

***Bacillus anthracis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 2**

Catalog No. NR-19726

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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-19726.

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-19726 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 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 2, NR-19726."

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 2 (QMG002843)¹

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX Clone ID
50383	A02	BA2858	hypothetical protein	Chromosome (NC_003997)	NP_845200.1	239679
48658	A03	BA5211	transcriptional regulator, PadR family	Chromosome (NC_003997)	NP_847393.1	239688
48908	A04	BA0986	hypothetical protein	Chromosome (NC_003997)	NP_843487.1	239698
51212	A05	BA5222	ABC transporter, ATP-binding protein	Chromosome (NC_003997)	NP_847403.1	239707
49300	A06	BA5204	hypothetical protein	Chromosome (NC_003997)	NP_847387.1	239718
49491	A07	BA0999	hypothetical protein	Chromosome (NC_003997)	NP_843500.1	239726
50557	A08	BA3577	glycosyl transferase, group 1 family protein	Chromosome (NC_003997)	NP_845847.1	242753
50688	A09	BA1167	hypothetical protein	Chromosome (NC_003997)	NP_843646.1	242761
50799	A10	BA3057	transcriptional regulator, AraC family	Chromosome (NC_003997)	NP_845387.1	242771
48739	A11	BA3573	hypothetical protein	Chromosome (NC_003997)	NP_845843.1	242780
50977	A12	BA3056	hypothetical protein	Chromosome (NC_003997)	NP_845386.1	242789
48297	B01	BA2887	hypothetical protein	Chromosome (NC_003997)	NP_845227.1	239672
48449	B02	BA2859	hypothetical protein	Chromosome (NC_003997)	NP_845201.1	239680
50704	B03	BA2872	degV family protein	Chromosome (NC_003997)	NP_845213.1	239689
51089	B04	BA5206	peptidase, M23/M37 family	Chromosome (NC_003997)	NP_847389.1	239699
49117	B05	BA5197	hypothetical protein	Chromosome (NC_003997)	NP_847380.1	239708
51620	B06	BA1008	DNA repair exonuclease family protein	Chromosome (NC_003997)	NP_843509.1	239719
51869	B07	BA5213	hypothetical protein	Chromosome (NC_003997)	NP_847395.1	239727
47796	B08	BA1173	hypothetical protein	Chromosome (NC_003997)	NP_843652.1	242754
50692	B09	BA1169	protein export protein prsA	Chromosome (NC_003997)	NP_843648.1	242763
48406	B10	BA1168	hypothetical protein	Chromosome (NC_003997)	NP_843647.1	242772
50917	B11	BA1192	oligopeptide ABC transporter, permease protein	Chromosome (NC_003997)	NP_843668.1	242781
49192	B12	BA3538	hypothetical protein	Chromosome (NC_003997)	NP_845815.1	242790
50331	C01	BA2868	glycosyl transferase, WecB/TagA/CpsF family	Chromosome (NC_003997)	NP_845209.1	239673
50436	C02	BA2890	methlytransferase, UbiE/COQ5 family	Chromosome (NC_003997)	NP_845230.1	239681
48657	C03	BA5210	hypothetical protein	Chromosome (NC_003997)	NP_847392.1	239690
48973	C04	BA2886	hypothetical protein	Chromosome (NC_003997)	NP_845226.1	239700
51519	C05	BA2869	peptidase, putative	Chromosome (NC_003997)	NP_845210.2	239709
49299	C06	BA5190	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_847373.1	239720

