

***Mycobacterium tuberculosis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 8**

**Catalog No. NR-19644**

This reagent is the tangible property of the U.S. Government.

**For research use only. Not for human use.**

**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 *attB* substrate (*attB*-PCR product or a linearized *attB* expression clone) with an *attP* substrate (pDONR™221) to create an *attL*-containing entry clone. The entry clone contains recombinational cloning sites, *attL1* and *attL2* 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-19644.

**Material Provided:**

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

**Packaging/Storage:**

NR-19644 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: 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 8, NR-19644.”

**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/bmb15/index.htm](http://www.cdc.gov/biosafety/publications/bmb15/index.htm).

**Disclaimers:**

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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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**Table 1: *Mycobacterium tuberculosis*, Gateway® Clones, Plate 8 (ZMTDH)<sup>1</sup>**

Clone	Well Position	ORF Length	Locus ID	Description (Gene name)	Accession Number	Average Depth of Coverage
75546	A01	2401	Rv0711	arylsulfatase AtsA (atsA)	NP_215225.1	4.06247397
75658	A02	2410	Rv2583c	GTP pyrophosphokinase (relA)	NP_217099.1	3.27344398
75758	A03	2425	Rv0585c	integral membrane protein	NP_215099.1	3.25113402
75734	A04	2431	Rv0908	metal cation transporter ATPase P-type CtpE (ctpE)	NP_215423.1	3.65734266
75822	A05	2464	Rv1821	preprotein translocase subunit SecA (secA2)	NP_216337.1	3.82102273
75669	A06	2602	Rv2047c	hypothetical protein Rv2047c	NP_216563.1	7.42928517
75835	A07	2605	Rv0987	adhesion component transport transmembrane protein ABC transporter	NP_215502.1	3.72360845
76013	A08	2914	Rv0402c	transmembrane transport protein MmpL1 (mmpL1)	NP_214916.1	3.00789293
75997	A09	2926	Rv2339	transmembrane transport protein MmpL9 (mmpL9)	NP_216855.1	3.27785373
75985	A10	2941	Rv0450c	transmembrane transport protein MmpL4 (mmpL4)	NP_214964.1	3.36688201
75913	A11	2944	Rv0507	transmembrane transport protein MmpL2 (mmpL2)	NP_215021.1	3.43546196
75977	A12	3265	Rv3479	hypothetical protein Rv3479	NP_217996.2	3.27105666
76062	B01	3307	Rv3823c	integral membrane transport protein (mmpL8)	NP_218340.1	3.60931358
76501	B02	130	MT1978	hypothetical protein MT1978	NP_336436.1	2
76345	B03	130	MT3102	hypothetical protein MT3102	NP_337617.1	2
76337	B04	133	MT1330	hypothetical protein MT1330	NP_335777.1	2
76509	B05	133	MT1790	hypothetical protein MT1790	NP_336249.1	2
76552	B06	133	MT2988.1	hypothetical protein MT2988.1	NP_337501.1	2
76445	B07	133	MT3449.2	hypothetical protein MT3449.2	NP_337979.1	2
76465	B08	133	MT3510.1	hypothetical protein MT3510.1	NP_338034.1	2
76513	B09	136	MT0325	hypothetical protein MT0325	NP_334733.1	2
76485	B10	136	MT0725	hypothetical protein MT0725	NP_335141.1	2
76561	B11	136	MT0835	hypothetical protein MT0835	NP_335263.1	2
76497	B12	136	MT1057.1	hypothetical protein MT1057.1	YP_061208.1	2
76184	C01	136	MT1342	hypothetical protein MT1342	NP_335790.1	2
76233	C02	136	MT2165.1	hypothetical protein MT2165.1	NP_336634.1	2
76297	C03	136	MT2803.1	tRNA delta(2)-isopentenylpyrophosphate transferase	NP_337302.1	2
76197	C04	139	MT1627	hypothetical protein MT1627	NP_336081.1	2
76353	C05	139	MT1717	hypothetical protein MT1717	NP_336171.1	2
76325	C06	139	MT2460	hypothetical protein MT2460	NP_336941.1	2
76457	C07	139	MT3580.1	hypothetical protein MT3580.1	NP_338124.1	2
76449	C08	142	MT3653.1	hypothetical protein MT3653.1	NP_338199.1	2
76533	C09	142	MT3952	hypothetical protein MT3952	NP_338505.1	2
76241	C10	142	MT3962	hypothetical protein MT3962	NP_338515.1	-
76213	C11	145	MT0827	hypothetical protein MT0827	NP_335255.1	2
76189	C12	145	MT1759	hypothetical protein MT1759	NP_336217.1	2
76489	D01	145	MT2011	hypothetical protein MT2011	NP_336469.1	2

