

***Mycobacterium tuberculosis* Gateway®
Clone Set, Recombinant in *Escherichia coli*, Plate 18**

Catalog No. NR-19654

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

For research use only. Not for use in humans.

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-19654.

Material Provided:

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

Packaging/Storage:

NR-19654 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 micrograms per milliliter 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 18, NR-19654.”

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](#). 6th ed. Washington, DC: U.S. Government Printing Office, 2020; see 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 18 (ZMTLH)¹

Clone	Well Position	ORF Length	Locus ID	Description	Accession Number	Average Depth of Coverage
41177	A01	553	Rv1939	oxidoreductase	NP_216455.1	3.508137432
41172	A02	553	Rv1306	F0F1 ATP synthase subunit B	NP_215822.1	2
41175	A03	553	Rv1585c	phiRv1 phage protein	NP_216101.1	3.128390597
41167	A04	553	Rv0335c	PE family protein	YP_177717.1	2
41178	A05	553	Rv2290	lipoprotein lppO	NP_216806.1	2
41171	A06	553	Rv0556	transmembrane protein	NP_215070.1	2
41170	A07	553	Rv0443	hypothetical protein	NP_214957.1	3.927667269
41185	A08	556	Rv3278c	transmembrane protein	NP_217795.1	3.877697842
41187	A09	556	Rv3669	transmembrane protein	NP_218186.1	3.888489209
41188	A10	559	Rv0057	hypothetical protein	NP_214571.1	2
41191	A11	559	Rv2668	hypothetical protein	NP_217184.1	2
41190	A12	559	Rv1954c	hypothetical protein	NP_216470.1	3.799642218
41192	B01	559	Rv2878c	soluble secreted antigen MPT53 precursor	NP_217394.1	2
41204	B02	562	Rv3072c	hypothetical protein	NP_217588.1	2
41206	B03	562	Rv3275c	phosphoribosylaminoimidazole carboxylase catalytic subunit	NP_217792.1	4.268683274
41201	B04	562	Rv2311	hypothetical protein	NP_216827.1	4.259786477
41200	B05	562	Rv1930c	hypothetical protein	NP_216446.1	3.003558719
41195	B06	562	Rv0254c	bifunctional cobinamide kinase/cobinamide phosphate guanylyltransferase	NP_214768.1	2
41198	B07	562	Rv0481c	hypothetical protein	NP_214995.1	2
41199	B08	562	Rv1758	cutinase Cut1	NP_216274.1	2
41203	B09	562	Rv2747	N-acetylglutamate synthase	NP_217263.1	3.886120996
41207	B10	562	Rv3525c	siderophore-binding protein	NP_218042.1	2
41196	B11	562	Rv0461	hypothetical protein	NP_214975.1	4.544483986
41214	B12	565	Rv0901	hypothetical protein	NP_215416.1	3.49380531
41212	C01	565	Rv0745	hypothetical protein	NP_215259.1	2
41215	C02	565	Rv1234	hypothetical protein	NP_215750.1	2
41220	C03	565	Rv2270	lipoprotein lppN	NP_216786.1	2
41209	C04	565	Rv0740	hypothetical protein	NP_215254.1	4.325663717
41221	C05	565	Rv2330c	lipoprotein LppP	NP_216846.1	2
41232	C06	568	Rv3902c	hypothetical protein	NP_218419.1	3.913732394
41225	C07	568	Rv2539c	shikimate kinase	NP_217055.1	2
41227	C08	568	Rv3114	hypothetical protein	NP_217630.1	2
41226	C09	568	Rv2907c	16S rRNA-processing protein RimM	NP_217423.1	2
41230	C10	568	Rv3572	hypothetical protein	NP_218089.1	2
41223	C11	568	Rv1884c	resuscitation-promoting factor RpfC	NP_216400.1	2
41224	C12	568	Rv2140c	hypothetical protein	NP_216656.1	2
9986	D02	568	Rv2943A	transposase	YP_177680.1	4.146126761
41239	D03	571	Rv3281	hypothetical protein	NP_217798.1	3.098073555
41233	D04	571	Rv0735	RNA polymerase sigma factor SigL	NP_215249.1	2
41235	D05	571	Rv2256c	hypothetical protein	NP_216772.1	2

