

Antimicrobial Resistance Panel 9: *Escherichia coli* Raetz Pathway Mutants

Catalog No. NR-55648

Product Description:

NR-55648 consists of an 8-member panel of *Escherichia coli* (*E. coli*) controlled expression mutant strains generated by disrupting the native copy of the indicated Lipid IVA biosynthesis pathway genes. The disrupted gene is expressed *in trans* in a plasmid under the control of *lac* promoter.

NR-51863 was produced by inoculation of deposited material into Tryptic Soy broth with 50 µg/mL kanamycin which was used to inoculate a Tryptic Soy agar with 50 µg/mL kanamycin plate, and both were grown at 37°C in an aerobic atmosphere for 1 day. After a hold at room temperature for 1 day, the material from the initial growth was passaged in Tryptic Soy broth with 50 µg/mL kanamycin for 1 day at 37°C in an aerobic atmosphere.

NR-51864, NR-51884, NR-51942 and NR-51944 were produced by inoculation of deposited material into Tryptic Soy broth with 50 µg/mL kanamycin and grown for 1 day at 37°C in an aerobic atmosphere. Broth inoculum was added to Tryptic Soy agar with 50 µg/mL kanamycin kolles, which were grown for 1 day at 37°C in an aerobic atmosphere.

NR-51865, NR-51941 and NR-51943 were produced by inoculation of 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.

Quality control testing was completed under propagation conditions unless otherwise noted.

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Table 1: Panel Components

COMPONENT NUMBER	STRAIN	MUTATION	LOT NUMBER	MANUFACTURING DATE
NR-51863	NB27082-CDU0019	<i>lpxD::Kan^R / pTU433 (pMMB, PtaclacUV5::Ec_lpxD)</i>	70047241	16SEP2021
NR-51864	NB27177-JRW0021	<i>ΔtolC::FRT, ΔlpxK::FRT, FL66-84MA (IPTG inducible lpxKPa, Kan^R)</i>	70047253	15SEP2021
NR-51865	NB27177-JWM0002	<i>ΔtolC lpxK::Kan^R / pMM14 (Plac::lpxK)</i>	70047255	16SEP2021
NR-51884	NB27082-TUP0006	<i>ΔlpxA::Kan^R / pTU406 (pMMB, CmR, PlacUV5::EclpxA)</i>	70047247	22SEP2021
NR-51941	NB27082-TUP0001	<i>ΔlpxD::frit / pTU433 (Plac::lpxD)</i>	70047243	16SEP2021
NR-51942	NB27082-TUP0005	<i>ΔlpxA::Kan^R / pTU406 (Plac::lpxA)</i>	70047245	22SEP2021
NR-51943	NB27354-TUT0035	<i>Δcdh::FRT ΔtolC::FRT</i>	70043420	23APR2021
NR-51944	NB27176-JWM0004	<i>ΔlpxK::Kan^R / pMM14 (Plac::lpxK)</i>	70047251	15SEP2021

Table 2: *Escherichia coli*, Strain NB27082-CDU0019 (NR-51863)

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, translucent and cream Motile <i>E. coli</i> (99.9%)

TEST	SPECIFICATIONS	RESULTS
Antibiotic Susceptibility Profile BD BBL™ Sensi-Disc™ susceptibility test disc 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Novobiocin Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Azithromycin Ciprofloxacin Fusidic Acid Polymixin B Rifampin	Report results Report results Report results Report results Report results	≥ 6 mm 3 µg/mL 0.032 µg/mL ≥ 256 µg/mL 0.125 µg/mL ≥ 32 µg/mL
Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ¹ Confirmation of deletion	≥ 70% for species identification Deletion confirmed	<i>E. coli</i> (74.1%) Pending
Purity 7 days on Tryptic Soy agar at 37°C in an aerobic atmosphere with and without 5% CO ₂	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Viability	Growth	Growth

¹Relatedness 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.

Table 3: *Escherichia coli*, Strain NB27177-JRW0021 (NR-51864)

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 Motile <i>E. coli</i> (99.9%)
Antibiotic Susceptibility Profile BD BBL™ Sensi-Disc™ susceptibility test disc 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Novobiocin Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Azithromycin Ciprofloxacin Fusidic Acid Polymixin B Rifampin	Report results Report results Report results Report results Report results	22 mm 1.5 µg/mL 0.004 µg/mL 4 µg/mL ≤ 0.064 µg/mL 6 µg/mL
Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ¹ Confirmation of deletions	≥ 70% for species identification Deletions confirmed	<i>E. coli</i> (75.1%) Pending
Purity 7 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	Growth consistent with expected colony morphology
Viability	Growth	Growth

¹Relatedness 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.

Table 4: *Escherichia coli*, Strain NB27177-JWM0002 (NR-51865)

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, translucent and cream Motile <i>E. coli</i> (99.9%)
Antibiotic Susceptibility Profile BD BBL™ Sensi-Disc™ susceptibility test disc 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Novobiocin Etest [®] antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Azithromycin Ciprofloxacin Fusidic Acid Polymixin B Rifampin	Report results Report results Report results Report results Report results	19 mm 1 µg/mL 0.004 µg/mL 96 µg/mL 0.125 µg/mL 12 µg/mL
Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ¹ Confirmation of deletions	≥ 70% for species identification Deletions confirmed	<i>E. coli</i> (75.1%) Pending
Purity 7 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	Growth consistent with expected colony morphology
Viability	Growth	Growth

¹Relatedness 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.

