

***Bacillus anthracis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 13**

**Catalog No. NR-19737**

This reagent is the tangible property of the U.S. Government.

**For research use only. Not for human use.**

**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-19737.

**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-19737 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 13, NR-19737."

**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](http://www.cdc.gov/biosafety/publications/bmb15/index.htm).

**Disclaimers:**

You are authorized to use this product for research use only. It is not intended for human use.

Use of this product is subject to the terms and conditions of the BEI Resources Material Transfer Agreement (MTA). The MTA is available on our Web site at [www.beiresources.org](http://www.beiresources.org).

While BEI Resources uses reasonable efforts to include accurate and up-to-date information on this product sheet, neither ATCC® nor the U.S. Government makes any warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. Neither ATCC® nor the U.S. Government warrants that such information has been confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling, use and disposal. ATCC® and the U.S. Government are not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to ensure authenticity and reliability of materials on deposit, the U.S. Government, ATCC®, their suppliers and contributors to BEI

Resources are not liable for damages arising from the misidentification or misrepresentation of products.

**Use Restrictions:**

**This material is distributed for internal research, non-commercial purposes only.** This material, its product or its derivatives may not be distributed to third parties. Except as performed under a U.S. Government contract, individuals contemplating commercial use of the material, its products or its derivatives must contact the contributor to determine if a license is required. U.S. Government contractors may need a license before first commercial sale.

**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.

ATCC® is a trademark of the American Type Culture Collection



**Table 1: *Bacillus anthracis*, Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 13 (QMG002854)<sup>1</sup>**

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
47707	A02	BA5018	hypothetical protein	Chromosome (NC_003997)	NP_847212.1	244367
47852	A03	BA5456	hypothetical protein	Chromosome (NC_003997)	NP_847624.1	244377
50881	A04	BA2672	transporter, EamA family	Chromosome (NC_003997)	NP_845031.1	244386
51179	A05	BA2678	hypothetical protein	Chromosome (NC_003997)	NP_845037.1	244398
48553	A06	BA2105	hypothetical protein	Chromosome (NC_003997)	NP_844504.1	244409
48892	A07	BA5022	ATP synthase protein I, putative	Chromosome (NC_003997)	NP_847215.1	244423
49429	A08	BA2685	mutT/nudix family protein	Chromosome (NC_003997)	NP_845042.1	244435
51823	A09	BA2673	chitinase	Chromosome (NC_003997)	NP_845032.1	244446
47968	A10	BA4877	proA domain protein	Chromosome (NC_003997)	NP_847078.1	245542
48195	A11	BA2539	hypothetical protein	Chromosome (NC_003997)	NP_844907.1	245556
48704	A12	BA0673	hypothetical protein	Chromosome (NC_003997)	NP_843200.1	245565
49588	B01	BA1404	hypothetical protein	Chromosome (NC_003997)	NP_843861.1	241967
47799	B02	BA2674	hypothetical protein	Chromosome (NC_003997)	NP_845033.1	244369
50630	B03	BA5003	ABC transporter, ATP-binding protein, putative	Chromosome (NC_003997)	NP_847197.1	244378
48153	B04	BA2104	hypothetical protein	Chromosome (NC_003997)	NP_844503.1	244387
48394	B05	BA2677	hypothetical protein	Chromosome (NC_003997)	NP_845036.1	244399
48617	B06	BA0791	PTS system, cellobiose-specific IIA component	Chromosome (NC_003997)	NP_843310.1	244411
49121	B07	BA0782	hypothetical protein	Chromosome (NC_003997)	NP_843303.1	244425
51735	B08	BA2664	permease, putative	Chromosome (NC_003997)	NP_845024.1	244436
49884	B09	BA2106	hypothetical protein	Chromosome (NC_003997)	NP_844505.1	244449
50470	B10	BA0647	hypothetical protein	Chromosome (NC_003997)	NP_843178.1	245543
50848	B11	BA0657	oligopeptide ABC transporter, permease protein	Chromosome (NC_003997)	NP_843187.1	245557
51038	B12	BA0658	oligopeptide ABC transporter, permease protein	Chromosome (NC_003997)	NP_843188.1	245566
49662	C01	BA3715	thiJ/pfpl family protein	Chromosome (NC_003997)	NP_845978.1	241969
50377	C02	BA0781	membrane protein, putative	Chromosome (NC_003997)	NP_843302.1	244370
47970	C03	BA5016	hypothetical protein	Chromosome (NC_003997)	NP_847210.1	244379
50900	C04	BA5027	hypothetical protein	Chromosome (NC_003997)	NP_847220.1	244388
51203	C05	BA4998	membrane protein, putative	Chromosome (NC_003997)	NP_847192.1	244400
48621	C06	BA2103	hypothetical protein	Chromosome (NC_003997)	NP_844502.1	244413
51555	C07	BA0787	major facilitator family transporter	Chromosome (NC_003997)	NP_843307.1	244426
49436	C08	BA5010	bacterial transferase family protein	Chromosome (NC_003997)	NP_847204.1	244437