Clone	Well Position	ORF Length	Locus ID	Description	Accession Number	Average Depth of Coverage
41238	D06	571	Rv2651c	phiRv2 prophage protease	NP_217167.1	2
41241	D07	571	Rv3471c	hypothetical protein	NP_217988.1	2
41242	D08	571	Rv3847	hypothetical protein	NP_218364.1	2
41240	D09	571	Rv3324c	molybdenum cofactor biosynthesis protein C	NP_217841.2	2
41234	D10	571	Rv1227c	hypothetical protein	NP_215743.1	2
41247	D11	574	Rv3429	PPE family protein	YP_177973.1	2
41248	D12	574	Rv3780	hypothetical protein	NP_218297.1	2
41246	E01	574	Rv0651	50S ribosomal protein L10	NP_215165.1	3.893728223
41250	E02	577	Rv0382c	orotate phosphoribosyltransferase	YP_177723.1	4.372616984
41253	E03	577	Rv1988	methyltransferase		3.760831889
41255	E04	577	Rv2630	hypothetical protein	NP_217146.1	4.285961872
41252	E05	577	Rv0719	50S ribosomal protein L6	NP_215233.1	2
41249	E06	577	Rv0340	hypothetical protein	NP_214854.1	2
41261	E07	580	Rv1231c	hypothetical protein	NP_215747.1	2
41256	E09	580	Rv0047c	hypothetical protein	NP_214561.1	2
41263	E11	580	Rv1275	lipoprotein LprC	NP_215791.1	2.644827586
41273	E12	583	Rv2321c	ornithine aminotransferase	NP_216837.1	2
41271	F01	583	Rv0984	pterin-4-alpha-carbinolamine dehydratase MoaB2	NP_215499.1	2
41277	F03	583	Rv2843	hypothetical protein	NP_217359.1	3.159519726
41270	F04	583	Rv0762c	hypothetical protein	NP_215276.1	2
41280	F05	583	Rv3841	bacterioferritin BfrB	NP_218358.1	2
41265	F06	583	Rv0262c	aminoglycoside 2'-N-acetyltransferase AAC (AAC(2')-IC)	NP_214776.1	2
41272	F07	583	Rv1957	hypothetical protein	NP_216473.1	2
41290	F08	586	Rv1677	lipoprotein DsbF	NP_216193.1	4.327645051
41292	F09	586	Rv2367c	putative metalloprotease	NP_216883.1	2
41286	F10	586	Rv0738	hypothetical protein	NP_215252.1	1.796928328
41294	F11	586	Rv3584	lipoprotein LpqE	NP_218101.1	2
41293	F12	586	Rv3033	hypothetical protein	NP_217549.1	2
41291	G01	586	Rv1732c	hypothetical protein	NP_216248.1	2
41283	G02	586	Rv0137c	methionine sulfoxide reductase A	NP_214651.1	2
41282	G03	586	Rv0009	iron-regulated peptidyl-prolyl cis-trans isomerase A	NP_214523.1	3.109215017
41284	G04	586	Rv0219	transmembrane protein	NP_214733.1	2
41285	G06	586	Rv0513	transmembrane protein	NP_215027.1	2.537542662
41305	G07	589	Rv3222c	hypothetical protein	NP_217738.1	2
41307	G08	589	Rv3867	hypothetical protein	NP_218384.1	2
41306	G09	589	Rv3361c	hypothetical protein	NP_217878.1	2
41303	G10	589	Rv2838c	ribosome-binding factor A	NP_217354.1	2
41304	G11	589	Rv2842c	hypothetical protein	NP_217358.1	2
41296	G12	589	Rv0098	hypothetical protein	NP_214612.1	2.36672326
41311	H01	592	Rv3614c	hypothetical protein	NP_218131.1	2
41308	H02	592	Rv0177	MCE associated protein	NP_214691.1	2
41310	H03	592	Rv3146	NADH dehydrogenase subunit B	NP_217662.1	2
41309	H04	592	Rv3054c	hypothetical protein	NP_217570.1	2
41320	H05	595	Rv2390c	hypothetical protein	NP_216906.1	3.173109244
41324	H06	595	Rv2882c	ribosome recycling factor	NP_217398.1	3.287394958
41314	H08	595	Rv1274	lipoprotein LprB	NP_215790.1	2
41313	H09	595	Rv1228	lipoprotein LpqX	NP_215744.1	2
41316	H10	595	Rv1749c	integral membrane protein	NP_216265.1	1.83697479
41312	H11	595	Rv0743c	hypothetical protein	NP_215257.1	2

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