

***Mycobacterium tuberculosis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 6**

Catalog No. NR-19642

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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-19642.

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-19642 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 6, NR-19642.”

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 6 (ZMTDF)¹

Clone	Well Position	ORF Length	Locus ID	Description (Gene name)	Accession Number	Average Depth of Coverage
73969	A01	1270	Rv1968	MCE-family protein MCE3C (mce3C)	NP_216484.1	2.71259843
74230	A02	1270	Rv2153c	undecaprenyldiphospho-muramoylpentapeptide beta-N- acetylglucosaminyltransferase (murG)	NP_216669.1	1.29133858
74007	A03	1270	Rv3899c	hypothetical protein Rv3899c	NP_218416.1	3.44251969
73981	A04	1273	Rv0597c	hypothetical protein Rv0597c	NP_215111.1	1.17910448
74245	A05	1276	Rv0914c	acetyl-CoA acetyltransferase	NP_215429.1	2.92319749
74278	A06	1279	Rv2627c	hypothetical protein Rv2627c	NP_217143.1	2.08131353
74153	A07	1279	Rv2943	IS1533 transposase	NP_217459.1	2.69898358
74011	A08	1279	Rv3578	arsenical PUMP integral membrane protein ArsB2 (arsB2)	NP_218095.1	1.30492572
74093	A09	1279	Rv3621c	PPE family protein (PPE65)	YP_177998.1	2.57154027
74105	A10	1279	Rv3712	ligase	NP_218229.1	2.84519156
73940	A11	1282	Rv1872c	L-lactate dehydrogenase (cytochrome) LldD2 (lldD2)	NP_216388.1	1.14898596
74242	A12	1285	Rv3775	lipase LipE (lipE)	NP_218292.1	2.06692607
74161	B01	1291	Rv0545c	inorganic phosphate transporter (pitA)	NP_215059.1	1.15491867
74053	B02	1300	Rv1426c	esterase LipO (lipO)	NP_215942.1	1.76
74067	B03	1303	Rv3190c	hypothetical protein Rv3190c	NP_217706.1	3.09593246
74071	B04	1309	Rv0915c	PPE family protein (PPE14)	YP_177765.1	3.12605042
73995	B05	1313	Rv1969	MCE-family protein MCE3D (mce3D)	NP_216485.1	3.09444021
74051	B06	1318	Rv2677c	protoporphyrinogen Oxidase (hemY)	YP_177675.1	1.82397572
74305	B07	1324	Rv1771	oxidoreductase	NP_216287.1	1.10574018
74221	B08	1324	Rv2266	cytochrome P450 124 CYP124 (cyp124)	NP_216782.1	2.78776435
73936	B09	1333	Rv0896	type II citrate synthase (gltA)	NP_215411.1	1.38934734
74273	B10	1339	Rv1095	PhoH-like protein PhoH2 (phosphate starvation-inducible protein PsiH) (phoH)	NP_215611.1	2.6982823
74013	B11	1339	Rv2673	integral membrane protein	NP_217189.1	2.78416729
73941	B12	1339	Rv3545c	cytochrome P450 125 (cyp125)	NP_218062.1	2.74159821
74295	C01	1342	Rv0576	ArsR family transcriptional regulator	NP_215090.1	2.78539493
74089	C02	1345	Rv1232c	hypothetical protein Rv1232c	NP_215748.1	2.20817844
73958	C03	1345	Rv1783	hypothetical protein Rv1783	NP_216299.1	2.07806691
74019	C04	1351	Rv1568	adenosylmethionine--8-amino-7-oxononanoate transaminase (bioA)	NP_216084.1	2.45965951
74027	C05	1354	Rv2246	3-oxoacyl-(acyl carrier protein) synthase II (kasB)	NP_216762.1	2.95199409
74185	C06	1354	Rv2681	hypothetical protein Rv2681	NP_217197.1	2.6661743
74234	C07	1357	Rv0449c	hypothetical protein Rv0449c	NP_214963.1	2.0412675
74285	C08	1357	Rv2041c	sugar-binding lipoprotein	NP_216557.1	2.76934414
73926	C09	1360	Rv2318	periplasmic sugar-binding lipoprotein UspC (uspC)	NP_216834.1	1.94926471
73962	C10	1360	Rv3693	hypothetical protein Rv3693	NP_218210.1	1.82647059
74445	C11	1363	Rv0393	13E12 repeat family protein	NP_214907.1	2.26118855

