

***Mycobacterium tuberculosis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 13**

Catalog No. NR-19649

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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 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 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 *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-19649.

Material Provided:

Each well of the 96-well plate contains approximately 40 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-19649 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 18 to 24 hours.

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 13, NR-19649.”

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.

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 13 (ZMTLC)¹

Clone	Well Position	ORF Length	Locus ID	Description (Gene name)	Accession Number	Average Depth of Coverage
10112	A01	352	Rv1290A	hypothetical protein	NP_215806.1	3.005681818
10098	A02	352	Rv3022A	PPE family protein	YP_177684.1	4.022727273
40352	A03	355	Rv0027	hypothetical protein	NP_214541.1	2.695774648
40361	A04	355	Rv2274c	hypothetical protein	NP_216790.1	2
40356	A05	355	Rv1495	hypothetical protein	NP_216011.1	2.205633803
40362	A06	355	Rv2375	hypothetical protein	NP_216891.1	2
40354	A07	355	Rv0715	50S ribosomal protein L24	NP_215229.1	2
40353	A08	355	Rv0531	hypothetical protein	NP_215045.1	2
40358	A09	355	Rv2044c	hypothetical protein	NP_216560.1	3.171830986
40364	A10	355	Rv3592	hypothetical protein	NP_218109.1	1.892957746
40355	A11	355	Rv0948c	hypothetical protein	NP_215463.1	2
40363	A12	355	Rv2644c	hypothetical protein	NP_217160.1	2
40365	B01	358	Rv1103c	hypothetical protein	NP_215619.1	2
40368	B02	358	Rv3706c	proline rich protein	NP_218223.1	3.268156425
40366	B03	358	Rv1344	acyl carrier protein	NP_215860.1	2
10124	B04	358	Rv1087A	hypothetical protein	YP_177637.1	3.156424581
40375	B06	361	Rv3488	hypothetical protein	NP_218005.1	2
40376	B07	361	Rv3891c	ESAT-6 like protein EsxD	NP_218408.1	2
40370	B08	361	Rv1117	hypothetical protein	NP_215633.1	2
40373	B09	361	Rv2653c	phiRv2 prophage protein	NP_217169.1	2
40372	B10	361	Rv2481c	hypothetical protein	NP_216997.1	1.900277008
40371	B11	361	Rv2034	ArsR-type repressor protein	NP_216550.1	2
40374	B12	361	Rv3065	multidrug-transport integral membrane protein MMR	YP_177922.1	2
40400	C01	364	Rv3381c	transposase	NP_217898.1	2.315934066
40386	C02	364	Rv1804c	hypothetical protein	NP_216320.1	2
40394	C03	364	Rv2798c	hypothetical protein	NP_217314.1	2
40381	C04	364	Rv1177	ferredoxin FdxC	NP_215693.1	2
40398	C05	364	Rv3325	transposase	NP_217842.1	3.302197802
40389	C06	364	Rv2278	transposase	NP_216794.1	2
40402	C07	364	Rv3474	transposase IS6110	NP_217991.1	2
40384	C08	364	Rv1757c	putative transposase	NP_216273.1	2
40383	C09	364	Rv1489c	hypothetical protein	YP_177646.1	-
40385	C10	364	Rv1763	putative transposase	NP_216279.1	2
40392	C11	364	Rv2480c	transposase	NP_216996.1	2
40395	C12	364	Rv2815c	transposase	NP_217331.1	3.318681319
40382	D01	364	Rv1370c	transposase	NP_215886.1	2