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
50084	C09	BA5004	membrane protein, putative	Chromosome (NC_003997)	NP_847198.1	244453
48088	C10	BA2525	hypothetical protein	Chromosome (NC_003997)	NP_844894.1	245546
48338	C11	BA2540	hypothetical protein	Chromosome (NC_003997)	NP_844908.1	245558
48715	C12	BA4878	hypothetical protein	Chromosome (NC_003997)	NP_847079.1	245567
49703	D01	BA1423	3-isopropylmalate dehydratase, small subunit	Chromosome (NC_003997)	NP_843880.1	241971
47814	D02	BA2679	hypothetical protein	Chromosome (NC_003997)	NP_845038.1	244371
50713	D03	BA2668	glycerophosphoryl diester phosphodiesterase, putative	Chromosome (NC_003997)	NP_845028.1	244380
48171	D04	BA0805	cotJA protein	Chromosome (NC_003997)	NP_843323.1	244389
51213	D05	BA5461	cytochrome d ubiquinol oxidase, subunit II, putative	Chromosome (NC_003997)	NP_847629.1	244402
51413	D06	BA2684	DNA polymerase III, beta subunit	Chromosome (NC_003997)	NP_845041.1	244414
49144	D07	BA2683	mutT/nudix family protein	Chromosome (NC_003997)	NP_845040.1	244427
51746	D08	BA0793	PTS system, cellobiose-specific IIC component	Chromosome (NC_003997)	NP_843312.1	244438
50139	D09	BA2665	hypothetical protein	Chromosome (NC_003997)	NP_845025.1	244455
50553	D10	BA0683	undecaprenol kinase family protein	Chromosome (NC_003997)	NP_843210.1	245547
50910	D11	BA0669	ribose ABC transporter, ribose-binding protein	Chromosome (NC_003997)	NP_843197.1	245560
51046	D12	BA0664	ribose operon repressor	Chromosome (NC_003997)	NP_843192.1	245568
49720	E01	BA1427	imidazoleglycerol-phosphate dehydratase	Chromosome (NC_003997)	NP_843884.1	241973
50442	E02	BA0807	hypothetical protein	Chromosome (NC_003997)	NP_843325.1	244372
47969	E03	BA5015	hypothetical protein	Chromosome (NC_003997)	NP_847209.1	244381
48209	E04	BA5023	hypothetical protein	Chromosome (NC_003997)	NP_847216.1	244391
48444	E05	BA0794	hypothetical protein	Chromosome (NC_003997)	NP_843313.1	244403
51440	E06	BA5012	glycosyl transferase, group 1 family protein	Chromosome (NC_003997)	NP_847206.1	244416
51580	E07	BA2680	oxalate:formate antiporter, putative	Chromosome (NC_003997)	NP_845039.1	244428
49498	E08	BA2670	hypothetical protein	Chromosome (NC_003997)	NP_845030.1	244439
47741	E09	BA0655	hypothetical protein	Chromosome (NC_003997)	NP_843185.1	245536
48087	E10	BA2523	DNA-binding protein	Chromosome (NC_003997)	NP_844892.1	245548
48562	E11	BA4871	DNA-binding protein	Chromosome (NC_003997)	NP_847072.1	245561
48718	E12	BA0676	hypothetical protein	Chromosome (NC_003997)	NP_843203.1	245569
47629	F01	BA2660	hypothetical protein	Chromosome (NC_003997)	NP_845021.1	244363
47851	F02	BA4999	hypothetical protein	Chromosome (NC_003997)	NP_847193.1	244373
48076	F03	BA5020	hypothetical protein	Chromosome (NC_003997)	NP_847214.1	244383
48226	F04	BA0778	hypothetical protein	Chromosome (NC_003997)	NP_843300.1	244393
48548	F05	BA0792	PTS system, cellobiose-specific IIB component	Chromosome (NC_003997)	NP_843311.1	244405
48699	F06	BA5458	transposase, IS605 family, OrfA	Chromosome (NC_003997)	NP_847626.1	244419
49157	F07	BA2110	hypothetical protein	Chromosome (NC_003997)	NP_844508.1	244429
51756	F08	BA0802	branched-chain amino acid transport system II carrier protein	Chromosome (NC_003997)	NP_843320.1	244440
50287	F09	BA2557	hypothetical protein	Chromosome (NC_003997)	NP_844925.1	245537
50652	F10	BA4902	transcriptional regulator, LysR family	Chromosome (NC_003997)	NP_847101.1	245551

