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Streptococcus pneumoniae Gateway[®] Clone Set, Recombinant in *Escherichia coli*, Plate 7

Catalog No. NR-19574

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Contributor:

Pathogen Functional Genomics Resource Center at the J. Craig Venter Institute

Manufacturer:

BEI Resources

Product Description:

Clone plates are replicated using a BioMek[®] FX robot. Production in the 96-well format has increased risk of crosscontamination between adjacent wells. Individual clones should be purified (e.g. single colony isolation and purification using good microbiological practices) and sequence-verified prior to use. BEI Resources only confirms the clone plate orientation and viability of randomly picked clones. BEI Resources does not confirm or validate individual clone identities provided by the contributor.

The Streptococcus pneumoniae (S. pneumoniae) Gateway[®] clone set consists of approximately 2029 sequence validated clones from S. pneumonia, strain TIGR4 cloned in *Escherichia coli* (*E. coli*) DH10B-T1 cells.¹ Each open reading frame was constructed in vector <u>pDONRTM221</u> (InvitrogenTM) with a native start codon and no stop codon. The sequence was validated by full length sequencing of each clone with greater than 1X coverage and a mutation rate of less than 0.2%. Detailed information about each clone is shown in Table 1.

Information related to the use of Gateway[®] Clones can be obtained from InvitrogenTM. Recombination was facilitated through an *att*B substrate (*att*B-PCR product or a linearized *att*B expression clone) with an *att*P substrate (pDONRTM221) to create an *att*L-containing entry clone. The entry clone contains recombinational cloning sites, *att*L1 and *att*L2 to facilitate gene transfer into a destination vector, M13 forward and reverse priming sites for sequencing and a kanamycin resistance gene for selection. Please refer to the InvitrogenTM <u>Gateway[®] Technology Manual</u> for additional details.

Plate orientation and viability were confirmed for NR-19574.

Material Provided:

Each inoculated well of the 96-well plate contains approximately 60 μ L of *E. coli* culture (strain DH10B-T1) in Luria Bertani (LB) broth containing 50 μ g/mL kanamycin supplemented with 15% glycerol.

Packaging/Storage:

NR-19574 was packaged aseptically in a 96-well plate. The product is provided frozen and should be stored at -80°C or colder immediately upon arrival. For long-term storage, the vapor phase of a liquid nitrogen freezer is recommended. Freeze-thaw cycles should be avoided.

Growth Conditions:

Media:

LB broth containing 50 µg/mL kanamycin LB agar containing 50 µg/mL kanamycin Incubation: Temperature: 37°C. Atmosphere: Aerobic <u>Propagation</u>:

- 1. Scrape top of frozen well with a pipette tip and streak onto agar plate.
- 2. Incubate the plates at 37°C for 18 to 24 hours.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Streptococcus pneumoniae* Gateway[®] Clone Set, Recombinant in *Escherichia coli*, Plate 7, NR-19574."

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. <u>Biosafety in</u> <u>Microbiological and Biomedical Laboratories</u>. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

Disclaimers:

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References:

 Kwon, K., et al. "A Correlation Analysis of Protein Characteristics Associated with Genome-Wide High Throughput Expression and Solubility of Streptococcus pneumoniae Proteins." <u>Protein Expr. Purif.</u> 55 (2007): 368-378. PubMed: 17703947.

ATCC[®] is a trademark of the American Type Culture Collection.



Table 1: Streptococcus pneumoniae Gateway[®] Clone Set, Recombinant in Escherichia coli, Plate 7 (YSPCG)¹