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX Clone ID
49523	C07	BA5212	hypothetical protein	Chromosome (NC_003997)	NP_847394.1	239728
50592	C08	BA3069	hypothetical protein	Chromosome (NC_003997)	NP_845399.1	242755
48040	C09	BA3570	hypothetical protein	Chromosome (NC_003997)	NP_845840.1	242764
50812	C10	BA3543	transcriptional regulator, LysR family	Chromosome (NC_003997)	NP_845819.1	242773
50926	C11	BA1184	3-oxoacyl-(acyl-carrier-protein) synthase III	Chromosome (NC_003997)	NP_843661.1	242783
49235	C12	BA3512	membrane protein, putative	Chromosome (NC_003997)	NP_845792.1	242792
48334	D01	BA0988	hypothetical protein	Chromosome (NC_003997)	NP_843489.1	239674
50468	D02	BA5217	ABC transporter, ATP-binding protein	Chromosome (NC_003997)	NP_847399.1	239682
50823	D03	BA5205	lipoic acid synthetase	Chromosome (NC_003997)	NP_847388.1	239691
51115	D04	BA1004	alcohol dehydrogenase, zinc-containing	Chromosome (NC_003997)	NP_843505.1	239701
49135	D05	BA5194	hypothetical protein	Chromosome (NC_003997)	NP_847377.1	239710
51715	D06	BA5216	hypothetical protein	Chromosome (NC_003997)	NP_847398.1	239721
49596	D07	BA5203	phosphoglycerate mutase family protein, putative	Chromosome (NC_003997)	NP_847386.1	239730
47920	D08	BA1170	hypothetical protein	Chromosome (NC_003997)	NP_843649.1	242756
50693	D09	BA1171	membrane protein, putative	Chromosome (NC_003997)	NP_843650.1	242765
48593	D10	BA3064	hypothetical protein	Chromosome (NC_003997)	NP_845394.1	242774
48952	D11	BA3576	hypothetical protein	Chromosome (NC_003997)	NP_845846.1	242784
51031	D12	BA3544	quinone oxidoreductase	Chromosome (NC_003997)	NP_845820.1	242793
50341	E01	BA2865	hypothetical protein	Chromosome (NC_003997)	NP_845206.1	239675
48550	E02	BA1003	hypothetical protein	Chromosome (NC_003997)	NP_843504.1	239683
50989	E03	BA5240	L-lactate dehydrogenase	Chromosome (NC_003997)	NP_847419.1	239693
51127	E04	BA2867	hypothetical protein	Chromosome (NC_003997)	NP_845208.1	239703
51412	E05	BA1010	transposase, IS605 family	Chromosome (NC_003997)	NP_843511.1	239711
49475	E06	BA2863	hypothetical protein	Chromosome (NC_003997)	NP_845204.1	239722
49683	E07	BA5199	lipoprotein, putative	Chromosome (NC_003997)	NP_847382.1	239732
50656	E08	BA1181	UDP-glucose 4-epimerase-like protein	Chromosome (NC_003997)	NP_843658.1	242757
48065	E09	BA1172	hypothetical protein	Chromosome (NC_003997)	NP_843651.1	242766
50869	E10	BA3061	transporter, EamA family	Chromosome (NC_003997)	NP_845391.1	242775
50938	E11	BA1195	oligopeptide ABC transporter, ATP-binding protein	Chromosome (NC_003997)	NP_843671.1	242785
49241	E12	BA3060	mutT/nudix family protein	Chromosome (NC_003997)	NP_845390.1	242794
48401	F01	BA5196	hypothetical protein	Chromosome (NC_003997)	NP_847379.1	239676
50537	F02	BA5219	ABC transporter, substrate-binding protein, putative	Chromosome (NC_003997)	NP_847400.1	239684
48767	F03	BA2870	hypothetical protein	Chromosome (NC_003997)	NP_845211.1	239694
49084	F04	BA5214	nifU domain protein	Chromosome (NC_003997)	NP_847396.1	239704
51435	F05	BA0994	response regulator	Chromosome (NC_003997)	NP_843495.1	239713
51837	F06	BA2861	sensor histidine kinase SrrB, putative	Chromosome (NC_003997)	NP_845203.1	239723
49713	F07	BA5191	NAD(P)H dehydrogenase, quinone family	Chromosome (NC_003997)	NP_847374.1	239734
47945	F08	BA1182	hypothetical protein	Chromosome (NC_003997)	NP_843659.1	242758
50747	F09	BA3065	hypothetical protein	Chromosome (NC_003997)	NP_845395.1	242767
48640	F10	BA3589	hypothetical protein	Chromosome (NC_003997)	NP_845859.1	242776
48990	F11	BA3059	hypothetical protein	Chromosome (NC_003997)	NP_845389.1	242786
51043	F12	BA3574	bile acid transporter family protein	Chromosome (NC_003997)	NP_845844.1	242795

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX Clone ID
50390	G01	BA5192	phosphatase,haloacid dehalogenase family	Chromosome (NC_003997)	NP_847375.1	239677
48578	G02	BA1001	hypothetical protein	Chromosome (NC_003997)	NP_843502.1	239685
51030	G03	BA2888	inosine-uridine preferring nucleoside hydrolase family protein	Chromosome (NC_003997)	NP_845228.1	239695
51173	G04	BA5239	hypothetical protein	Chromosome (NC_003997)	NP_847418.1	239705
49217	G05	BA0985	lipoprotein, putative	Chromosome (NC_003997)	NP_843486.1	239714
49503	G06	BA5237	lipoprotein, putative	Chromosome (NC_003997)	NP_847416.1	239724
50543	G07	BA1174	hydrolase, haloacid dehalogenase-like family	Chromosome (NC_003997)	NP_843653.1	242751
50679	G08	BA3557	transcriptional regulator, LysR family	Chromosome (NC_003997)	NP_845830.1	242759
48204	G09	BA1161	hypothetical protein	Chromosome (NC_003997)	NP_843642.1	242768
50871	G10	BA3588	lipoprotein, putative	Chromosome (NC_003997)	NP_845858.1	242777
50967	G11	BA3560	glycerophosphoryl diester phosphodiesterase, putative	Chromosome (NC_003997)	NP_845833.1	242787
49252	G12	BA3058	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_845388.1	242796
48405	H01	BA0997	hypothetical protein	Chromosome (NC_003997)	NP_843498.1	239678
50561	H02	BA5220	ABC transporter, substrate-binding protein, putative	Chromosome (NC_003997)	NP_847401.1	239686
51052	H03	BA5195	fructose-1,6-bisphosphatase, class II	Chromosome (NC_003997)	NP_847378.1	239697
49079	H04	BA2873	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_845214.1	239706
51617	H05	BA2882	major facilitator family transporter	Chromosome (NC_003997)	NP_845223.1	239717
51846	H06	BA2866	MTA/SAH nucleosidase/ phosphatase, putative	Chromosome (NC_003997)	NP_845207.1	239725
47691	H07	BA3579	hypothetical protein	Chromosome (NC_003997)	NP_845849.1	242752
47962	H08	BA1175	hypothetical protein	Chromosome (NC_003997)	NP_843654.1	242760
48248	H09	BA3555	hypothetical protein	Chromosome (NC_003997)	NP_845828.1	242770
50921	H10	BA3587	glyoxalase family protein	Chromosome (NC_003997)	NP_845857.1	242779
49125	H11	BA3063	yail/yqxD family protein	Chromosome (NC_003997)	NP_845393.1	242788

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