

***Bacillus anthracis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 9**

Catalog No. NR-19733

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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 *Bacillus anthracis* (*B. anthracis*) Gateway® clone set consists of 58 plates which contain 5341 sequence validated clones from *B. anthracis*, strains Ames (5139 clones), Sterne (107 clones; contains plasmid pXO1 only) and A2012 (95 clones; contains plasmid pXO2 only) 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 library was independently cloned and sequence verified by the [Harvard Institute of Proteomics](#). 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 a Harvard-modified *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-19733.

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-19733 was packaged aseptically in 96-well plates. 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 containing 50 µg/mL kanamycin

LB 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 24 hours.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Bacillus anthracis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 9, NR-19733."

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. Read, T. D., et al. "The Genome Sequence of *Bacillus anthracis* Ames and Comparison to Closely Related Bacteria." *Nature* 423 (2003): 81-86. PubMed: 12721629.
2. Read, T. D., et al. "Comparative Genome Sequencing for Discovery of Novel Polymorphisms in *Bacillus anthracis*." *Science* 296 (2002): 2028-2033. PubMed: 12004073.

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Table 1: *Bacillus anthracis*, Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 9 (QMG002850)¹

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
48029	A02	BA4349	hypothetical protein	Chromosome (NC_003997)	NP_846581.1	247852
50514	A03	BA3893	cell wall hydrolase, putative	Chromosome (NC_003997)	NP_846140.1	247860
50904	A04	BA3911	renal dipeptidase family protein	Chromosome (NC_003997)	NP_846158.1	247872
48818	A05	BA0120	ribosomal protein L14	Chromosome (NC_003997)	NP_842688.1	247881
51465	A06	BA4352	acetylornithine aminotransferase	Chromosome (NC_003997)	NP_846584.1	247892
49369	A07	BA0127	ribosomal protein S5	Chromosome (NC_003997)	NP_842695.1	247901
49812	A08	BA1510	negative regulator of competence MecA, putative	Chromosome (NC_003997)	NP_843961.1	247912
50083	A09	BA3888	hypothetical protein	Chromosome (NC_003997)	NP_846135.1	247928
47731	A10	BA2325	hypothetical protein	Chromosome (NC_003997)	NP_844710.1	246533
50006	A11	BA2296	CoA-transferase, beta subunit	Chromosome (NC_003997)	NP_844681.1	246543
47996	A12	BA0448	hypothetical protein	Chromosome (NC_003997)	NP_842989.1	246553
50159	B01	BA3903	bacteriocin ABC transporter, ATP-binding protein, putative	Chromosome (NC_003997)	NP_846150.1	247839
48042	B02	BA3889	hypothetical protein	Chromosome (NC_003997)	NP_846136.1	247853
48316	B03	BA0104	ribosomal protein L7A family	Chromosome (NC_003997)	NP_842672.1	247861
48565	B04	BA0109	ribosomal protein S10	Chromosome (NC_003997)	NP_842677.1	247873
51105	B05	BA3891	1-phosphatidylinositol phosphodiesterase	Chromosome (NC_003997)	NP_846138.1	247882
49120	B06	BA0129	ribosomal protein L15	Chromosome (NC_003997)	NP_842697.1	247893
49526	B07	BA0125	ribosomal protein L6	Chromosome (NC_003997)	NP_842693.1	247903
51832	B08	BA4345	Na ⁺ /H ⁺ antiporter NhaC	Chromosome (NC_003997)	NP_846577.1	247913
47626	B09	BA1845	hypothetical protein	Chromosome (NC_003997)	NP_844264.1	246525
49814	B10	BA2324	hypothetical protein	Chromosome (NC_003997)	NP_844709.1	246534
47917	B11	BA0447	hypothetical protein	Chromosome (NC_003997)	NP_842988.1	246544
48047	B12	BA0440	hypothetical protein	Chromosome (NC_003997)	NP_842981.1	246555
50199	C01	BA3921	oxidoreductase, short-chain dehydrogenase/reductase family	Chromosome (NC_003997)	NP_846168.1	247841
50465	C02	BA3919	ACT domain protein	Chromosome (NC_003997)	NP_846166.1	247854
48325	C03	BA3920	hypothetical protein	Chromosome (NC_003997)	NP_846167.1	247863
50986	C04	BA4351	ornithine carbamoyltransferase	Chromosome (NC_003997)	NP_846583.1	247874
48879	C05	BA4342	hypothetical protein	Chromosome (NC_003997)	NP_846575.1	247883
51530	C06	BA4339	8-amino-7-oxononanoate synthase	Chromosome (NC_003997)	NP_846572.1	247894
51700	C07	BA3922	zinc protease, insulinase family	Chromosome (NC_003997)	NP_846169.1	247904

