

***Bacillus anthracis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 11**

Catalog No. NR-19735

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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-19735.

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-19735 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 11, NR-19735."

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 11 (QM002852)¹

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX Clone ID
47777	A02	BA4550	hypothetical protein	Chromosome (NC_003997)	NP_846772.1	247055
50122	A03	BA0338	hypothetical protein	Chromosome (NC_003997)	NP_842885.1	247064
50418	A04	BA2202	transposase, IS110 family, OrfA	Chromosome (NC_003997)	NP_844594.1	247074
51098	A05	BA0334	hypothetical protein	Chromosome (NC_003997)	NP_842881.1	247088
51378	A06	BA0342	amidohydrolase amhX	Chromosome (NC_003997)	NP_842889.1	247098
48796	A07	BA2189	hypothetical protein	Chromosome (NC_003997)	NP_844581.1	247109
51818	A08	BA2213	sensor histidine kinase	Chromosome (NC_003997)	NP_844605.1	247119
49544	A09	BA4099	prophage LambdaBa02, site-specific recombinase, phage integrase	Chromosome (NC_003997)	NP_846338.2	247134
50386	A10	BA4247	hypothetical protein	Chromosome (NC_003997)	NP_846482.1	246176
47902	A11	BA4244	hlyC domain protein	Chromosome (NC_003997)	NP_846479.1	246185
48389	A12	BA0520	hypothetical protein	Chromosome (NC_003997)	NP_843056.1	246195
49986	B01	BA2241	hemolysin III	Chromosome (NC_003997)	NP_844632.1	246864
47776	B02	BA4131	hypothetical protein	Chromosome (NC_003997)	NP_846370.1	247057
47901	B03	BA4123	hypothetical protein	Chromosome (NC_003997)	NP_846362.1	247065
47964	B04	BA1729	hypothetical protein	Chromosome (NC_003997)	NP_844164.1	247075
51308	B05	BA0330	polysaccharide deacetylase-like protein	Chromosome (NC_003997)	NP_842877.1	247090
48580	B06	BA1723	hypothetical protein	Chromosome (NC_003997)	NP_844158.1	247099
51664	B07	BA1731	permease, putative	Chromosome (NC_003997)	NP_844166.1	247110
48967	B08	BA1726	hypothetical protein	Chromosome (NC_003997)	NP_844161.1	247120
49480	B09	BA1732	lipoprotein, putative	Chromosome (NC_003997)	NP_844167.1	247135
50426	B10	BA4760	electron transfer flavoprotein, beta subunit	Chromosome (NC_003997)	NP_846966.1	246178
50504	B11	BA4748	chemotaxis protein MotA	Chromosome (NC_003997)	NP_846956.1	246188
50786	B12	BA4745	ABC transporter, ATP-binding protein	Chromosome (NC_003997)	NP_846954.1	246196
49991	C01	BA0377	thiamine-phosphate pyrophosphorylase	Chromosome (NC_003997)	NP_842921.1	246866
49810	C02	BA0343	hypothetical protein	Chromosome (NC_003997)	NP_842890.1	247058
50136	C03	BA2205	hypothetical protein	Chromosome (NC_003997)	NP_844597.1	247066
50417	C04	BA2188	hypothetical protein	Chromosome (NC_003997)	NP_844580.1	247076
48353	C05	BA4117	hypothetical protein	Chromosome (NC_003997)	NP_846356.1	247091
51485	C06	BA4093	hypothetical protein	Chromosome (NC_003997)	NP_846332.1	247100

