet mount) | Report results | Non-motile |
| Hemolysis | Report results | β-hemolytic |
| Catalase | Positive | Positive |
| VITEK® MS (MALDI-TOF) | S. aureus | S. aureus (99.9%) |
| Antibiotic Susceptibility Profile ¹ Etest [®] antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar | | |
| Kanamycin | Resistant | Resistant (≥ 256 µg per mL) |
| Tobramycin | Resistant | Sensitive (0.09 µg per mL) ² |
| Thermo Scientific™ Susceptibility Test Disc 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar | | |
| Neomycin | Resistant | 76 mm ³ |
| Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ⁴ Next-Generation Sequencing (NGS) analysis for antimicrobial resistance genes ⁶ | ≥ 70% for species identification | S. aureus (99.4%) ⁵ |
| Kanamycin | Resistant | Resistant |
| Neomycin | Resistant | Resistant |
| Tobramycin | Resistant | Inconclusive ⁷ |
| Purity (post-freeze) 8 days at 37°C in an aerobic atmosphere with and without 5% CO ₂ on Tryptic Soy agar with 5% defibrinated sheep blood | | Growth consistent with expected colony morphology |
| Viability (post-freeze) | Growth | Growth |

¹Minimum Inhibitory Concentration (MIC); Clinical & Laboratory Standards Institute (CLSI) M100-S28 (2018) or European Committee on Antimicrobial Susceptibility Testing (EUCAST) (2023)

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²S. aureus, strain AJUL5 was deposited as resistant to tobramycin, but showed a MIC of 0.09 μg per mL (interpreted as sensitive) for this antibiotic during QC testing.

³No CLSI or EUCAST interpretations of this antibiotic for *S. aureus* are currently available.



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⁴Relatedness between bacterial strains has traditionally been determined using DDH. For additional information refer to Auch, A. F., et al. "Digital DNA-DNA Hybridization for Microbial Species Delineation by Means of Genome-to-Genome Sequence Comparison." <u>Stand. Genomic Sci.</u> 2 (2010): 117-134. PubMed: 21304684.

⁵The whole genome of *S. aureus*, strain AJUL5 (contig total length approximately 2.66 megabase pairs) was sequenced using the Illumina® MiSeq® system.

⁶In silico analysis of NGS data for antimicrobial resistance genes was performed using the Bacterial and Viral Bioinformatics Resource Center (BV-BRC), ResFinder and Pathogenwatch genome analysis tools.

7S. aureus, strain AJUL5 was deposited as resistant to tobramycin. In silico analysis determined the predicted phenotype of this antibiotic as sensitive and resistant, respectively. No data for this antibiotic is available in the BV-BRC.

/Sonia Bjorum Brower/ Sonia Bjorum Brower

30 JAN 2023

Technical Manager or designee, ATCC Federal Solutions

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