Table 5: *Escherichia coli*, Strain NB27082-TUP0006 (NR-51884)

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, translucent and cream Motile <i>E. coli</i> (99.9%)
Antibiotic Susceptibility Profile BD BBL™ Sensi-Disc™ susceptibility test disc 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Novobiocin Etest [®] antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Azithromycin Ciprofloxacin	Report results Report results Report results	≤ 6 mm 3 µg/mL 0.008 µg/mL

TEST	SPECIFICATIONS	RESULTS
Fusidic Acid Polymixin B Rifampin	Report results Report results Report results	≥ 256 µg/mL 0.125 µg/mL ≥ 32 µg/mL
Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ¹ Confirmation of deletion	≥ 70% for species identification Deletion confirmed	<i>E. coli</i> (75.1%) Pending
Purity 7 days on Tryptic Soy agar at 37°C in an aerobic atmosphere with and without 5% CO ₂	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Viability	Growth	Growth

¹Relatedness 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.

Table 6: *Escherichia coli*, Strain NB27082-TUP0001(NR-51941)

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, translucent and cream Motile <i>E. coli</i> (99.9%)
Antibiotic Susceptibility Profile BD BBL™ Sensi-Disc™ susceptibility test disc 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Novobiocin Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Azithromycin Ciprofloxacin Fusidic Acid Polymixin B Rifampin	Report results Report results Report results Report results Report results	≤ 6 mm 4 µg/mL 0.016 µg/mL ≥ 256 µg/mL 0.125 µg/mL ≥ 32 µg/mL
Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ¹ Confirmation of deletion	≥ 70% for species identification Deletion confirmed	<i>E. coli</i> (75.1%) Pending
Purity 7 days at 37°C in an aerobic atmosphere with 5% CO ₂ on Tryptic Soy agar with 5% defibrinated sheep blood	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Viability	Growth	Growth

¹Relatedness 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.

Table 7: *Escherichia coli*, Strain NB27082-TUP0005 (NR-51942)

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis Cellular morphology Colony morphology Motility (wet mount)	Gram-negative rods Report results Report results	Gram-negative rods Circular, convex, entire, smooth, and cream Motile

TEST	SPECIFICATIONS	RESULTS
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 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Novobiocin Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Azithromycin Ciprofloxacin Fusidic Acid Polymixin B Rifampin	Report results Report results Report results Report results Report results	≤ 6 mm 2 µg/mL 0.008 µg/mL ≥ 256 µg/mL ≤ 0.125 µg/mL ≥ 32 µg/mL
Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ¹ Confirmation of deletion	≥ 70% for species identification Deletion confirmed	<i>E. coli</i> (75.1%) Pending
Purity 7 days at 37°C in an aerobic atmosphere with 5% CO ₂ on Tryptic Soy agar with 5% defibrinated sheep blood	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Viability	Growth	Growth

¹Relatedness 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.

Table 8: *Escherichia coli*, NB27354-TUT0035 (NR-51943)

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 Motile <i>E. coli</i> (99.9%)
Antibiotic Susceptibility Profile BD BBL™ Sensi-Disc™ susceptibility test disc 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Gatifloxacin Novobiocin Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Clindamycin Erythromycin Linezolid Rifampin Tetracyclin Trimethoprim Sensititre™ Gram Negative GNX2F Colistin	Report results Report results Report results Report results Report results Report results Report results Report results Report results	34 to 35 mm 11 mm 1.5 µg/mL 4 µg/mL 6 to 8 µg/mL 16 µg/mL 1.5 µg/mL 0.64 µg/mL ≤ 0.25 µg/mL
Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ¹ Confirmation of deletions	≥ 70% for species identification Deletions confirmed	<i>E. coli</i> (75.4%) Pending

TEST	SPECIFICATIONS	RESULTS
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

¹Relatedness 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.

Table 9: *Escherichia coli*, Strain NB27176-JWM0004 (NR-51944)

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 Motile <i>E. coli</i> (99.9%)
Antibiotic Susceptibility Profile BD BBL™ Sensi-Disc™ susceptibility test disc 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Novobiocin Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar Azithromycin Ciprofloxacin Fusidic Acid Polymixin B Rifampin	Report results Report results Report results Report results Report results	≤ 6 mm 3 µg/mL 0.012 µg/mL ≥ 256 µg/mL ≤ 0.064 µg/mL 12 µg/mL
Genotypic Analysis Digital DNA-DNA hybridization (dDDH) ¹	≥ 70% for species identification	<i>E. coli</i> (75.1%)
Confirmation of deletion	Deletion confirmed	Pending
Purity 7 days on Tryptic Soy agar with 5% defibrinated sheep blood at 37°C in an aerobic atmosphere with and without 5% CO ₂	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Viability	Growth	Growth

¹Relatedness 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.

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