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX CloneID
50909	F11	BA0659	oligopeptide ABC transporter, ATP-binding protein	Chromosome (NC_003997)	NP_843189.1	245562
51109	F12	BA4889	hypothetical protein	Chromosome (NC_003997)	NP_847089.1	245570
50262	G01	BA0800	ABC transporter, permease protein, putative	Chromosome (NC_003997)	NP_843319.1	244364
50522	G02	BA2667	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_845027.1	244374
50855	G03	BA5024	transporter, EamA family	Chromosome (NC_003997)	NP_847217.1	244384
51014	G04	BA5001	conserved hypothetical protein TIGR01212	Chromosome (NC_003997)	NP_847195.1	244394
48557	G05	BA2659	hypothetical protein	Chromosome (NC_003997)	NP_845020.1	244407
48779	G06	BA0806	hypothetical protein	Chromosome (NC_003997)	NP_843324.1	244421
51597	G07	BA5028	sensor histidine kinase, putative	Chromosome (NC_003997)	NP_847221.1	244430
49648	G08	BA0803	cotJC protein	Chromosome (NC_003997)	NP_843321.1	244443
47831	G09	BA2544	hypothetical protein	Chromosome (NC_003997)	NP_844912.1	245538
50674	G10	BA0677	phospholipase C	Chromosome (NC_003997)	NP_843204.1	245553
48652	G11	BA2554	hypothetical protein	Chromosome (NC_003997)	NP_844922.1	245563
48929	G12	BA0666	ribose ABC transporter protein	Chromosome (NC_003997)	NP_843194.1	245573
47668	H01	BA2666	hypothetical protein	Chromosome (NC_003997)	NP_845026.1	244365
47853	H02	BA5459	hypothetical protein	Chromosome (NC_003997)	NP_847627.1	244375
48121	H03	BA5026	hypothetical protein	Chromosome (NC_003997)	NP_847219.1	244385
48386	H04	BA5000	hypothetical protein	Chromosome (NC_003997)	NP_847194.1	244397
51320	H05	BA0789	hypothetical protein	Chromosome (NC_003997)	NP_843308.1	244408
51550	H06	BA0783	transcription antiterminator, LytR family	Chromosome (NC_003997)	NP_843304.1	244422
49417	H07	BA5014	molybdenum cofactor biosynthesis protein B, putative	Chromosome (NC_003997)	NP_847208.1	244433
49666	H08	BA5002	hypothetical protein	Chromosome (NC_003997)	NP_847196.1	244445
50477	H09	BA2551	enoyl-CoA hydratase/isomerase family protein	Chromosome (NC_003997)	NP_844919.1	245541
48154	H10	BA2549	hypothetical protein	Chromosome (NC_003997)	NP_844917.1	245554
50995	H11	BA0648	hypothetical protein	Chromosome (NC_003997)	NP_843179.1	245564

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