Clone	Well Position	ORF Length	Locus ID	Description (Gene name)	Accession Number	Average Depth of Coverage
76521	D02	148	MT0450	hypothetical protein MT0450	NP_334859.1	2
76357	D03	148	MT1107	hypothetical protein MT1107	NP_335549.1	2
76280	D04	148	MT2364.1	hypothetical protein MT2364.1	NP_336837.1	2
76421	D05	148	MT3520	hypothetical protein MT3520	NP_338044.1	2
76477	D06	151	MT2438	hypothetical protein MT2438	NP_336918.1	2
76229	D07	154	MT1367.1	hypothetical protein MT1367.1	NP_335817.1	2
76462	D08	154	MT1909	hypothetical protein MT1909	NP_336366.1	2
76365	D09	154	MT3427.1	hypothetical protein MT3427.1	NP_337956.1	2
76246	D10	157	MT1116	hypothetical protein MT1116	NP_335558.1	2
76270	D11	157	MT1166	hypothetical protein MT1166	NP_335609.1	2
76437	D12	157	MT1285	hypothetical protein MT1285	NP_335729.1	2
76201	E01	157	MT1488	hypothetical protein MT1488	NP_335937.1	2
76261	E02	157	MT3449.1	hypothetical protein MT3449.1	NP_337978.1	2
76520	E03	160	MT0598	hypothetical protein MT0598	NP_335007.1	1.98125
76193	E04	160	MT1096.2	hypothetical protein MT1096.2	NP_335538.1	-
76369	E05	160	MT1760	hypothetical protein MT1760	NP_336218.1	2
76429	E06	160	MT2027	hypothetical protein MT2027	NP_336488.1	2
76537	E07	163	MT0159	hypothetical protein MT0159	NP_334569.1	2
76331	E08	166	MT2514	hypothetical protein MT2514	NP_336997.1	2
76341	E09	166	MT3145.1	hypothetical protein MT3145.1	NP_337664.1	2
76209	E10	169	MT0291.2	hypothetical protein MT0291.2	NP_334699.1	2
76362	E11	169	MT2722	hypothetical protein MT2722	NP_337222.1	2
76333	E12	169	MT3762	hypothetical protein MT3762	NP_338312.1	2.83431953
76413	F01	172	MT3273	hypothetical protein MT3273	NP_337799.1	2
76321	F02	172	MT3378	hypothetical protein MT3378	NP_337906.1	-
76405	F03	172	MT3744	hypothetical protein MT3744	NP_338290.1	2
76301	F04	175	MT0009	hypothetical protein MT0009	NP_334417.1	2
76305	F05	175	MT0521.1	hypothetical protein MT0521.1		1.92
76397	F06	175	MT1822	hypothetical protein MT1822	NP_336278.1	-
76556	F07	175	MT2015	hypothetical protein MT2015	NP_336473.1	2
76317	F08	178	MT0768.1	hypothetical protein MT0768.1	NP_335190.1	2
76417	F09	178	MT1025.1	hypothetical protein MT1025.1	NP_335459.1	-
76442	F10	178	MT1401	hypothetical protein MT1401	NP_335850.1	2
76544	F11	178	MT3032	hypothetical protein MT3032	NP_337544.1	2
76377	F12	178	MT3135	hypothetical protein MT3135	NP_337652.1	2
76381	G01	178	MT3207	hypothetical protein MT3207	NP_337733.1	2
76274	G02	184	MT0771	hypothetical protein MT0771	NP_335193.1	2
76259	G03	184	MT1192	hypothetical protein MT1192	NP_335635.1	2
76285	G04	184	MT2653	hypothetical protein MT2653	NP_337152.1	2
76253	G05	184	MT2993	hypothetical protein MT2993	NP_337506.1	1.60869565
76266	G06	187	MT0204.1	hypothetical protein MT0204.1	NP_334611.1	2
76185	G07	187	MT1798	hypothetical protein MT1798	NP_336257.1	2
76393	G08	187	MT2547.2	hypothetical protein MT2547.2	NP_337035.1	2
76228	G09	190	MT0012	hypothetical protein MT0012	NP_334420.1	2
76402	G10	190	MT0494	hypothetical protein MT0494	NP_334903.1	-
76281	G11	190	MT2007	hypothetical protein MT2007	NP_336465.1	1.91052632
76505	G12	190	MT2371	hypothetical protein MT2371	NP_336850.1	2
76545	H01	190	MT3921	hypothetical protein MT3921	NP_338473.1	2
76453	H02	193	MT0291.1	hypothetical protein MT0291.1	NP_334698.1	2
76409	H03	193	MT1650	hypothetical protein MT1650	NP_336105.1	2
76482	H04	193	MT2370.1	hypothetical protein MT2370.1	NP_336847.1	-
76385	H05	193	MT3535	hypothetical protein MT3535	NP_338061.1	2
76557	H06	196	MT2736.1	hypothetical protein MT2736.1	NP_337238.1	2

Clone	Well Position	ORF Length	Locus ID	Description (Gene name)	Accession Number	Average Depth of Coverage
76530	H07	196	MT3131.1	hypothetical protein MT3131.1	NP_337648.1	2
76221	H08	199	MT0932	hypothetical protein MT0932	NP_335365.1	2
76351	H09	199	MT3718.2	hypothetical protein MT3718.2	NP_338265.1	2
76250	H10	202	MT3210	hypothetical protein MT3210	NP_337736.1	2
76433	H11	205	MT1821.1	hypothetical protein MT1821.1	NP_336277.1	2
76218	H12	205	MT1839.1	hypothetical protein MT1839.1	NP_336297.1	2

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