Mycobacterium tuberculosis Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 5

Catalog No. NR-19641

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Contributor:

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

Manufacturer:

BEI Resources

Product Description:

Production in the 96-well format has increased risk of cross-contamination 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 does not confirm or validate individual mutants provided by the contributor.

The *Mycobacterium tuberculosis (M. tuberculosis)* Gateway® clone set consists of 42 plates which contain 3724 sequence validated clones (3294 *M. tuberculosis,* strain H37Rv clones supplemented with 430 unique open reading frames (ORF) from *M. tuberculosis,* strain CDC1551) cloned in *Escherichia coli* (*E. coli*) DH10B-T1 cells. Each ORF was recombined in vector pDONR™221 with an ATG start codon and no stop codon. The sequence was validated by full length sequencing of each entry 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 Invitrogen™. Recombination was facilitated through an *att*B substrate (*att*B-PCR product or a linearized *att*B expression clone) with an *att*P substrate (pDONR™221) 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 Invitrogen™ Gateway® Technology Manual for additional details.

Plate orientation and viability were confirmed for NR-19641.

Material Provided:

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

Packaging/Storage:

NR-19641 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 or agar containing 50 µg/mL kanamycin
Incubation:
Temperature: *E. coli*, strain DH10B-T1 clones should be grown at 37°C.
Atmosphere: Aerobic
Propagation:
1. Scrape top of frozen well with a pipette tip and streak onto agar plate.
2. Incubate the plates at 37°C for 1 day.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Mycobacterium tuberculosis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 5, NR-19641."

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. Biosafety in Microbiological and Biomedical Laboratories. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

Disclaimers:

You are authorized to use this product for research use only. It is not intended for human use.

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References:

1. Cole, S. T., et al. "Deciphering the Biology of *Mycobacterium tuberculosis* from the Complete Genome Sequence." Nature 393 (1998): 537-544. PubMed: 9634230.
2. Camus, J. C., et al. "Re-Annotation of the Genome Sequence of *Mycobacterium tuberculosis* H37Rv." Microbiology 148 (2002): 2967-2973. PubMed 12368430.

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Support Provided by NIAID

Table 1: *Mycobacterium tuberculosis*, Gateway® Clones, Plate 5 (ZMTDE)[1]

Clone	Well Position	ORF Length	Locus ID	Description	Accession Number	Average Depth of Coverage
73387	A01	1030	Rv1681	molybdopterin biosynthesis protein MoeX	NP_216197.1	2.621359223
73419	A02	1030	Rv3694c	transmembrane protein	NP_218211.1	3.186407767
73351	A03	1033	Rv3261	LPPG:FO 2-phospho-L-lactate transferase	NP_217778.1	1.181994192
73165	A04	1033	Rv3464	dTDP-glucose 4,6-dehydratase RMLB	NP_217981.1	1.460793804
73251	A05	1036	Rv1285	sulfate adenylyltransferase subunit 2	NP_215801.1	3.283783784
73790	A06	1039	Rv2424c	transposase	NP_216940.1	3.204042348
73767	A07	1039	Rv3001c	ketol-acid reductoisomerase	NP_217517.1	3.769008662
73743	A08	1042	Rv1912c	oxidoreductase FADB5	NP_216428.1	1.657389635
73802	A09	1060	Rv0129c	secreted antigen 85-C FBPC (85C) (antigen 85 complex C) (AG58C) (Mycolyl transferase 85C) (fibronectin-binding protein C)	YP_177694.1	2.737735849
73748	A10	1060	Rv0885	hypothetical protein	NP_215400.1	1.63490566
73834	A11	1060	Rv3194c	hypothetical protein	NP_217710.1	3.544339623
73898	A12	1062	Rv2568c	hypothetical protein	NP_217084.1	2.425612053
73787	B01	1063	Rv1043c	hypothetical protein	NP_215559.1	3.645343368
73705	B02	1072	Rv2592c	Holliday junction DNA helicase RuvB	NP_217108.1	2.014925373
73711	B03	1078	Rv0536	UDP-glucose 4-epimerase	YP_177737.1	3.130797774
73878	B04	1078	Rv1168c	PPE family protein	YP_177791.1	1.386827458
73761	B05	1078	Rv1520	sugar transferase	NP_216036.1	2.045454545
73679	B06	1078	Rv3056	DNA polymerase IV	NP_217572.1	3.046382189
73633	B07	1081	Rv0791c	hypothetical protein	NP_215306.1	2.061054579
73736	B08	1081	Rv1523	methyltransferase	NP_216039.1	1.55226642
73755	B09	1081	Rv2167c	transposase	NP_216683.1	3.183163737
73830	B10	1087	Rv0072	glutamine-transport transmembrane protein ABC transporter	NP_214586.1	3.530818767
73567	B11	1096	Rv3659c	hypothetical protein	YP_178003.1	2.832116788
73883	B12	1099	Rv1940	riboflavin biosynthesis protein ribA1 (GTP cyclohydrolase II)	YP_177851.1	3.295723385
73541	C01	1105	Rv0125	serine protease PepA	NP_214639.1	1.31040724
73674	C02	1105	Rv2571c	transmembrane alanine and valine and leucine rich protein	NP_217087.1	2.857918552
73694	C03	1108	Rv2777c	hypothetical protein		1.324909747
73557	C04	1111	Rv2678c	uroporphyrinogen decarboxylase	NP_217194.1	1.96039604
73682	C05	1114	Rv3586	DNA integrity scanning protein DisA	NP_218103.1	3.527827648
73686	C06	1117	Rv1184c	hypothetical protein	NP_215700.1	1.368845121
73779	C07	1117	Rv2156c	phospho-N-acetylmuramoyl-pentapeptide-transferase	NP_216672.1	3.290062668
73915	C08	1117	Rv3044	FEIII-dicitrate-binding periplasmic lipoprotein	NP_217560.1	3.347358997
73855	C09	1120	Rv3035	hypothetical protein	NP_217551.1	2.814285714
73895	C10	1126	Rv0542c	O-succinylbenzoic acid--CoA ligase	NP_215056.1	2.376554174