Clone	Well Position	ORF Length	Locus ID	Description (Gene name)	Accession Number	Average Depth of Coverage
74403	C12	1375	Rv2508c	hypothetical protein Rv2508c	NP_217024.1	2.76363636
74646	D01	1378	Rv0902c	two component sensor histidine kinase PRRB (prbB)	NP_215417.1	2.00435414
74542	D02	1378	Rv3371	hypothetical protein Rv3371	NP_217888.1	1.73222061
74381	D03	1387	Rv3835	hypothetical protein Rv3835	NP_218352.1	2.58399423
74326	D04	1405	Rv0955	integral membrane protein	NP_215470.1	2.46690391
74449	D05	1405	Rv1257c	oxidoreductase	NP_215773.1	3.59572954
74613	D06	1417	Rv2791c	transposase	NP_217307.1	3.27875794
74642	D07	1420	Rv3881c	hypothetical protein Rv3881c	NP_218398.1	2.66056338
74511	D08	1423	Rv1726	oxidoreductase	NP_216242.1	4.11665495
74661	D09	1423	Rv2615c	PE-PGRS family protein (PE_PGRS45)	YP_177895.1	2.91848208
74333	D10	1429	Rv0096	PPE family protein (PPE1)	YP_177690.1	3.31980406
74621	D11	1429	Rv0573c	nicotinate phosphoribosyltransferase	NP_215087.1	3.51574528
74371	D12	1429	Rv1537	DNA polymerase IV (dinX)	NP_216053.2	3.5703289
74386	E01	1432	Rv0983	serine protease PepD (pepD)	NP_215498.1	2.47835196
74349	E02	1432	Rv1077	cystathionine beta-synthase CBS (Serine sulfhydrylase) (Beta-thionase) (hemoprotein H-450) (cbs)	YP_177782.1	3.2122905
74658	E03	1435	Rv0296c	sulfatase	YP_177712.1	2.58885017
74549	E04	1438	Rv2462c	trigger factor (tig)	NP_216978.1	3.59318498
74628	E05	1441	Rv3448	integral membrane protein	NP_217965.1	1.10825815
74319	E06	1444	Rv1614	prolipoprotein diacylglycerol transferase (lgt)	NP_216130.1	3.64265928
74577	E07	1444	Rv3820c	polyketide synthase associated protein PapA2 (papA2)	YP_178020.1	3.55955679
74514	E08	1447	Rv0221	hypothetical protein Rv0221	NP_214735.1	2.5252246
74358	E09	1447	Rv0261c	integral membrane nitrite extrusion protein NarK3 (narK3)	NP_214775.1	2.57636489
74619	E10	1447	Rv2812	transposase	NP_217328.1	3.96199032
74601	E11	1447	Rv3580c	cysteinyI-tRNA synthetase (cysS)	YP_177992.1	1.02280581
74397	E12	1450	Rv0392c	membrane NADH dehydrogenase (ndhA)	NP_214906.1	1.01172414
74650	F01	1450	Rv2522c	hypothetical protein Rv2522c	NP_217038.1	1.93103448
74683	F02	1450	Rv2974c	hypothetical protein Rv2974c	NP_217490.1	2.62758621
74606	F03	1450	Rv3450c	hypothetical protein Rv3450c	NP_217967.1	2.5937931
74671	F04	1459	Rv1576c	phiRV1 phage protein	NP_216092.1	3.91432488
74421	F05	1459	Rv2888c	Amidase (amiC)	NP_217404.1	3.60246744
74537	F06	1462	Rv1098c	fumarate hydratase (fumC)	NP_215614.1	3.02941176
74379	F07	1465	Rv2655c	phiRv2 prophage protein	NP_217171.1	3.90443686
74673	F08	1465	Rv3375	amidase AmiD (amiD)	NP_217892.1	3.46416382
74405	F09	1465	Rv3764c	two component sensor kinase	NP_218281.1	3.48191126
74534	F10	1480	Rv2425c	hypothetical protein Rv2425c	NP_216941.1	2.23243243
74589	F11	1480	Rv3869	hypothetical protein Rv3869	NP_218386.1	2.76216216
74630	F12	1480	Rv3907c	poly(A) polymerase (pcnA)	YP_178026.1	2.49324324
74559	G01	1483	Rv0591	MCE-family protein MCE2C (mce2C)	NP_215105.1	3.55428186
74354	G02	1483	Rv2434c	transmembrane protein	NP_216950.1	2.57518543
74495	G03	1495	Rv1844c	6-phosphogluconate dehydrogenase (gnd1)	YP_177848.1	3.63745819
74419	G04	1498	Rv1129c	transcriptional regulator protein	NP_215645.1	3.81041389
74361	G05	1501	Rv0442c	PPE family protein (PPE10)	YP_177726.1	3.39573618
74529	G06	1501	Rv1226c	hypothetical protein Rv1226c	NP_215742.1	3.42771486
74637	G07	1507	Rv2127	L-asparagine permease ansP1 (ansP1)	YP_177863.1	3.3384207
74329	G08	1507	Rv3854c	monooxygenase ETHA (ethA)	NP_218371.1	3.52753816
74677	G09	1516	Rv0143c	transmembrane protein	NP_214657.1	3.26253298
74505	G10	1516	Rv2519	PE family protein (PE26)	YP_177888.1	3.18205805
74437	G11	1516	Rv3059	cytochrome P450 136 (cyp136)	NP_217575.1	2.94129288
74434	G12	1519	Rv0343	isoniazid inducible gene protein INIC (iniC)	NP_214857.1	2.57077024
74690	H01	1519	Rv3303c	flavoprotein disulfide reductase (lpdA)	NP_217820.1	2.3805135

