

***Bacillus anthracis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 17**

Catalog No. NR-19741

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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-19741.

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-19741 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 17, NR-19741."

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 17 (QMG002858)¹

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
47842	A02	BA2416	hypothetical protein	Chromosome (NC_003997)	NP_844793.1	245990
50381	A03	BA1953	hydrolase, alpha/beta fold family	Chromosome (NC_003997)	NP_844362.1	246001
48758	A04	BA0538	hypothetical protein	Chromosome (NC_003997)	NP_843073.1	246012
50883	A05	BA4318	lolS protein	Chromosome (NC_003997)	NP_846551.1	246023
51060	A06	BA4766	iron compound ABC transporter, iron compound-binding protein	Chromosome (NC_003997)	NP_846972.1	246035
49427	A07	BA2412	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_844789.1	246044
49792	A08	BA2401	hypothetical protein	Chromosome (NC_003997)	NP_844778.1	246060
49714	A09	BA5380	ATP-dependent Clp protease, proteolytic subunit ClpP	Chromosome (NC_003997)	NP_847553.1	247997
48255	A10	BA5350	hypothetical protein	Chromosome (NC_003997)	NP_847524.1	248012
50991	A11	BA5382	hypothetical protein	Chromosome (NC_003997)	NP_847555.1	248023
48464	A12	BA3504	hypothetical protein	Chromosome (NC_003997)	NP_845784.1	248034
49359	B01	BA2631	hypothetical protein	Chromosome (NC_003997)	NP_844993.1	244269
50241	B02	BA1958	oxidoreductase, short-chain dehydrogenase/reductase family	Chromosome (NC_003997)	NP_844366.1	245993
48398	B03	BA4330	hypothetical protein	Chromosome (NC_003997)	NP_846563.1	246002
48765	B04	BA2403	hypothetical protein	Chromosome (NC_003997)	NP_844780.1	246014
49088	B05	BA0537	transcriptional regulator, Fur family	Chromosome (NC_003997)	NP_843072.1	246024
49280	B06	BA4323	DNA-binding protein, CopG family	Chromosome (NC_003997)	NP_846556.1	246036
49458	B07	BA4301	hypothetical protein	Chromosome (NC_003997)	NP_846534.1	246046
51705	B08	BA4305	xanthine/uracil permease family protein	Chromosome (NC_003997)	NP_846538.1	246061
48018	B09	BA1123	hypothetical protein	Chromosome (NC_003997)	NP_843609.1	247998
50739	B10	BA5379	hypothetical protein	Chromosome (NC_003997)	NP_847552.1	248013
48323	B11	BA3507	hypothetical protein	Chromosome (NC_003997)	NP_845787.1	248024
48463	B12	BA3502	hypothetical protein	Chromosome (NC_003997)	NP_845782.1	248036
49448	C01	BA4973	molybdopterin-guanine dinucleotide biosynthesis protein MobB	Chromosome (NC_003997)	NP_847168.1	244271
47985	C02	BA2408	hypothetical protein	Chromosome (NC_003997)	NP_844785.1	245994
48447	C03	BA1947	hypothetical protein	Chromosome (NC_003997)	NP_844356.1	246004