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX Clone ID
48830	C07	BA4126	prophage LambdaBa02, repressor protein	Chromosome (NC_003997)	NP_846365.1	247112
49106	C08	BA2197	hypothetical protein	Chromosome (NC_003997)	NP_844589.1	247122
49610	C09	BA4552	ComE operon protein 2	Chromosome (NC_003997)	NP_846774.1	247137
47841	C10	BA1896	hypothetical protein	Chromosome (NC_003997)	NP_844307.1	246179
48142	C11	BA1869	hypothetical protein	Chromosome (NC_003997)	NP_844285.1	246189
48478	C12	BA0524	small acid-soluble spore protein, gamma-type	Chromosome (NC_003997)	NP_843060.1	246197
50018	D01	BA2220	hydrolase, haloacid dehalogenase-like family	Chromosome (NC_003997)	NP_844612.1	246868
47788	D02	BA2200	hypothetical protein	Chromosome (NC_003997)	NP_844592.1	247059
47928	D03	BA4101	hypothetical protein	Chromosome (NC_003997)	NP_846340.1	247067
50456	D04	BA2201	site-specific recombinase, resolvase family, putative	Chromosome (NC_003997)	NP_844593.1	247078
48419	D05	BA2191	hypothetical protein	Chromosome (NC_003997)	NP_844583.1	247093
48623	D06	BA0328	multidrug resistance protein, Smr family	Chromosome (NC_003997)	NP_842875.1	247101
51686	D07	BA2193	TPR domain protein	Chromosome (NC_003997)	NP_844585.1	247113
49160	D08	BA4112	prophage LambdaBa02, deoxyuridine 5'-triphosphate nucleotidohydr	Chromosome (NC_003997)	NP_846351.1	247124
47627	D09	BA1870	hypothetical protein	Chromosome (NC_003997)	NP_844286.1	246171
50450	D10	BA4750	D-alanyl-D-alanine carboxypeptidase family protein	Chromosome (NC_003997)	NP_846957.1	246180
50552	D11	BA0516	recX domain protein	Chromosome (NC_003997)	NP_843052.1	246190
48477	D12	BA0523	yfhS protein	Chromosome (NC_003997)	NP_843059.1	246199
47655	E01	BA2182	hypothetical protein	Chromosome (NC_003997)	NP_844575.1	247049
49973	E02	BA2195	hypothetical protein	Chromosome (NC_003997)	NP_844587.1	247060
50165	E03	BA0337	hypothetical protein	Chromosome (NC_003997)	NP_842884.1	247068
50529	E04	BA4120	prophage LambdaBa02, DNA replication protein DnaC, putative	Chromosome (NC_003997)	NP_846359.1	247080
51349	E05	BA0331	polysaccharide deacetylase-like protein	Chromosome (NC_003997)	NP_842878.1	247094
51549	E06	BA0332	nucleoside transporter, NupC family	Chromosome (NC_003997)	NP_842879.1	247104
48878	E07	BA4095	hypothetical protein	Chromosome (NC_003997)	NP_846334.1	247114
51835	E08	BA0333	RNA methyltransferase, TrmA family	Chromosome (NC_003997)	NP_842880.1	247125
50362	E09	BA1859	oxidoreductase	Chromosome (NC_003997)	NP_844276.1	246172
47857	E10	BA0518	hypothetical protein	Chromosome (NC_003997)	NP_843054.1	246181
48246	E11	BA1897	hypothetical protein	Chromosome (NC_003997)	NP_844308.1	246191
50886	E12	BA1898	membrane protein, putative	Chromosome (NC_003997)	NP_844309.1	246200
49722	F01	BA1725	transcriptional regulator, TetR family	Chromosome (NC_003997)	NP_844160.1	247050
47824	F02	BA4114	hypothetical protein	Chromosome (NC_003997)	NP_846353.1	247061
47923	F03	BA1724	hypothetical protein	Chromosome (NC_003997)	NP_844159.1	247069
50586	F04	BA4098	hypothetical protein	Chromosome (NC_003997)	NP_846337.1	247082
48460	F05	BA2204	hypothetical protein	Chromosome (NC_003997)	NP_844596.1	247095
48728	F06	BA4130	prophage LambdaBa02, repressor protein	Chromosome (NC_003997)	NP_846369.1	247105
51720	F07	BA2186	aspartyl-tRNA synthetase	Chromosome (NC_003997)	NP_844578.1	247115

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX Clone ID
49233	F08	BA2203	transposase, IS110 family, OrfB	Chromosome (NC_003997)	NP_844595.1	247126
47666	F09	BA1871	hypothetical protein	Chromosome (NC_003997)	NP_844287.1	246173
50462	F10	BA0534	ABC transporter, permease protein, putative	Chromosome (NC_003997)	NP_843069.1	246182
50603	F11	BA4746	acid phosphatase	Chromosome (NC_003997)	NP_846955.1	246192
48505	F12	BA4242	hypothetical protein	Chromosome (NC_003997)	NP_846477.1	246201
47737	G01	BA4102	hypothetical protein	Chromosome (NC_003997)	NP_846341.1	247053
50091	G02	BA2212	DNA-binding response regulator	Chromosome (NC_003997)	NP_844604.1	247062
50210	G03	BA4121	prophage LambdaBa02, DNA replication protein	Chromosome (NC_003997)	NP_846360.1	247070
50830	G04	BA4115	prophage LambdaBa02, RNA polymerase sigma-F factor, putative	Chromosome (NC_003997)	NP_846354.1	247084
51367	G05	BA0339	mandelate racemase/muconate lactonizing enzyme family protein	Chromosome (NC_003997)	NP_842886.1	247096
51581	G06	BA1730	DNA-binding protein	Chromosome (NC_003997)	NP_844165.1	247106
48886	G07	BA2215	hypothetical protein	Chromosome (NC_003997)	NP_844607.1	247116
49257	G08	BA4100	hypothetical protein	Chromosome (NC_003997)	NP_846339.1	247128
50370	G09	BA4753	succinate dehydrogenase, iron-sulfur protein	Chromosome (NC_003997)	NP_846959.1	246174
47876	G10	BA0519	hypothetical protein	Chromosome (NC_003997)	NP_843055.1	246183
48365	G11	BA0526	hypothetical protein	Chromosome (NC_003997)	NP_843061.1	246193
50902	G12	BA1860	hydrolase, alpha/beta fold family	Chromosome (NC_003997)	NP_844277.1	246202
49772	H01	BA4553	comE operon protein 1	Chromosome (NC_003997)	NP_846775.1	247054
47847	H02	BA4128	hypothetical protein	Chromosome (NC_003997)	NP_846367.1	247063
47938	H03	BA4118	hypothetical protein	Chromosome (NC_003997)	NP_846357.1	247071
48183	H04	BA4097	hypothetical protein	Chromosome (NC_003997)	NP_846336.1	247085
48482	H05	BA4116	hypothetical protein	Chromosome (NC_003997)	NP_846355.1	247097
48764	H06	BA2199	hypothetical protein	Chromosome (NC_003997)	NP_844591.1	247107
48911	H07	BA2207	hypothetical protein	Chromosome (NC_003997)	NP_844599.1	247118
49309	H08	BA2184	hypothetical protein	Chromosome (NC_003997)	NP_844577.1	247132
47676	H09	BA1867	hypothetical protein	Chromosome (NC_003997)	NP_844284.1	246175
50475	H10	BA1879	peptidase, M23/M37 family	Chromosome (NC_003997)	NP_844294.1	246184
50712	H11	BA1857	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_844274.1	246194

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