

Antimicrobial Resistance Panel 11: *Escherichia coli* Resistance-Nodulation-Division (RND) Efflux Pumps Mutants

Catalog No. NR-55650

Product Description:

NR-55650 consists of a three-member panel of *Escherichia coli* (*E. coli*), strain BW25113 derivatives in which genes encoding Resistance-Nodulation-Division (RND) family efflux pumps were deleted. Additionally, one strain, NR-51862, was serially passaged in the presence of argyirin B, a small molecule inhibitor of LpxA, resulting in a substitution mutation in LpxA (Q73L) that exhibits reduced susceptibility to argyirin B. Each panel component listed in Table 1 was produced by inoculation of the deposited material into Tryptic Soy broth and grown for 1 day at 37°C in an aerobic atmosphere. Broth inoculum was added to Tryptic Soy agar kolles, which were grown for 1 day at 37°C in an aerobic atmosphere to produce each respective lot. Quality control testing was completed under propagation conditions unless otherwise noted.

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Table 1: Panel Components

| COMPONENT NUMBER | DESCRIPTION | GENOTYPE | LOT NUMBER | MANUFACTURING DATE |
|------------------|---|---|------------|--------------------|
| NR-51823 | <i>E. coli</i> , Strain NB27079-CDY0099 | Δ acrB, Δ acrD, Δ acrF, Δ emrB, Δ emrY, Δ entS, Δ mdtF, Δ mdtBC, Δ macB | 70043438 | 07APR2021 |
| NR-51883 | <i>E. coli</i> , Strain NB27079-CDY0154 | Δ acrAB::Km ^R , Δ acrD, Δ acrF, Δ emrB, Δ emrY, Δ entS, Δ macB, Δ mdtBC, Δ mdtF, | 70047261 | 15SEP2021 |
| NR-51862 | <i>E. coli</i> , Strain NB27079-TUP0093 | Δ acrAB::Km ^R , Δ acrD, Δ acrF, Δ emrB, Δ emrY, Δ entS, Δ macB, Δ mdtBC, Δ mdtF, LpxA(Q73L) | 70047263 | 09SEP2021 |

Table 2: *E. coli*, Strain NB27079-CDY0099 (NR-51923)

| TEST | SPECIFICATIONS | RESULTS |
|---|--------------------|--|
| Phenotypic Analysis | | |
| Cellular morphology | Gram-negative rods | Gram-negative rods |
| Colony morphology | Report results | Circular, convex, entire, smooth and cream (Figure 1a) |
| Motility (wet mount) | Report results | Motile |
| VITEK® MS (MALDI-TOF) | <i>E. coli</i> | <i>E. coli</i> (99.9%) |
| Antibiotic Susceptibility Profile | | |
| BD BBL™ Sensi-Disc™ susceptibility test disc ¹ | | |
| Gatifloxacin | Report results | 31 to 35 mm |
| Novobiocin | Report results | 25 mm |
| Etest® antibiotic test strips ¹ | | |
| Erythromycin | Report results | 1.5 to 2 µg/mL |
| Kanamycin | Report results | 1.5 µg/mL |
| Linezolid | Report results | 4 to 6 µg/mL |
| Rifampin | Report results | 3 µg/mL |
| Tetracycline | Report results | 0.75 µg/mL |
| Trimethoprim | Report results | 0.05 µg/mL |
| Sensititre™ System ² | | |
| Colistin | Report results | ≤ 0.025 µg/mL |

| TEST | SPECIFICATIONS | RESULTS |
|--|---|---|
| Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ³ Deletion of <i>acrB</i> , <i>acrD</i> , <i>acrF</i> , <i>emrB</i> , <i>emrY</i> , <i>entS</i> , <i>macB</i> , <i>mdtBC</i> and <i>mdtF</i> | ≥ 70% for species identification Deletions confirmed | <i>E. coli</i> (75.4%) Pending |
| Purity 7 days at 37°C in an aerobic atmosphere with | Growth consistent with expected colony morphology | Growth consistent with expected colony morphology |
| Viability | Growth | Growth |

¹1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar

²Sensititre™ System Gram Negative GN2F AST Plate (Thermo Scientific™)

³Relatedness 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." *Stand. Genomic Sci.* 2 (2010): 117-134. PubMed: 21304684. *E. coli*, strain DSM 30083 (GenBank: KK583188.1) was used for dDDH analysis.

