

***Rickettsia prowazekii*, Strain Madrid E, Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 6**

Catalog No. NR-19454

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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 *Rickettsia prowazekii* (*R. prowazekii*), strain Madrid E, Gateway® clone set consists of approximately 750 sequence validated clones from *R. prowazekii*, strain Madrid E, cloned in *Escherichia coli* (*E. coli*) DH10B-T1 cells. Each open reading frame was constructed in vector [pDONR™221](#) (Invitrogen™) with an ATG start codon and no stop codon. The sequence was validated by full length sequencing of each 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-19454.

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-19454 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 24 hours.

Citation:

Acknowledgment for publications should read “The following reagent was obtained through BEI Resources, NIAID, NIH: *Rickettsia prowazekii*, Strain Madrid E, Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 6, NR-19454.”

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:

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References:

1. Andersson, S. G., et al. "The Genome Sequence of *Rickettsia prowazekii* and the Origin of Mitochondria." *Nature* 396 (1998): 133-140. PubMed: 9823893.

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Table 1: *Rickettsia prowazekii*, Strain Madrid E, Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 6 (ZRPAF)¹

Clone	Well Position	Locus ID	Description	ORF Length	Accession Number	Average Depth of Coverage
46415	A01	RP576	protein export protein PRSA precursor (prsA)	883	NP_220947.1	3.654977
46479	A02	RP686	4-hydroxybenzoate polyprenyltransferase	883	NP_221047.1	2.507412
46282	A03	RP332	hypothetical protein RP332	889	NP_220715.1	3.391451
46482	A04	RP496	hypothetical protein RP496	892	NP_220872.1	1.9787
46197	A05	RP059	stage 0 sporulation protein J (spo0J)	895	NP_220453.1	4.158659
46346	A06	RP123	HFLC protein (hflC)	895	NP_220515.1	2.167598
46290	A07	RP581	hypothetical protein RP581	895	NP_220952.1	2.185475
46227	A08	RP254	UDP-3-O-	901	NP_220639.1	3.826859
46311	A09	RP489	hypothetical protein RP489	901	NP_220866.1	4.135405
46299	A10	RP850	glycyl-tRNA synthetase subunit alpha	904	NP_221198.1	3.568584
46471	A11	RP428	branched-chain amino acid aminotransferase	907	NP_220809.1	1.942668
46463	A12	RP218	hypothetical protein RP218	913	NP_220604.1	4.143483
46239	B01	RP432	succinyl-CoA synthetase subunit alpha	913	NP_220813.1	4.46988
46215	B02	RP303	RNA polymerase factor sigma-32	916	NP_220687.1	1.870087
46247	B03	RP444	hypothetical protein RP444	916	NP_220825.1	1.995633
46475	B04	RP682	hypothetical protein RP682	916	NP_221043.1	1.99345
46351	B05	RP301	FAD-dependent thymidylate synthase	919	NP_220685.1	3.732318
46363	B06	RP429	dihydrodipicolinate synthase	919	NP_220810.1	4.131665
46106	B07	RP005	hypothetical protein RP005	922	NP_220401.1	2.614967
46486	B08	RP118	GTP-binding protein Era	922	NP_220510.1	1.573753
46191	B09	RP673	hypothetical protein RP673	922	NP_221034.1	3.801518
46431	B10	RP086	30S ribosomal protein S2	925	NP_220479.1	1.977297
46127	B11	RP742	lipoyl synthase	928	NP_221094.1	4.588362
46186	B12	RP786	hypothetical protein RP785	928	NP_221136.1	2.02694
46159	C01	RP295	hypothetical protein RP295	931	NP_220679.1	3.988185
46427	C02	RP014	hypothetical protein RP014	934	NP_220409.1	2.762313
46145	C03	RP466	porphobilinogen deaminase	934	NP_220846.1	4.167024
46391	C04	RP832	protein P34 (p34)	937	NP_221181.1	3.533618
46459	C05	RP016	hypothetical protein RP016	943	NP_220411.1	1.962884
46411	C06	RP209	methionyl-tRNA formyltransferase	946	NP_220597.1	2
46451	C07	RP258	hypothetical protein RP258	946	NP_220643.1	3.991543
46367	C08	RP367	hypothetical protein RP367	946	NP_220750.1	4.062368
46235	C09	RP740	hemolysin C (tlyC)	946	NP_221092.1	3.884778
46286	C10	RP525	protease IV (sppA)	952	NP_220898.1	2.163866