	Well		Description	ORF	Accession	Average Depth of
Clone	Position	Locus ID	Description	Length	Number	Coverage
23031	A01	SP0319	hypothetical protein SP_0319	387	NP_344855.1	2.89405685
23034	A02	SP1987	ABC transporter ATP-binding protein	871	NP_346414.1	2
23035	A03	SP2060	pyrrolidone-carboxylate peptidase	1291	NP_346484.1	3.04570101
23036	A04	SP0860	pyrrolidone-carboxylate peptidase	831	NP_345348.1	8.89410349
23037	A05	SP1677	hypothetical protein SP_1677	835	NP_346116.1	5.4994012
23038	A06	SP1125	phosphoserine phosphatase	1113	-	10.4851752
23039	A07	SP0940	replication initiator protein	897	-	8.66778149
23040	A08	SP0600	ABC transporter ATP-binding protein	618	NP_345112.1	3.21359223
23041	A09	SP0707	ABC transporter ATP-binding protein	696	NP_345210.1	13.0833333
23042	A10	SP1466	hemolysin	151	NP_345920.1	2
23043	A11	SP1139	hypothetical protein SP_1139	1134	NP_345609.1	8.12962963
23046	A12	SP0954	competence protein CeIA	915	NP_345435.1	6.26120219
23047	B01	SP1638	iron-dependent transcriptional regulator	667	NP_346078.1	4.98650675
23048	B02	SP2088	phosphate transport system regulatory protein PhoU	1381	NP_346508.1	1.19840695
23049	B03	SP0215	30S ribosomal protein S3	306	NP_344755.1	5.02614379
23050	B04	SP1395	phosphate transport system regulatory protein PhoU	1749	NP_345853.1	12.4636935
23051	B05	SP2193	DNA-binding response regulator	2032	NP_346604.1	3.81643701
23053	B06	SP1983	ribulose-phosphate 3-epimerase	862	NP_346410.1	2
23055	B07	SP0754	acetoin utilization protein AcuB	738	NP_345253.1	2.47154472
23056	B08	SP0603	DNA-binding response regulator VncR	621	NP_345115.1	4.99355878
23057	B09	SP0607	amino acid ABC transporter permease	624	NP_345119.1	4.44551282
23058	B10	SP0891	type I restriction-modification system, S subunit	858	NP_345378.1	8.08041958
23059	B11	SP1827	hypothetical protein SP_1827	253	NP_346260.1	2
23060	B12	SP1808	type IV prepilin peptidase	202	NP_346241.1	1.91089109
23061	C01	SP2207	competence protein ComF	2119	NP_346618.1	3.18074563
23062	C02	SP0843	deoxyribose-phosphate aldolase	813	NP_345334.1	9.54120541
23063	C03	SP0079	Trk family potassium uptake protein	201	NP_344628.1	2.95522388
23065	C04	SP0144	hypothetical protein SP_0144	252	NP_344686.1	9.99206349
23067	C05	SP2035	3-keto-L-gulonate-6-phosphate decarboxylase	1117	NP_346460.1	1.65801253
23069	C06	SP0716	transcriptional regulator	699	NP_345217.1	13.0629471
23070	C07	SP0252	fructose-6-phosphate aldolase	336	NP_344791.1	2.69940476

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Clone	Well Position	Locus ID	Description	ORF Length	Accession Number	Average Depth of Coverage
23071	C08	SP1665	yImE protein	787	NP_346104.1	4.69377382
23073	C09	SP0106	L-serine dehydratase, iron-sulfur- dependent, beta subunit	231	NP_344653.1	14.8398268
23074	C10	SP1603	cytidylate kinase	439	NP_346047.1	4
23076	C11	SP0798	DNA-binding response regulator CiaR	774	NP_345295.1	7.60594315
23079	C12	SP1377	3-dehydroquinate dehydratase	1673	NP_345835.1	13.1978482
23080	D01	SP0710	amino acid ABC transporter permease	696	NP_345212.1	12.6508621
23084	D02	SP1610	hypothetical protein SP_1610	469	NP_346054.1	3
23085	D03	SP1575	hypothetical protein SP_1575	316	NP_346021.1	4
23086	D04	SP0181	hypothetical protein SP_0181	282	NP_344722.1	4.22695035
23088	D05	SP1088	DNA repair protein RadC	1056	NP_345560.1	10.0625
23089	D06	SP2117	hypothetical protein SP_2117	1516	NP_346536.1	3.02704485
23090	D07	SP0127	hypothetical protein SP_0127	240	NP_344673.1	14.9208333
23092	D08	SP0349	capsular polysaccharide biosynthesis protein Cps4D	423	NP_344882.1	2.42789598
23093	D09	SP1982	hypothetical protein SP_1982	862	NP_346409.1	2
23094	D10	SP2033	L-ribulose-5-phosphate 4-epimerase	1099	NP_346458.1	1.69608735
23095	D11	SP0760	hypothetical protein SP_0760	741	NP_345258.1	7.03238866
23097	D12	SP1089	glutamine amidotransferase, class I	1056	NP_345561.1	9.68465909
23099	E01	SP0376	DNA-binding response regulator	450	NP_344903.1	4.38
23100	E02	SP0631	50S ribosomal protein L1	636	NP_345142.1	5.39622642
23103	E03	SP0823	amino acid ABC transporter permease	795	NP_345315.1	9.12830189
23104	E04	SP1838	glycosyl transferase	283	NP_346271.1	2
23105	E05	SP0152	ABC transporter permease	258	NP_344694.1	13.9922481
23106	E06	SP1655	phosphoglyceromutase	739	NP_346094.1	4.52774019
23107	E07	SP0991	5'-methylthioadenosine/S- adenosylhomocysteine nucleosidase	948	NP_345470.1	8.88396624
23108	E08	SP0914	-	873	-	7.27147766
23109	E09	SP0722	transcriptional activator TenA	702	NP_345223.1	13.0669516
23110	E10	SP0240	phosphoglycerate mutase family protein	318	NP_344780.1	4.8427673
23111	E11	SP0348	capsular polysaccharide biosynthesis protein Cps4C	423	NP_344881.1	2.43026005
23113	E12	SP0288	hypothetical protein SP_0288	363	NP_344826.1	3.27823691
23114	F01	SP1616	allulose-6-phosphate 3-epimerase	478	NP_346057.1	4
23115	F02	SP0255	acetyltransferase	339	NP_344794.1	2.82300885
23116	F03	SP1685	N-acetylmannosamine-6-phosphate 2- epimerase	874	NP_346124.1	4.17162471
23117	F04	SP1248	ribonuclease III	1260	NP_345713.1	7.48333333
23118	F05	SP2097	2,3,4,5-tetrahydropyridine-2, 6-carboxylate N-succinyltransferase,	1402	NP_346516.1	2.99500713
23119	F06	SP0793	3-ketoacyl-(acyl-carrier-protein) reductase	771	NP_345290.1	9.30479896
23120	F07	SP0385	hypothetical protein SP_0385	456	NP_344912.1	5.82017544
23121	F08	SP0083	DNA-binding response regulator	204	NP_344631.1	-
23122	F09	SP1415	glucosamine-6-phosphate isomerase	1890	 NP_345873.1	13.5507937
23123	F10	SP0786	ABC transporter ATP-binding protein	765	 NP_345283.1	9.46797386
23124	F11	SP0701	orotidine 5'-phosphate decarboxylase	690	 NP_345204.1	13.0231884
23125	F12	SP1867	NAD-dependent epimerase/dehydratase family protein	385	NP_346299.1	2
23126	G01	SP1330	N-acetylmannosamine-6-phosphate 2- epimerase	1389	NP_345788.1	20.9366451

