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

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

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis		
Cellular morphology	Gram-negative rods	Gram-negative rods
Colony morphology	Report results	Circular, low convex, entire, smooth, translucent and cream
Motility (wet mount)	Report results	Motile
VITEK® MS (MALDI-TOF)	*E. coli*	*E. coli* (99.9%)

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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	Report results	≥ 6 mm
Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar		
Azithromycin	Report results	3 µg/mL
Ciprofloxacin	Report results	0.032 µg/mL
Fusidic Acid	Report results	≥ 256 µg/mL
Polymixin B	Report results	0.125 µg/mL
Rifampin	Report results	≥ 32 µg/mL
Genotypic Analysis		
Digital DNA-DNA hybridization (dDDH)[1]	≥ 70% for species identification	*E. coli* (74.1%)
Confirmation of deletion	Deletion confirmed	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

[1]Relatedness 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	Gram-negative rods	Gram-negative rods
Colony morphology	Report results	Circular, convex, entire, smooth, and cream
Motility (wet mount)	Report results	Motile
VITEK® MS (MALDI-TOF)	*E. coli*	*E. coli* (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	Report results	22 mm
Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar		
Azithromycin	Report results	1.5 µg/mL
Ciprofloxacin	Report results	0.004 µg/mL
Fusidic Acid	Report results	4 µg/mL
Polymixin B	Report results	≤ 0.064 µg/mL
Rifampin	Report results	6 µg/mL
Genotypic Analysis		
Digital DNA-DNA hybridization (dDDH)[1]	≥ 70% for species identification	*E. coli* (75.1%)
Confirmation of deletions	Deletions confirmed	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

[1]Relatedness 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	Gram-negative rods	Gram-negative rods
Colony morphology	Report results	Circular, low convex, entire, smooth, translucent and cream
Motility (wet mount)	Report results	Motile
VITEK® MS (MALDI-TOF)	*E. coli*	*E. coli* (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	Report results	19 mm
Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar		
Azithromycin	Report results	1 µg/mL
Ciprofloxacin	Report results	0.004 µg/mL
Fusidic Acid	Report results	96 µg/mL
Polymixin B	Report results	0.125 µg/mL
Rifampin	Report results	12 µg/mL
Genotypic Analysis		
Digital DNA-DNA hybridization (dDDH)[1]	≥ 70% for species identification	*E. coli* (75.1%)
Confirmation of deletions	Deletions confirmed	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

[1]Relatedness 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	Gram-negative rods	Gram-negative rods
Colony morphology	Report results	Circular, low convex, entire, smooth, translucent and cream
Motility (wet mount)	Report results	Motile
VITEK® MS (MALDI-TOF)	*E. coli*	*E. coli* (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	Report results	≤ 6 mm
Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar		
Azithromycin	Report results	3 µg/mL
Ciprofloxacin	Report results	0.008 µg/mL

TEST	SPECIFICATIONS	RESULTS
Fusidic Acid	Report results	≥ 256 µg/mL
Polymixin B	Report results	0.125 µg/mL
Rifampin	Report results	≥ 32 µg/mL
Genotypic Analysis		
Digital DNA-DNA hybridization (dDDH)[1]	≥ 70% for species identification	*E. coli* (75.1%)
Confirmation of deletion	Deletion confirmed	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

[1]Relatedness 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	Gram-negative rods	Gram-negative rods
Colony morphology	Report results	Circular, low convex, entire, smooth, translucent and cream
Motility (wet mount)	Report results	Motile
VITEK® MS (MALDI-TOF)	*E. coli*	*E. coli* (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	Report results	≤ 6 mm
Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar		
Azithromycin	Report results	4 µg/mL
Ciprofloxacin	Report results	0.016 µg/mL
Fusidic Acid	Report results	≥ 256 µg/mL
Polymixin B	Report results	0.125 µg/mL
Rifampin	Report results	≥ 32 µg/mL
Genotypic Analysis		
Digital DNA-DNA hybridization (dDDH)[1]	≥ 70% for species identification	*E. coli* (75.1%)
Confirmation of deletion	Deletion confirmed	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

[1]Relatedness 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	Gram-negative rods	Gram-negative rods
Colony morphology	Report results	Circular, convex, entire, smooth, and cream
Motility (wet mount)	Report results	Motile

TEST	SPECIFICATIONS	RESULTS
VITEK® MS (MALDI-TOF)	*E. coli*	*E. coli* (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	Report results	≤ 6 mm
Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar		
Azithromycin	Report results	2 µg/mL
Ciprofloxacin	Report results	0.008 µg/mL
Fusidic Acid	Report results	≥ 256 µg/mL
Polymixin B	Report results	≤ 0.125 µg/mL
Rifampin	Report results	≥ 32 µg/mL
Genotypic Analysis		
Digital DNA-DNA hybridization (dDDH)[1]	≥ 70% for species identification	*E. coli* (75.1%)
Confirmation of deletion	Deletion confirmed	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

[1]Relatedness 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	Gram-negative rods	Gram-negative rods
Colony morphology	Report results	Circular, low convex, entire, smooth and cream
Motility (wet mount)	Report results	Motile
VITEK® MS (MALDI-TOF)	*E. coli*	*E. coli* (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	Report results	34 to 35 mm
Novobiocin	Report results	11 mm
Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar		
Clindamycin	Report results	1.5 µg/mL
Erythromycin	Report results	4 µg/mL
Linezolid	Report results	6 to 8 µg/mL
Rifampin	Report results	16 µg/mL
Tetracyclin	Report results	1.5 µg/mL
Trimethoprim	Report results	0.64 µg/mL
Sensititre™ Gram Negative GNX2F		
Colistin	Report results	≤ 0.25 µg/mL
Genotypic Analysis		
Digital DNA-DNA hybridization (dDDH)[1]	≥ 70% for species identification	*E. coli* (75.4%)
Confirmation of deletions	Deletions confirmed	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

[1]Relatedness 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	Gram-negative rods	Gram-negative rods
Colony morphology	Report results	Circular, convex, entire, smooth, and cream
Motility (wet mount)	Report results	Motile
VITEK® MS (MALDI-TOF)	*E. coli*	*E. coli* (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	Report results	≤ 6 mm
Etest® antibiotic test strips 1 day at 35°C in an aerobic atmosphere on Mueller Hinton agar		
Azithromycin	Report results	3 µg/mL
Ciprofloxacin	Report results	0.012 µg/mL
Fusidic Acid	Report results	≥ 256 µg/mL
Polymixin B	Report results	≤ 0.064 µg/mL
Rifampin	Report results	12 µg/mL
Genotypic Analysis		
Digital DNA-DNA hybridization (dDDH)[1]	≥ 70% for species identification	*E. coli* (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

[1]Relatedness 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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