Clone	Well Position	ORF Length	Locus ID	Description (Gene name)	Accession Number	Average Depth of Coverage
74502	H02	1522	Rv0255c	cobyric acid synthase (cobQ1)	YP_177703.1	2.4434954
74429	H03	1522	Rv1867	acetyl-CoA acetyltransferase	NP_216383.1	3.08081472
74413	H04	1522	Rv2152c	UDP-N-acetylmuramate--L-alanine ligase (murC)	NP_216668.1	3.53088042
74321	H05	1522	Rv3011c	aspartyl/glutamyl-tRNA amidotransferase subunit A (gatA)	NP_217527.1	2.51445466
74465	H06	1522	Rv3293	piperideine-6-carboxylic acid dehydrogenase (pcd)	YP_177953.1	2.84362681
74485	H07	1525	Rv0904c	putative acetyl-coenzyme A carboxylase carboxyl transferase (subunit BETA) ACCD3 (ACCASE BETA chain) (accD3)	NP_215419.1	3.33377049
74598	H08	1525	Rv2725c	GTP-binding protein HflX (hflX)	NP_217241.1	2.54557377
74341	H09	1525	Rv3083	monooxygenase	NP_217599.1	3.32131148
74930	H10	1525	Rv3175	amidase	NP_217691.1	2.44918033
74890	H11	1528	Rv0109	PE-PGRS family protein (PE_PGRS1)	YP_177692.1	1.56479058
74945	H12	1531	Rv1376	hypothetical protein Rv1376	NP_215892.1	3.29196604

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