

***Escherichia coli* K-12, Strain IM08B**

**Catalog No. NR-49806**

**Product Description:** *Escherichia coli* (*E. coli*) K-12, strain IM08B contains the *hsdM* (methylase) and *hsdS* (specificity) genes from *Staphylococcus aureus* NRS384 clonal complex 8 (CC8). This insertion mutant was produced in *E. coli* K-12, strain DC10B ( $\Delta dcm$ ). *E. coli* K-12, strain IM08B was deposited as resistant to streptomycin.

**Lot<sup>1</sup>: 70015547**

**Manufacturing Date: 01JUN2018**

TEST	SPECIFICATIONS	RESULTS
<b>Phenotypic Analysis</b> Cellular morphology Colony morphology <sup>2</sup>  Motility (wet mount) VITEK <sup>®</sup> 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%)
<b>Genotypic Analysis</b> Sequencing of 16S ribosomal RNA gene (~ 1460 base pairs)	≥ 99% sequence identity to <i>E. coli</i> K-12 strain (GenBank: NZ_CP014225.1)	99.9% sequence identity to <i>E. coli</i> K-12 strain (GenBank: NZ_CP014225.1) <sup>3</sup>
<b>Analysis of <i>hsdMS</i> by PCR Assay<sup>4,5</sup></b> <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
<b>Analysis of <i>hsdMS</i> by Sequence Analysis<sup>4,5</sup></b> <i>hsdM2</i> and <i>hsdS2</i> (~ 1710 base pairs) <i>hsdS1</i> (~ 1390 base pairs)	Consistent with depositor sequence Consistent with depositor sequence	Consistent with depositor sequence Consistent with depositor sequence
<b>Confirmation of Streptomycin Resistance<sup>2</sup></b>	Growth	Growth
<b>Purity (post-freeze)<sup>6</sup></b>	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
<b>Viability (post-freeze)<sup>2</sup></b>	Growth	Growth

<sup>1</sup>NR-49806 lot 70015547 was produced by the inoculation of BEI Resources NRS-49806 lot 63849806 into Tryptic Soy broth with 25 µg/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/mL streptomycin kolles, which were grown for 1 day at 37°C in an aerobic atmosphere to produce this lot.

<sup>2</sup>1 day at 37°C in an aerobic atmosphere on Tryptic Soy agar with 25 µg/mL streptomycin

<sup>3</sup>Also consistent with other *Escherichia* and *Shigella* species

<sup>4</sup>Performed on BEI Resources NRS-49806 lot 63849806

<sup>5</sup>PCR primers used for amplification were IM434 forward primer 5'-ACTTTCTTTAAGGCTTAGAGTCAAGC-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.

<sup>6</sup>Purity of this lot was assessed for 7 days at 37°C in an aerobic atmosphere with 5% CO<sub>2</sub> on Tryptic Soy agar with 5% defibrinated sheep blood.

Figure 1: Colony Morphology



/Heather Couch/  
Heather Couch

Program Manager or designee, ATCC Federal Solutions

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