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Clone	Well Position	ORF Length	Locus ID	Description	Accession Number	Average Depth of Coverage
73798	C11	1132	Rv2231c	hypothetical protein	NP_216747.1	1.348939929
73630	C12	1132	Rv3468c	dTDP-glucose 4,6-dehydratase	YP_177974.1	3.617491166
73647	D01	1135	Rv1787	PPE family protein	YP_177833.1	2.903964758
73922	D02	1138	Rv3921c	putative inner membrane protein translocase component YidC	NP_218438.1	2.775922671
73614	D03	1141	Rv0205	transmembrane protein	NP_214719.1	2.862401402
73824	D04	1147	Rv1936	monooxygenase	NP_216452.1	1.401918047
73723	D05	1156	Rv2535c	cytoplasmic peptidase PepQ	NP_217051.1	3.553633218
73619	D06	1156	Rv2856	nickel-transport integral membrane protein	NP_217372.1	3.128027682
73639	D07	1159	Rv1516c	sugar transferase	NP_216032.2	3.391716997
73874	D08	1165	Rv2089c	dipeptidase PepE	NP_216605.1	1.32360515
73643	D09	1168	Rv0433	carboxylate-amine ligase	NP_214947.1	2.906678082
73597	D10	1168	Rv2439c	gamma-glutamyl kinase	NP_216955.1	1.927226027
73657	D11	1171	Rv1479	transcriptional regulatory protein MOXR1	YP_177816.1	1.935952178
73815	D12	1171	Rv3562	acyl-CoA dehydrogenase FADE31	NP_218079.1	3.108454313
73826	E01	1174	Rv0557	mannosyltransferase PIMB	NP_215071.1	3.27427598
73739	E02	1177	Rv2150c	cell division protein FtsZ	NP_216666.1	3.204757859
73794	E03	1177	Rv2184c	hypothetical protein	NP_216700.1	1.23364486
73689	E04	1180	Rv0062	endo-1,4-beta-glucanase	YP_177689.1	1.902542373
73918	E05	1183	Rv2951c	oxidoreductase	NP_217467.1	1.284023669
73850	E06	1186	Rv3161c	dioxygenase	NP_217677.1	1.336424958
73752	E07	1189	Rv0628c	enoyl-CoA hydratase	NP_215146.1	1.347350715
73671	E08	1189	Rv3332	N-acetylglucosamine-6-phosphate deacetylase	NP_217849.1	3.110176619
73583	E09	1192	Rv2164c	hypothetical protein	NP_216680.1	3.367449664
73627	E10	1192	Rv3495c	MCE-family lipoprotein LprN	NP_218012.1	2.944630872
73758	E11	1195	Rv2190c	hypothetical protein Rv2190c	NP_216706.1	2.389958159
73809	E12	1198	Rv0115	D-alpha-D-heptose-7-phosphate kinase	NP_214629.1	1.446577629
73902	F01	1198	Rv1569	8-amino-7-oxononanoate synthase	YP_177822.1	3.136894825
73807	F02	1198	Rv3540c	lipid-transfer protein	NP_218057.1	2.939899833
73714	F03	1198	Rv3556c	acetyl-CoA acetyltransferase	NP_218073.1	3.373956594
73866	F04	1201	Rv2868c	4-hydroxy-3-methylbut-2-en-1-yl diphosphate synthase	NP_217384.1	3.331390508
73546	F05	1207	Rv0823c	transcriptional regulatory protein	NP_215338.1	2.788732394
73729	F06	1207	Rv3305c	N-acyl-L-amino acid amidohydrolase	YP_177955.1	1.694283347
74077	F07	1210	Rv2072c	precorrin-6y methyltransferase CobL	NP_216588.1	2.924793388
73980	F08	1210	Rv3860	hypothetical protein	NP_218377.1	1.442975207
73989	F09	1213	Rv1196	PPE family protein	YP_177795.1	1.981038747
74152	F10	1213	Rv3546	acetyl-CoA acetyltransferase	NP_218063.1	1.480626546
74101	F11	1219	Rv1131	citrate synthase	NP_215647.1	2.671862182
74125	F12	1219	Rv1372	hypothetical protein	YP_177803.1	2.40607055
74083	G01	1219	Rv1785c	cytochrome P450 143	NP_216301.1	3.007383101
74173	G02	1219	Rv2495c	branched-chain alpha-keto acid dehydrogenase subunit E2	NP_217011.1	2.673502871
74203	G03	1222	Rv2500c	acyl-CoA dehydrogenase	NP_217016.1	3.103109656
73985	G04	1225	Rv0400c	acyl-CoA dehydrogenase FADE7	NP_214914.1	1.192653061
74250	G05	1225	Rv3618	monooxygenase	NP_218135.1	2.10122449
74085	G06	1228	Rv0694	L-lactate dehydrogenase (cytochrome) LldD1	NP_215208.1	1.199511401
73932	G07	1228	Rv2276	cytochrome P450 121 CYP121	NP_216792.1	1.457654723
74217	G08	1231	Rv2723	integral membrane protein	NP_217239.1	1.086108855
74137	G09	1234	Rv3778c	aminotransferase	NP_218295.1	1.06726094
74145	G10	1237	Rv2188c	hypothetical protein	NP_216704.2	2.73807599
73997	G11	1240	Rv1812c	dehydrogenase	NP_216328.1	3.044354839