Clone	Well Position	Locus ID	Description	ORF Length	Accession Number	Average Depth of Coverage
46295	C11	RP817	site-specific tyrosine recombinase XerC	952	NP_221166.1	3.013655
46167	C12	RP389	penicillin-binding protein DACF precursor (dacF)	955	NP_220770.1	3.118325
46434	D01	RP569	S-adenosyl-methyltransferase MraW	955	NP_220941.1	2.009424
46278	D02	RP443	hypothetical protein RP443	964	NP_220824.1	2.048755
46343	D03	RP248	UDP-N-acetylenolpyruvoylglucosamine reductase	967	NP_220633.1	3.619442
46319	D04	RP346	protoheme IX farnesyltransferase	967	NP_220729.1	4.459152
46355	D05	RP445	thioredoxin reductase (trxB1)	967	NP_220826.1	3.509824
46303	D06	RP515	bifunctional 5,10-methylene-tetrahydrofolate dehydrogenase/ 5,10-methylene-tetrahydrofolate cyclohydrolase	967	NP_220891.1	3.541882
46123	D07	RP114	preprotein translocase subunit SecF	970	NP_220506.1	3.297938
46447	D08	RP510	tRNA delta(2)-isopentenylpyrophosphate transferase	970	NP_220886.1	1.9
46370	D09	RP197	hypothetical protein RP197	976	NP_220585.1	2.155738
46439	D10	RP406	cytochrome C oxidase polypeptide II (coxB)	976	NP_220787.1	3.815574
46422	D11	RP846	pseudouridylate synthase	976	NP_221194.1	2.143443
46327	D12	RP376	malate dehydrogenase	979	NP_220759.1	3.552605
46378	E01	RP706	hypothetical protein RP706	979	NP_221065.1	1.930541
46359	E02	RP186	protease DO (htrA)	982	NP_220575.1	5.117108
46383	E03	RP442	NAD(P)H-dependent glycerol-3-phosphate dehydrogenase	985	NP_220823.1	3.93002
46258	E04	RP402	hypothetical protein RP402	988	NP_220783.1	2.071862
46407	E05	RP772	3-oxoacyl-(acyl carrier protein) synthase III	988	NP_221123.1	3.245951
46403	E06	RP121	hypothetical protein RP121	991	NP_220513.1	1.961655
46198	E07	RP339	minor teichoic acids biosynthesis protein ggab (ggab)	991	NP_220722.1	3.696266
46271	E08	RP505	arabinose-5-phosphate isomerase	994	NP_220881.1	4.172032
46243	E09	RP701	hypothetical protein RP701	994	NP_221061.1	3.28672
46183	E10	RP735	acyl-carrier-protein S-malonyltransferase	994	NP_221088.1	3.34004
46203	E11	RP390	rare lipoprotein A precursor (rlpA)	997	NP_220771.1	3.38014
46250	E12	RP249	D-alanine--D-alanine ligase	1000	NP_220634.1	2.008
46223	F01	RP305	hypothetical protein RP305	1003	NP_220689.1	1.918245
46208	F02	RP853	hypothetical protein RP853	1003	NP_221201.1	3.360917
46257	F03	RP372	hypothetical protein RP372	1006	NP_220755.1	4.120278
46322	F04	RP512	ribonucleotide-diphosphate reductase subunit beta	1009	NP_220888.1	1.751239
46155	F05	RP719	tetraacyldisaccharide 4'-kinase	1012	NP_221074.1	1.866601
46175	F06	RP261	pyruvate dehydrogenase E1 component, alpha subunit precursor (pdhA)	1015	NP_220646.1	3.285714
46335	F07	RP262	pyruvate dehydrogenase subunit beta	1015	NP_220647.1	2.669634
46399	F08	RP825	undecaprenyl-phosphate alpha-N-acetylglucosaminyltransferase (mraY2)	1015	NP_221174.1	4.153695
46468	F09	RP479	octaprenyl-diphosphate synthase (ispB)	1018	NP_220858.1	4.049116
46178	F10	RP564	hypothetical protein RP564	1018	NP_220936.1	2.128684

Clone	Well Position	Locus ID	Description	ORF Length	Accession Number	Average Depth of Coverage
46375	F11	RP468	tryptophanyl-tRNA synthetase	1027	NP_220847.1	3.251217
46339	F12	RP539	delta-aminolevulinic acid dehydratase	1027	NP_220912.1	3.822785
46231	G01	RP395	hypothetical protein RP395	1036	NP_220776.1	3.939189
46266	G02	RP449	threonine dehydratase	1036	NP_220830.1	3.925676
46262	G03	RP092	hypothetical protein RP092	1039	NP_220485.1	1.787295
46146	G04	RP257	cytochrome C oxidase assembly protei (coxW)	1039	NP_220642.1	4.638114
46134	G05	RP292	type IV secretion system ATPase VirB11	1039	NP_220676.1	4.682387
46315	G06	RP472	hypothetical protein RP472	1039	NP_220851.1	1.748797
46111	G07	RP514	thioredoxin reductase (trxB2)	1039	NP_220890.1	3.304139
46419	G08	RP361	site-specific tyrosine recombinase XerD	1042	NP_220744.2	1.712092
46170	G09	RP316	aspartate-semialdehyde dehydrogenase	1051	NP_220699.1	1.683159
46394	G10	RP340	hypothetical protein RP340	1051	NP_220723.1	4.782112
46590	G11	RP414	CAPM protein (capM2)	1051	NP_220795.1	1.747859
46535	G12	RP217	cytochrome D ubiquinol oxidase subunit II (cydB)	1054	NP_220603.1	1.767552
46721	H01	RP738	poly-BETA-hydroxybutyrate polymerase (phbC1)	1054	NP_221090.1	5.132827
46563	H02	RP796	NADH dehydrogenase subunit H	1054	NP_221146.1	1.832068
46627	H03	RP322	hypothetical protein RP322	1057	NP_220705.1	3.190161
46614	H04	RP635	DNA-directed RNA polymerase subunit alpha	1057	NP_220999.1	1.748344
46687	H05	RP761	recombinase A	1057	NP_221113.1	2.552182
46862	H06	RP081	190 kDa antigen precursor	1060	NP_220474.1	1.712264
46498	H07	RP333	capsular polysaccharide biosynthesis protein CapD	1060	NP_220716.1	4.876415
46710	H08	RP386	Holliday junction DNA helicase RuvB	1063	NP_220767.1	1.632173
46642	H09	RP452	isopentenyl pyrophosphate isomerase	1063	NP_220833.1	3.877705
46623	H10	RP122	HFLK protein (hflK)	1069	NP_220514.1	4.624883
46638	H11	RP885	uroporphyrinogen decarboxylase	1072	NP_221231.1	4.130597
46503	H12	RP009	UDP-3-O-	1075	NP_220405.1	1.655814

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