

***Bacillus anthracis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 8**

Catalog No. NR-19732

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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-19732.

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-19732 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 8, NR-19732."

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 8 (QMG002849)¹

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
49038	A02	BA5129	hypothetical protein	Chromosome (NC_003997)	NP_847315.1	240452
49290	A03	BA2777	hypothetical protein	Chromosome (NC_003997)	NP_845126.1	240460
49966	A04	BA0905	hypothetical protein	Chromosome (NC_003997)	NP_843415.1	240472
50425	A05	BA4188	hydrolase, haloacid dehalogenase-like family	Chromosome (NC_003997)	NP_846424.1	246612
50885	A06	BA1831	cysteine synthase A	Chromosome (NC_003997)	NP_844250.1	246622
51131	A07	BA4149	hydrolase, carbon-nitrogen family	Chromosome (NC_003997)	NP_846387.1	246637
48691	A08	BA1782	transposase, IS605 family, OrfA	Chromosome (NC_003997)	NP_844208.1	246646
51416	A09	BA4193	peptidase, M20/M25/M40 family	Chromosome (NC_003997)	NP_846429.1	246655
49362	A10	BA4160	hypothetical protein	Chromosome (NC_003997)	NP_846398.1	246666
49693	A11	BA4144	phosphoglycerate mutase family protein	Chromosome (NC_003997)	NP_846382.1	246678
51932	A12	BA1822	carbon starvation protein A, putative	Chromosome (NC_003997)	NP_844243.1	246687
51226	B01	BA2775	TPP-dependent acetoin dehydrogenase E1 beta-subunit	Chromosome (NC_003997)	NP_845124.1	240445
51453	B02	BA5138	aminotransferase, classes I and II	Chromosome (NC_003997)	NP_847324.1	240453
49410	B03	BA2779	hypothetical protein	Chromosome (NC_003997)	NP_845128.1	240462
50008	B04	BA2791	ribose 5-phosphate isomerase	Chromosome (NC_003997)	NP_845140.1	240474
47755	B05	BA4176	hypothetical protein	Chromosome (NC_003997)	NP_846414.1	246613
48095	B06	BA4163	hypothetical protein	Chromosome (NC_003997)	NP_846401.1	246625
48446	B07	BA1820	hypothetical protein	Chromosome (NC_003997)	NP_844241.1	246638
51359	B08	BA1792	sensor histidine kinase, putative	Chromosome (NC_003997)	NP_844217.1	246647
49122	B09	BA1788	hypothetical protein	Chromosome (NC_003997)	NP_844214.1	246656
49425	B10	BA1832	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_844251.1	246668
51792	B11	BA1784	D-serine dehydratase	Chromosome (NC_003997)	NP_844210.1	246679
50103	B12	BA1838	hypothetical protein	Chromosome (NC_003997)	NP_844257.1	246690
48893	C01	BA5141	kinase-associated protein B	Chromosome (NC_003997)	NP_847327.1	240446
49094	C02	BA2781	mutT/nudix family protein	Chromosome (NC_003997)	NP_845130.1	240454
51785	C03	BA5143	arsenical pump family protein	Chromosome (NC_003997)	NP_847329.1	240463
50036	C04	BA0904	membrane protein, putative	Chromosome (NC_003997)	NP_843414.1	240476
47754	C05	BA4174	hypothetical protein	Chromosome (NC_003997)	NP_846412.1	246615
50996	C06	BA1842	dehydrogenase, putative	Chromosome (NC_003997)	NP_844261.1	246630
51158	C07	BA1852	ketol-acid reductoisomerase	Chromosome (NC_003997)	NP_844269.1	246639