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Clone	Well Position	ORF Length	Locus ID	Description	Accession Number	Average Depth of Coverage
74047	G12	1243	Rv3644c	DNA polymerase III subunit delta'	NP_218161.1	3.585679807
73946	H01	1246	Rv0593	MCE-family lipoprotein LprL	NP_215107.1	1.33788122
74302	H02	1246	Rv1627c	lipid-transfer protein	NP_216143.1	1.337078652
74117	H03	1249	Rv2542	hypothetical protein	NP_217058.1	2.15692554
74299	H04	1252	Rv1302	undecapaprenyl-phosphate alpha-N-acetylglucosaminyltransferase rfe (UDP-GlcNAc transferase)	NP_215818.1	3.090255591
73949	H05	1252	Rv1653	bifunctional ornithine acetyltransferase/N-acetylglutamate synthase protein	NP_216169.1	2.748402556
74035	H06	1255	Rv1074c	acetyl-CoA acetyltransferase	NP_215590.1	3.000796813
74281	H07	1255	Rv1256c	cytochrome P450 130 CYP130	NP_215772.1	1.152191235
74003	H08	1255	Rv3330	penicillin-binding protein DacB1	NP_217847.1	3.419123506
74258	H09	1261	Rv2067c	hypothetical protein	NP_216583.1	2.087232355
74210	H10	1261	Rv3786c	hypothetical protein	NP_218303.1	2.069785884
74058	H11	1264	Rv2518c	lipoprotein LppS	NP_217034.1	2.053797468
74141	H12	1267	Rv3640c	transposase	NP_218157.1	1.055248619

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

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