

Escherichia coli K-12, Strain IM08B

Catalog No. NR-49806

Product Description:

Escherichia coli (*E. coli*) K-12, strain IM08B is an insertion mutant produced in *E. coli* K-12, strain DC10B via recombination-mediated genetic engineering (recombineering), and contains the *hsdM* (methylase) and *hsdS* (specificity) genes from *Staphylococcus aureus* (*S. aureus*), strain NRS384 clonal complex 8 (CC8). NR-49806 was produced by inoculation of BEI Resources seed lot 63849806 into Tryptic Soy broth with 25 µg per mL streptomycin and grown for 1 day at 37°C in an aerobic atmosphere. Broth inoculum was added to Tryptic Soy agar with 25 µg per mL streptomycin kolles, which were 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: 70039948

Manufacturing Date: 11NOV2020

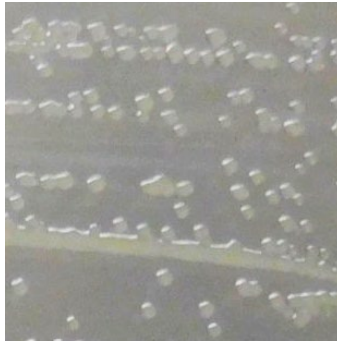
TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis Cellular morphology Colony morphology Motility (wet mount) VITEK® MS (MALDI-TOF)	Gram-negative rods Report results Report results <i>E. coli</i>	Gram-negative rods Circular, low convex, entire, smooth and cream (Figure 1) Motile <i>E. coli</i> (99.9%)
Genotypic Analysis Sequencing of 16S ribosomal RNA gene (~ 1480 base pairs)	≥ 99% sequence identity to <i>E. coli</i> K-12 strain (GenBank: NZ_CP014225.1)	99.5% sequence identity to <i>E. coli</i> K-12 strain (GenBank: NZ_CP014225.1) ¹
Analysis of <i>hsdMS</i> by PCR Assay^{2,3} <i>hsdM2</i> and <i>hsdS2</i> (CC8) <i>hsdS1</i> (CC8)	~ 3400 base pair amplicon ~ 1770 base pair amplicon	~ 3400 base pair amplicon ~ 1770 base pair amplicon
Analysis of <i>hsdMS</i> by Sequence Analysis^{2,3} <i>hsdM2</i> and <i>hsdS2</i> (CC8) <i>hsdS1</i> (CC8)	Consistent with depositor sequence Consistent with depositor sequence	Consistent with depositor sequence Consistent with depositor sequence
Confirmation of Streptomycin Resistance 1 day at 37°C in an aerobic atmosphere on Tryptic Soy agar with 25 µg per mL streptomycin	Growth	Growth
Purity 7 days at 37°C in an aerobic atmosphere with and without 5% CO ₂ on Tryptic Soy agar	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Viability	Growth	Growth

¹Also consistent with other *Escherichia* and *Shigella* species

²Performed on BEI Resources NRS-49806 lot 63849806

³PCR primers used for amplification were IM434 forward primer 5'-ACTTCTTTAAGGCTTAGAGTCAAGC-3', IM435 reverse primer 5'-TTTAACGCCACGTTCACTCTTTGC-3', 179 forward primer 5'-CGGCCATTTATACAGGAAAAGCCTA-3' and 180 reverse primer 5'-GTTACCTTCTATAGAGAGTGGTG-3'. For additional information, refer to Monk, I., et al. "Complete Bypass of Restriction Systems for Major *Staphylococcus aureus* Lineages." *mBio* 26 (2015): e00308-15. PubMed: 26015493.

Figure 1: Colony Morphology



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Program Manager or designee, ATCC Federal Solutions

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