Clone	Well Position	ORF Length	Locus ID	Description (Gene name)	Accession Number	Average Depth of Coverage
40393	D02	364	Rv2648	transposase IS6110	NP_217164.1	2
40378	D03	364	Rv0795	transposase IS6110	NP_215310.1	1.60989011
40390	D04	364	Rv2348c	hypothetical protein	NP_216864.1	2
40397	D05	364	Rv3186	transposase	NP_217702.1	2
40396	D06	364	Rv3184	transposase	NP_217700.1	2
40388	D07	364	Rv2168c	transposase	NP_216684.1	2
40391	D08	364	Rv2354	transposase	NP_216870.1	2
40387	D09	364	Rv2105	transposase	NP_216621.1	1.612637363
40412	D10	367	Rv1942c	hypothetical protein	NP_216458.1	2
40413	D11	367	Rv2273	transmembrane protein	NP_216789.1	2
40416	D12	367	Rv3612c	hypothetical protein	NP_218129.1	2
40409	E01	367	Rv0692	hypothetical protein	NP_215206.1	-
40411	E02	367	Rv1351	hypothetical protein	NP_215867.1	2.689373297
40415	E03	367	Rv3183	transcriptional regulatory protein	NP_217699.1	1.662125341
40432	E04	370	Rv3686c	hypothetical protein	NP_218203.1	2
40418	E05	370	Rv0759c	hypothetical protein	NP_215273.1	2
40424	E06	370	Rv1390	DNA-directed RNA polymerase subunit omega	NP_215906.1	2
40433	E07	370	Rv3898c	hypothetical protein	NP_218415.1	2
40426	E08	370	Rv2639c	hypothetical protein	NP_217155.1	2
40422	E09	370	Rv1174c	low molecular weight T-cell antigen TB8.4	NP_215690.1	2
40417	E10	370	Rv0156	NAD(P) transhydrogenase subunit alpha	NP_214670.1	2
40431	E12	370	Rv3453	transmembrane protein	NP_217970.1	2
40419	F01	370	Rv0850	transposase	NP_215365.1	2
40425	F02	370	Rv2269c	hypothetical protein	NP_216785.1	1.905405405
40420	F03	370	Rv0991c	putative serine rich protein	NP_215506.1	3.594594595
40423	F04	370	Rv1214c	PE family protein	YP_177797.1	2
40437	F06	373	Rv3819	hypothetical protein	NP_218336.1	2
40434	F07	373	Rv1291c	hypothetical protein	NP_215807.1	3.292225201
40435	F08	373	Rv2050	hypothetical protein	NP_216566.1	2
40436	F09	373	Rv3746c	PE family protein	YP_178011.1	2
10070	F10	373	Rv1990A	transcriptional regulatory protein	NP_216506.1	2.975871314
40441	F11	376	Rv2745c	transcriptional regulatory protein	NP_217261.1	2
40442	F12	376	Rv2919c	nitrogen regulatory protein P-II GLNB	NP_217435.1	2
40440	G01	376	Rv1036c	truncated IS1560 transposase	NP_215552.1	2
40443	G02	376	Rv3004	low molecular weight protein antigen 6 (CFP-6)	NP_217520.1	2
40438	G03	376	Rv0559c	hypothetical protein	NP_215073.1	2
40444	G04	376	Rv3316	succinate dehydrogenase cytochrome B-556 subunit	NP_217833.1	2
40439	G05	376	Rv0665	hypothetical protein	NP_215179.1	2
40445	G06	376	Rv3597c	iron-regulated LSR2 protein precursor	NP_218114.1	2
40446	G07	379	Rv0572c	hypothetical protein	NP_215086.1	2
40450	G08	379	Rv2365c	hypothetical protein	NP_216881.1	2
40451	G09	379	Rv2816c	hypothetical protein	NP_217332.1	2
40449	G10	379	Rv1990c	dehydrogenase	YP_177656.1	2
40448	G11	379	Rv1271c	hypothetical protein	NP_215787.1	2
40447	G12	379	Rv1136	enoyl-CoA hydratase	NP_215652.1	2
40452	H01	379	Rv2904c	50S ribosomal protein L19	NP_217420.1	2
10075	H02	379	Rv2160c	hypothetical protein	NP_216676.1	2
40458	H03	382	Rv3182	hypothetical protein	NP_217698.1	2
40454	H04	382	Rv1991c	hypothetical protein	NP_216507.1	2
40460	H05	382	Rv3632	hypothetical protein	NP_218149.1	2
40453	H06	382	Rv0081	transcriptional regulatory protein	NP_214595.1	2

Clone	Well Position	ORF Length	Locus ID	Description (Gene name)	Accession Number	Average Depth of Coverage
40455	H07	382	Rv2007c	ferredoxin FDXA	NP_216523.1	2
40459	H08	382	Rv3353c	hypothetical protein	NP_217870.2	2
40461	H09	385	Rv0039c	transmembrane protein	NP_214553.1	1.981818182
40473	H10	385	Rv3636	transposase	NP_218153.1	2
40467	H11	385	Rv1606	phosphoribosyl-AMP cyclohydrolase	NP_216638.2	2
40469	H12	385	Rv1805c	hypothetical protein	NP_216321.1	2

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