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Clone	Well Position	Locus ID	Description	ORF Length	Accession Number	Average Depth of Coverage
23127	G02	SP1653	ABC transporter ATP-binding protein	712	NP_346092.1	5.19382022
23129	G03	SP0184	hypothetical protein SP_0184	285	NP_344725.1	4.2245614
23130	G04	SP0075	phosphorylase Pnp/Udp family protein	198	NP_344624.1	-
23132	G05	SP1227	DNA-binding response regulator	1239	NP_345693.1	7.16545601
23134	G06	SP2184	glycerol uptake facilitator protein	2011	NP_346595.1	2.28791646
23135	G07	SP2171	zinc ABC transporter ATP-binding protein	1918	NP_346585.1	3.46089677
23136	G08	SP1668	hypothetical protein SP_1668	790	NP_346107.1	5.41012658
23137	G09	SP1271	2-C-methyl-D-erythritol 4-phosphate cytidylyltransferase	1302	NP_345735.1	11.9093702
23138	G10	SP1972	hypothetical protein SP_1972	835	NP_346399.1	2
23139	G11	SP1446	GntR family transcriptional regulator	133	NP_345900.1	4
23141	G12	SP0044	phosphoribosylaminoimidazole- succinocarboxamide synthase	114	NP_344593.1	2
23143	H01	SP0658	cytochrome c-type biogenesis protein CcdA	654	NP_345163.1	5.09633028
23144	H02	SP1823	MgtC/SapB family protein	244	NP_346256.1	2
23145	H03	SP1885	trehalose operon transcriptional repressor	472	NP_346317.1	2
23147	H04	SP0835	purine nucleoside phosphorylase	804	NP_345326.1	8.19776119
23148	H05	SP1285	16S rRNA methyltransferase GidB	1329	NP_345749.1	12.4680211
23150	H06	SP0698	hypothetical protein SP_0698	690	NP_345202.1	11.2333333
23151	H07	SP1710	nitroreductase family protein	1030	NP_346148.1	4.26116505
23153	H08	SP0058	GntR family transcriptional regulator	180	NP_344607.1	-
23155	H09	SP0629	hypothetical protein SP_0629	636	NP_345140.1	4.17767296
23158	H10	SP0779	tRNA (guanine-N(1)-)- methyltransferase	762	NP_345277.1	8.35301837
23160	H11	SP1874	ribosomal large subunit pseudouridine synthase B	424	NP_346306.1	1.48584906
23161	H12	SP1648	manganese ABC transporter ATP- binding protein	700	NP_346088.1	5.93142857

¹All information in this table was provided by J. Craig Venter Institute at the time of deposition.