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
49894	C08	BA0110	ribosomal protein L3	Chromosome (NC_003997)	NP_842678.1	247916
49516	C09	BA1846	peptide methionine sulfoxide reductase	Chromosome (NC_003997)	NP_844265.1	246526
49854	C10	BA4213	hypothetical protein	Chromosome (NC_003997)	NP_846448.1	246536
47931	C11	BA0441	hypothetical protein	Chromosome (NC_003997)	NP_842982.1	246546
50385	C12	BA4204	oxidoreductase, short chain dehydrogenase/reductase family	Chromosome (NC_003997)	NP_846440.1	246556
47934	D01	BA1517	hypothetical protein	Chromosome (NC_003997)	NP_843968.1	247842
48080	D02	BA0118	ribosomal protein L29	Chromosome (NC_003997)	NP_842686.1	247855
50723	D03	BA3909	pyruvate ferredoxin oxidoreductase, beta subunit, putative	Chromosome (NC_003997)	NP_846156.1	247866
48574	D04	BA0121	ribosomal protein L24	Chromosome (NC_003997)	NP_842689.1	247875
48942	D05	BA0124	ribosomal protein S8	Chromosome (NC_003997)	NP_842692.1	247885
49161	D06	BA4348	transcriptional regulator, MarR family	Chromosome (NC_003997)	NP_846580.1	247895
49525	D07	BA0122	ribosomal protein L5	Chromosome (NC_003997)	NP_842690.1	247905
49964	D08	BA0131	adenylate kinase	Chromosome (NC_003997)	NP_842699.1	247918
47646	D09	BA4234	hypothetical protein	Chromosome (NC_003997)	NP_846469.1	246527
47767	D10	BA0431	hypothetical protein	Chromosome (NC_003997)	NP_842973.1	246537
47959	D11	BA0444	hypothetical protein	Chromosome (NC_003997)	NP_842985.1	246548
48085	D12	BA2318	DNA-binding protein	Chromosome (NC_003997)	NP_844703.1	246557
47975	E01	BA0128	ribosomal protein L30	Chromosome (NC_003997)	NP_842696.1	247844
50501	E02	BA4357	oxidoreductase, short-chain dehydrogenase/reductase family	Chromosome (NC_003997)	NP_846589.1	247856
48376	E03	BA0119	ribosomal protein S17	Chromosome (NC_003997)	NP_842687.1	247867
51056	E04	BA1516	L-asparaginase	Chromosome (NC_003997)	NP_843967.1	247876
49040	E05	BA0105	ribosomal protein S12	Chromosome (NC_003997)	NP_842673.1	247887
51524	E06	BA0108	translation elongation factor Tu	Chromosome (NC_003997)	NP_842676.1	247896
49542	E07	BA3906	spore coat protein E	Chromosome (NC_003997)	NP_846153.1	247907
49999	E08	BA4361	L-serine dehydratase, iron-sulfur-dependent, beta subunit	Chromosome (NC_003997)	NP_846592.1	247920
49537	E09	BA2287	hypothetical protein	Chromosome (NC_003997)	NP_844672.1	246528
47783	E10	BA0449	hypothetical protein	Chromosome (NC_003997)	NP_842990.1	246538
50324	E11	BA4222	hypothetical protein	Chromosome (NC_003997)	NP_846457.1	246549
48096	E12	BA4224	hypothetical protein	Chromosome (NC_003997)	NP_846459.1	246558
48011	F01	BA4356	hypothetical protein	Chromosome (NC_003997)	NP_846588.1	247846
48140	F02	BA1512	hypothetical protein	Chromosome (NC_003997)	NP_843963.1	247857
48442	F03	BA0114	ribosomal protein S19	Chromosome (NC_003997)	NP_842682.1	247869
51079	F04	BA1515	pyridine nucleotide-disulfide oxidoreductase family protein	Chromosome (NC_003997)	NP_843966.1	247878
51256	F05	BA1520	isopentenyl-diphosphate delta-isomerase	Chromosome (NC_003997)	NP_843971.1	247888
49231	F06	BA1514	N-acetylmuramoyl-L-alanine amidase, family 2	Chromosome (NC_003997)	NP_843965.1	247897
51790	F07	BA3894	hypothetical protein	Chromosome (NC_003997)	NP_846141.1	247909
49990	F08	BA0116	ribosomal protein S3	Chromosome (NC_003997)	NP_842684.1	247922
47658	F09	BA4221	hypothetical protein	Chromosome (NC_003997)	NP_846456.1	246529
47789	F10	BA2323	hypothetical protein	Chromosome (NC_003997)	NP_844708.1	246540
47966	F11	BA2305	hypothetical protein	Chromosome (NC_003997)	NP_844690.1	246550