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
48812	C08	BA4148	hypothetical protein	Chromosome (NC_003997)	NP_846386.1	246648
49147	C09	BA4196	CBS domain protein	Chromosome (NC_003997)	NP_846432.1	246658
51644	C10	BA1828	GTP-binding protein	Chromosome (NC_003997)	NP_844248.1	246669
49862	C11	BA4152	cytochrome c oxidase, subunit III	Chromosome (NC_003997)	NP_846390.1	246680
50156	C12	BA1836	polysaccharide deacetylase, putative	Chromosome (NC_003997)	NP_844255.1	246692
51255	D01	BA0897	Peptidase, family M20/M25/M40 protein	Chromosome (NC_003997)	NP_843408.1	240447
51551	D02	BA2774	dihydrolipoamide acetyltransferase	Chromosome (NC_003997)	NP_845123.1	240455
49534	D03	BA5139	superoxide dismutase, Cu-Zn	Chromosome (NC_003997)	NP_847325.1	240464
50203	D04	BA5145	hypothetical protein	Chromosome (NC_003997)	NP_847331.1	240484
50724	D05	BA4192	cytochrome d ubiquinol oxidase, subunit II-related protein	Chromosome (NC_003997)	NP_846428.1	246616
48240	D06	BA4179	hypothetical protein	Chromosome (NC_003997)	NP_846417.1	246631
48483	D07	BA4159	hypothetical protein	Chromosome (NC_003997)	NP_846397.1	246640
51520	D08	BA4158	cell division protein, FtsW/RodA/SpoVE family	Chromosome (NC_003997)	NP_846396.2	246649
51566	D09	BA1785	hypothetical protein	Chromosome (NC_003997)	NP_844211.1	246659
49456	D10	BA4198	hypothetical protein	Chromosome (NC_003997)	NP_846434.1	246670
51844	D11	BA1818	N-acetylmuramoyl-L-alanine amidase, family 4	Chromosome (NC_003997)	NP_844239.1	246681
50194	D12	BA1778	cytochrome c-type biogenesis protein CcdA	Chromosome (NC_003997)	NP_844204.1	246693
48972	E01	BA2802	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_845151.1	240448
49095	E02	BA2792	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_845141.1	240456
51839	E03	BA5147	glycyl-tRNA synthetase	Chromosome (NC_003997)	NP_847333.1	240467
50231	E04	BA5117	lipase family protein	Chromosome (NC_003997)	NP_847303.1	240486
47833	E05	BA4186	hypothetical protein	Chromosome (NC_003997)	NP_846422.1	246617
48271	E06	BA4207	hypothetical protein	Chromosome (NC_003997)	NP_846442.1	246633
51228	E07	BA4147	hypothetical protein	Chromosome (NC_003997)	NP_846385.1	246641
48924	E08	BA4178	hypothetical protein	Chromosome (NC_003997)	NP_846416.1	246650
51583	E09	BA1830	fosmidomycin resistance protein	Chromosome (NC_003997)	NP_844249.1	246661
51654	E10	BA1819	transporter, putative	Chromosome (NC_003997)	NP_844240.1	246672
49921	E11	BA1791	DNA-binding response regulator, LuxR family	Chromosome (NC_003997)	NP_844216.1	246682
50229	E12	BA4194	2,3,4,5-tetrahydropyridine-2-carboxylate N-succinyltransferase,	Chromosome (NC_003997)	NP_846430.1	246695
51366	F01	BA5107	N-acylamino acid racemase	Chromosome (NC_003997)	NP_847293.1	240449
51572	F02	BA2786	glycine betaine/L-proline ABC transporter, ATP-binding protein	Chromosome (NC_003997)	NP_845135.1	240457
49768	F03	BA2772	hypothetical protein	Chromosome (NC_003997)	NP_845121.1	240468
47642	F04	BA1824	hypothetical protein	Chromosome (NC_003997)	NP_844245.1	246609
50785	F05	BA4195	transcriptional regulator, LysR family	Chromosome (NC_003997)	NP_846431.1	246618
48309	F06	BA4145	hypothetical protein	Chromosome (NC_003997)	NP_846383.1	246634
48516	F07	BA4177	hypothetical protein	Chromosome (NC_003997)	NP_846415.1	246642

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
51370	F08	BA1794	ABC transporter, permease protein	Chromosome (NC_003997)	NP_844219.1	246651
51582	F09	BA1783	transposase, IS605 family, OrfB	Chromosome (NC_003997)	NP_844209.1	246663
49594	F10	BA4187	polypeptide deformylase	Chromosome (NC_003997)	NP_846423.1	246673
51888	F11	BA4181	pyruvate dehydrogenase complex E3 component, dihydrolipoamide de	Chromosome (NC_003997)	NP_846418.1	246683
50303	F12	BA1797	uridylate kinase	Chromosome (NC_003997)	NP_844222.1	246697
49020	G01	BA2807	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_845155.1	240450
49243	G02	BA5144	hypothetical protein	Chromosome (NC_003997)	NP_847330.1	240458
51843	G03	BA0893	membrane protein, putative	Chromosome (NC_003997)	NP_843404.1	240469
50380	G04	BA1823	hypothetical protein	Chromosome (NC_003997)	NP_844244.1	246610
50853	G05	BA4150	membrane protein, putative	Chromosome (NC_003997)	NP_846388.1	246620
51083	G06	BA1826	3-oxoacyl-(acyl-carrier-protein) synthase III, putative	Chromosome (NC_003997)	NP_844246.1	246635
51260	G07	BA4154	cytochrome c oxidase, subunit II	Chromosome (NC_003997)	NP_846392.1	246643
49092	G08	BA1816	mutT/nudix family protein	Chromosome (NC_003997)	NP_844237.1	246652
49307	G09	BA1789	RNA polymerase sigma-70 factor, ECF subfamily	Chromosome (NC_003997)	NP_844215.1	246664
49591	G10	BA1827	cbiN domain protein	Chromosome (NC_003997)	NP_844247.1	246675
49934	G11	BA1840	fibronectin-binding protein, putative	Chromosome (NC_003997)	NP_844259.1	246684
47615	G12	BA3890	hypothetical protein	Chromosome (NC_003997)	NP_846137.1	247838
51417	H01	BA5122	glucose-1-phosphate adenylyltransferase	Chromosome (NC_003997)	NP_847308.1	240451
51571	H02	BA0926	tellurium resistance protein, putative	Chromosome (NC_003997)	NP_843432.1	240459
49865	H03	BA5142	sporulation inhibitor KapD	Chromosome (NC_003997)	NP_847328.1	240470
47717	H04	BA1839	hypothetical protein	Chromosome (NC_003997)	NP_844258.1	246611
48043	H05	BA4197	hypothetical protein	Chromosome (NC_003997)	NP_846433.1	246621
48379	H06	BA1837	hypothetical protein	Chromosome (NC_003997)	NP_844256.1	246636
48668	H07	BA4151	cytochrome c oxidase, subunit IVB	Chromosome (NC_003997)	NP_846389.1	246644
51382	H08	BA4184	pyruvate dehydrogenase complex E1 component, alpha subunit	Chromosome (NC_003997)	NP_846421.1	246653
51590	H09	BA1796	cardiolipin synthetase domain protein	Chromosome (NC_003997)	NP_844221.1	246665
49590	H10	BA1779	thioredoxin family protein	Chromosome (NC_003997)	NP_844205.1	246677
50020	H11	BA4191	potassium uptake protein, TrkA family	Chromosome (NC_003997)	NP_846427.1	246686

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