Table 3: *E. coli*, Strain NB27079-CDY0154 (NR-51883)

| TEST | SPECIFICATIONS | RESULTS |
|---|--|--|
| Phenotypic Analysis Cellular morphology Colony morphology Motility BBL™ Motility Test Medium w/TTC Indicator for 1 day at 37°C in an aerobic atmosphere VITEK® MS (MALDI-TOF) | Gram-negative rods Report results Report results <i>E. coli</i> | Gram-negative rods Circular, convex, entire, smooth and cream (Figure 1b) Motile <i>E. coli</i> (99.9%) |
| Antibiotic Susceptibility Profile BD BBL™ Sensi-Disc™ susceptibility test disc ¹ Gatifloxacin Novobiocin Etest® antibiotic test strips ¹ Erythromycin Kanamycin Linezolid Rifampin Tetracycline Trimethoprim | Report results Report results Report results Report results Report results Report results Report results Report results | 32 to 34 mm 20 to 22 mm 30 µg/mL > 256 µg/mL 4 to 6 µg/mL 4 µg/mL 1 µg/mL 0.05 µg/mL |
| Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ² Deletion of <i>mdtBC</i> , <i>mdtF</i> , <i>macB</i> , <i>entS</i> , <i>emrY</i> , <i>emrB</i> , <i>acrF</i> , <i>acrD</i> , and <i>acrAB</i> | ≥ 70% for species identification Deletions confirmed | <i>E. coli</i> (75.2%) Pending |
| 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 |

¹1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar

²Relatedness 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." *Stand. Genomic Sci.* 2 (2010): 117-134. PubMed: 21304684. *E. coli*, strain DSM 30083 (GenBank: KK583188.1) was used for dDDH analysis.

Table 4: *E. coli*, Strain NB27079-TUP0093 (NR-51862)

| 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, convex, entire, smooth and cream (Figure 1c) Motile <i>E. Coli</i> (99.9%) |

| TEST | SPECIFICATIONS | RESULTS |
|---|--|---|
| Antibiotic Susceptibility Profile BD BBL™ Sensi-Disc™ susceptibility test disc ¹ Gatifloxacin Novobiocin Etest® antibiotic test strips ¹ Erythromycin Kanamycin Linezolid Rifampin Tetracycline Trimethoprim | Report results Report results Report results Report results Report results Report results Report results Report results | 35 to 36 mm 23 to 25 mm 0.064 µg/mL 96 µg/mL 1.0 to 1.5 µg/mL 0.125 µg/mL 0.25 µg/mL 0.023 µg/mL |
| Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ² Deletion of <i>mdtBC</i> , <i>mdtF</i> , <i>macB</i> , <i>entS</i> , <i>emrY</i> , <i>emrB</i> , <i>acrF</i> , <i>acrD</i> , and <i>acrAB</i> Confirmation of LpxA Gln73Leu SNP | ≥ 70% for species identification Deletions confirmed SNP confirmed | <i>E. coli</i> (> 75.2 %) Pending Pending |
| 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 |

¹1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar

²Relatedness 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." *Stand. Genomic Sci.* 2 (2010): 117-134. PubMed: 21304684. *E. coli*, strain DSM 30083 (GenBank: KK583188.1) was used for dDDH analysis.

Figure 1a: NR-51923 Colony Morphology

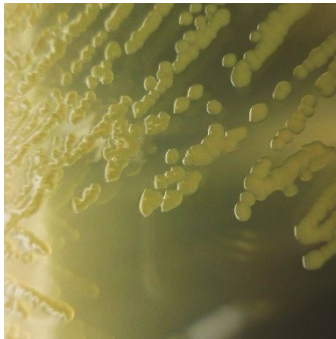
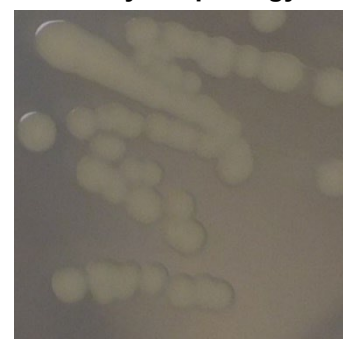


Figure 1b: NR-51883 Colony Morphology



Figure 1c: NR-51862 Colony Morphology



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31 MAY 2023

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