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
50651	F12	BA4227	maltosaccharide ABC transporter, permease protein	Chromosome (NC_003997)	NP_846462.1	246559
47993	G01	BA0123	ribosomal protein S14	Chromosome (NC_003997)	NP_842691.1	247848
50500	G02	BA3913	phosphoesterase family protein	Chromosome (NC_003997)	NP_846160.1	247858
50874	G03	BA3918	hypothetical protein	Chromosome (NC_003997)	NP_846165.1	247870
48791	G04	BA0126	ribosomal protein L18	Chromosome (NC_003997)	NP_842694.1	247879
49080	G05	BA3907	hypothetical protein	Chromosome (NC_003997)	NP_846154.1	247889
51600	G06	BA4354	glutamate N-acetyltransferase/amino-acid acetyltransferase	Chromosome (NC_003997)	NP_846586.1	247898
49692	G07	BA3917	CDP-diacylglycerol--glycerol-3-phosphate 3-phosphatidyltransferase	Chromosome (NC_003997)	NP_846164.1	247910
50011	G08	BA3900	lantibiotic biosynthesis DNA-binding response regulator, putative	Chromosome (NC_003997)	NP_846147.1	247924
47704	G09	BA2293	hypothetical protein	Chromosome (NC_003997)	NP_844678.1	246531
50007	G10	BA2303	hypothetical protein	Chromosome (NC_003997)	NP_844688.1	246541
47984	G11	BA2320	hypothetical protein	Chromosome (NC_003997)	NP_844705.1	246551
48084	G12	BA1844	hypothetical protein	Chromosome (NC_003997)	NP_844263.1	246560
48010	H01	BA3897	hypothetical protein	Chromosome (NC_003997)	NP_846144.1	247850
48256	H02	BA1513	hypothetical protein	Chromosome (NC_003997)	NP_843964.1	247859
48486	H03	BA0112	ribosomal protein L23	Chromosome (NC_003997)	NP_842680.1	247871
51095	H04	BA3898	choloylglycine hydrolase family protein	Chromosome (NC_003997)	NP_846145.1	247880
51444	H05	BA1519	ribosomal protein S1	Chromosome (NC_003997)	NP_843970.1	247890
51618	H06	BA3916	competence/damage-inducible protein CinA	Chromosome (NC_003997)	NP_846163.1	247900
51824	H07	BA3899	lantibiotic biosynthesis sensor histidine kinase, putative	Chromosome (NC_003997)	NP_846146.1	247911
50061	H08	BA1518	cytidylate kinase	Chromosome (NC_003997)	NP_843969.1	247926
49777	H09	BA0446	hypothetical protein	Chromosome (NC_003997)	NP_842987.1	246532
47904	H10	BA0430	hypothetical protein	Chromosome (NC_003997)	NP_842972.1	246542
50352	H11	BA2308	sporulation-control protein Spo0M, putative	Chromosome (NC_003997)	NP_844693.1	246552

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