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
50720	C04	BA2399	metallo-beta-lactamase family protein	Chromosome (NC_003997)	NP_844776.1	246015
49132	C05	BA4295	anti-sigma F factor antagonist	Chromosome (NC_003997)	NP_846528.1	246026
51133	C06	BA4336	biotin synthetase	Chromosome (NC_003997)	NP_846569.1	246037
49510	C07	BA2398	lipoprotein, putative	Chromosome (NC_003997)	NP_844775.1	246048
49872	C08	BA4291	stage V sporulation protein AA	Chromosome (NC_003997)	NP_846524.1	246062
48060	C09	BA3505	hypothetical protein	Chromosome (NC_003997)	NP_845785.1	248000
48294	C10	BA5321	hypothetical protein	Chromosome (NC_003997)	NP_847497.1	248014
48329	C11	BA5381	phosphocarrier protein HPr	Chromosome (NC_003997)	NP_847554.1	248026
51217	C12	BA5370	gapA transcriptional regulator CggR	Chromosome (NC_003997)	NP_847543.1	248037
50104	D01	BA1955	DNA-binding response regulator	Chromosome (NC_003997)	NP_844363.1	245985
50273	D02	BA2395	zinc transporter family protein	Chromosome (NC_003997)	NP_844772.1	245995
48581	D03	BA1940	carboxymuconolactone decarboxylase family protein	Chromosome (NC_003997)	NP_844349.1	246006
48846	D04	BA4335	hypothetical protein	Chromosome (NC_003997)	NP_846568.1	246016
50911	D05	BA2411	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_844788.1	246027
49317	D06	BA4327	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_846560.1	246038
49568	D07	BA2419	hypothetical protein	Chromosome (NC_003997)	NP_844796.1	246050
49886	D08	BA2397	hypothetical protein	Chromosome (NC_003997)	NP_844774.1	246065
48122	D09	BA5352	hypothetical protein	Chromosome (NC_003997)	NP_847526.1	248002
50777	D10	BA5384	hypothetical protein	Chromosome (NC_003997)	NP_847557.1	248015
51009	D11	BA5387	thioredoxin reductase	Chromosome (NC_003997)	NP_847560.1	248027
48493	D12	BA3503	hypothetical protein	Chromosome (NC_003997)	NP_845783.1	248038
47647	E01	BA4292	hypothetical protein	Chromosome (NC_003997)	NP_846525.1	245986
50293	E02	BA4338	bioH protein, putative	Chromosome (NC_003997)	NP_846571.1	245997
48616	E03	BA0540	hypothetical protein	Chromosome (NC_003997)	NP_843075.1	246008
48986	E04	BA2404	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_844781.1	246018
49142	E05	BA1941	transcriptional regulator, MarR family	Chromosome (NC_003997)	NP_844350.1	246028
51132	E06	BA4324	hydrolase, alpha/beta fold family, putative	Chromosome (NC_003997)	NP_846557.1	246039
49579	E07	BA2420	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_844797.1	246052
51872	E08	BA2418	protoporphyrinogen oxidase	Chromosome (NC_003997)	NP_844795.1	246066
48134	E09	BA5373	hypothetical protein	Chromosome (NC_003997)	NP_847546.1	248006
48298	E10	BA3023	hypothetical protein	Chromosome (NC_003997)	NP_845354.1	248016
48373	E11	BA3021	hypothetical protein	Chromosome (NC_003997)	NP_845352.1	248028
48502	E12	BA3039	hypothetical protein	Chromosome (NC_003997)	NP_845370.1	248039
50196	F01	BA2410	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_844787.1	245987
48113	F02	BA2422	cold shock protein CspA	Chromosome (NC_003997)	NP_844799.1	245998
50587	F03	BA4308	purine nucleoside phosphorylase	Chromosome (NC_003997)	NP_846541.1	246009
50780	F04	BA2413	membrane protein, putative	Chromosome (NC_003997)	NP_844790.1	246019
49154	F05	BA1948	lipoprotein, putative	Chromosome (NC_003997)	NP_844357.1	246030
49324	F06	BA2402	hypothetical protein	Chromosome (NC_003997)	NP_844779.1	246040
49619	F07	BA2414	DNA-binding protein	Chromosome (NC_003997)	NP_844791.1	246054
49949	F08	BA4332	riboflavin synthase, alpha subunit	Chromosome (NC_003997)	NP_846565.1	246067
50538	F09	BA5386	hypothetical protein	Chromosome (NC_003997)	NP_847559.1	248007
48314	F10	BA5371	glutaredoxin family protein	Chromosome (NC_003997)	NP_847544.1	248018
51025	F11	BA5325	hypothetical protein	Chromosome (NC_003997)	NP_847501.1	248029
51248	F12	BA3508	hypothetical protein	Chromosome (NC_003997)	NP_845788.1	248041

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
47757	G01	BA4317	hypothetical protein	Chromosome (NC_003997)	NP_846550.1	245988
50357	G02	BA4294	RNA polymerase sigma-F factor	Chromosome (NC_003997)	NP_846527.1	245999
48748	G03	BA4296	anti-sigma F factor antagonist	Chromosome (NC_003997)	NP_846529.1	246010
49023	G04	BA4290	stage V sporulation protein AB	Chromosome (NC_003997)	NP_846523.1	246020
50985	G05	BA2415	hypothetical protein	Chromosome (NC_003997)	NP_844792.1	246031
51138	G06	BA1949	microcin immunity protein MccF	Chromosome (NC_003997)	NP_844358.1	246041
49732	G07	BA1957	hypothetical protein	Chromosome (NC_003997)	NP_844365.1	246056
49968	G08	BA2409	chloramphenicol acetyltransferase	Chromosome (NC_003997)	NP_844786.1	246069
48151	G09	BA1082	hypothetical protein	Chromosome (NC_003997)	NP_843575.1	248008
48313	G10	BA5351	hypothetical protein	Chromosome (NC_003997)	NP_847525.1	248020
48402	G11	BA5354	hypothetical protein	Chromosome (NC_003997)	NP_847528.1	248032
48606	G12	BA3015	hypothetical protein	Chromosome (NC_003997)	NP_845346.1	248042
50214	H01	BA0541	conserved hypothetical protein TIGR01033	Chromosome (NC_003997)	NP_843076.1	245989
48310	H02	BA4329	hypothetical protein	Chromosome (NC_003997)	NP_846562.1	246000
50694	H03	BA1950	D-alanyl-D-alanine carboxypeptidase family protein	Chromosome (NC_003997)	NP_844359.1	246011
50788	H04	BA0546	hypothetical protein	Chromosome (NC_003997)	NP_843081.1	246021
49223	H05	BA4334	riboflavin synthase, beta subunit	Chromosome (NC_003997)	NP_846567.1	246034
49351	H06	BA0542	mutT/nudix family protein	Chromosome (NC_003997)	NP_843077.1	246042
49779	H07	BA1951	hypothetical protein	Chromosome (NC_003997)	NP_844360.1	246058
47954	H08	BA5348	hypothetical protein	Chromosome (NC_003997)	NP_847522.1	247996
48168	H09	BA5349	hypothetical protein	Chromosome (NC_003997)	NP_847523.1	248010
48306	H10	BA3012	hypothetical protein	Chromosome (NC_003997)	NP_845344.1	248022
51156	H11	BA5369	glyceraldehyde 3-phosphate dehydrogenase	Chromosome (NC_003997)	NP_847542.1	248033

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