

***Bacillus anthracis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 14**

Catalog No. NR-19738

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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-19738.

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-19738 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 14, NR-19738."

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 14 (QMG002855)¹

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX Clone ID
48880	A02	BA5393	membrane protein, putative	Chromosome (NC_003997)	NP_847566.1	242918
48747	A03	BA3525	hypothetical protein	Chromosome (NC_003997)	NP_845803.1	242904
50696	A04	BA3536	phenazine biosynthesis protein, PhzF family	Chromosome (NC_003997)	NP_845813.1	242893
48304	A05	BA1099	hypothetical protein	Chromosome (NC_003997)	NP_843592.1	242884
48173	A06	BA1144	spore germination protein GerPF	Chromosome (NC_003997)	NP_843628.1	242874
49992	A07	BA1129	S-layer protein, putative	Chromosome (NC_003997)	NP_843614.1	242863
47729	A08	BA3496	hypothetical protein	Chromosome (NC_003997)	NP_845776.1	242848
49952	A09	BA0670	transaldolase, putative	Chromosome (NC_003997)	NP_843198.1	245612
51751	A10	BA0680	oxidoreductase, FAD-binding	Chromosome (NC_003997)	NP_843207.1	245601
49204	A11	BA2558	hypothetical protein	Chromosome (NC_003997)	NP_844926.1	245590
51439	A12	BA4900	aminotransferase, class V	Chromosome (NC_003997)	NP_847099.1	245581
49173	B01	BA5394	hypothetical protein	Chromosome (NC_003997)	NP_847567.1	242928
48877	B02	BA3523	hypothetical protein	Chromosome (NC_003997)	NP_845801.1	242916
51067	B03	BA1111	HD domain protein	Chromosome (NC_003997)	NP_843598.1	242903
48480	B04	BA1143	hypothetical protein	Chromosome (NC_003997)	NP_843627.1	242892
50384	B05	BA3535	membrane protein, putative	Chromosome (NC_003997)	NP_845812.1	242883
48125	B06	BA1148	spore germination protein GerPB	Chromosome (NC_003997)	NP_843632.1	242872
47877	B07	BA1103	hypothetical protein	Chromosome (NC_003997)	NP_843594.1	242862
49529	B08	BA3530	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_845807.1	242847
49942	B09	BA0642	amino acid ABC transporter, permease protein	Chromosome (NC_003997)	NP_843173.1	245611
49473	B10	BA2555	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_844923.1	245600
51486	B11	BA0674	multidrug resistance protein, putative	Chromosome (NC_003997)	NP_843201.1	245589
49042	B12	BA0654	universal stress protein family	Chromosome (NC_003997)	NP_843184.1	245580
49085	C01	BA5398	lipoprotein, putative	Chromosome (NC_003997)	NP_847571.1	242926
48810	C02	BA1155	hypothetical protein	Chromosome (NC_003997)	NP_843637.1	242912
48712	C03	BA3494	hypothetical protein	Chromosome (NC_003997)	NP_845774.1	242902
50687	C04	BA1120	CAAX amino terminal protease family protein	Chromosome (NC_003997)	NP_843606.1	242891
50367	C05	BA3493	ABC transporter, ATP-binding protein	Chromosome (NC_003997)	NP_845773.1	242881
48111	C06	BA1135	cold shock protein CspA	Chromosome (NC_003997)	NP_843619.1	242870
47865	C07	BA3521	hypothetical protein	Chromosome (NC_003997)	NP_845799.1	242858

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX Clone ID
47669	C08	BA3499	hypothetical protein	Chromosome (NC_003997)	NP_845779.1	242846
51941	C09	BA0653	sulfate permease family protein	Chromosome (NC_003997)	NP_843183.1	245610
49462	C10	BA0649	dihydrofolate reductase family protein	Chromosome (NC_003997)	NP_843180.1	245598
49099	C11	BA4887	mutT/nudix family protein	Chromosome (NC_003997)	NP_847087.1	245588
51425	C12	BA4873	alanine dehydrogenase	Chromosome (NC_003997)	NP_847074.1	245579
49066	D01	BA1090	lipoprotein, putative	Chromosome (NC_003997)	NP_843583.1	242924
51221	D02	BA3498	hypothetical protein	Chromosome (NC_003997)	NP_845778.1	242911
50924	D03	BA5392	HPr(Ser) kinase/phosphatase	Chromosome (NC_003997)	NP_847565.1	242901
48468	D04	BA1153	hypothetical protein	Chromosome (NC_003997)	NP_843635.1	242890
48220	D05	BA3520	hypothetical protein	Chromosome (NC_003997)	NP_845798.1	242880
50118	D06	BA3510	cyclic nucleotide-binding domain protein	Chromosome (NC_003997)	NP_845790.1	242869
47768	D07	BA1100	hypothetical protein	Chromosome (NC_003997)	NP_843593.1	242856
50259	D08	BA2526	D-alanyl-D-alanine carboxypeptidase family protein	Chromosome (NC_003997)	NP_844895.1	245624
51909	D09	BA0643	amino acid ABC transporter, permease protein	Chromosome (NC_003997)	NP_843174.1	245608
49394	D10	BA2534	acetyltransferase, GNAT family	Chromosome (NC_003997)	NP_844902.1	245596
49089	D11	BA0645	HIT family protein	Chromosome (NC_003997)	NP_843176.1	245586
48969	D12	BA2527	hypothetical protein	Chromosome (NC_003997)	NP_844896.1	245578
49049	E01	BA3511	membrane protein, putative	Chromosome (NC_003997)	NP_845791.1	242922
48752	E02	BA3509	hypothetical protein	Chromosome (NC_003997)	NP_845789.1	242910
48688	E03	BA1096	hypothetical protein	Chromosome (NC_003997)	NP_843589.1	242900
50563	E04	BA5391	prolipoprotein diacylglyceryl transferase	Chromosome (NC_003997)	NP_847564.1	242889
50333	E05	BA3514	metallo-beta-lactamase family protein	Chromosome (NC_003997)	NP_845794.1	242879
48051	E06	BA1146	spore germination protein GerPD	Chromosome (NC_003997)	NP_843630.1	242868
49702	E07	BA1138	competence transcription factor, putative	Chromosome (NC_003997)	NP_843622.1	242855
50130	E08	BA4876	membrane protein, putative	Chromosome (NC_003997)	NP_847077.1	245622
49918	E09	BA4886	hypothetical protein	Chromosome (NC_003997)	NP_847086.1	245607
49377	E10	BA4890	thiol peroxidase	Chromosome (NC_003997)	NP_847090.1	245594
51457	E11	BA2560	sensor histidine kinase	Chromosome (NC_003997)	NP_844928.1	245585
51261	E12	BA0675	alcohol dehydrogenase, zinc-containing	Chromosome (NC_003997)	NP_843202.1	245577
51534	F01	BA1154	ornithine aminotransferase	Chromosome (NC_003997)	NP_843636.1	242921
51181	F02	BA3533	iron compound ABC transporter, permease protein	Chromosome (NC_003997)	NP_845810.1	242909
48662	F03	BA1121	hypothetical protein	Chromosome (NC_003997)	NP_843607.1	242898
48450	F04	BA3522	hypothetical protein	Chromosome (NC_003997)	NP_845800.1	242888
48203	F05	BA1149	spore germination protein GerPA	Chromosome (NC_003997)	NP_843633.1	242878
50037	F06	BA1127	S-layer protein, putative	Chromosome (NC_003997)	NP_843613.1	242867
47742	F07	BA1092	hypothetical protein	Chromosome (NC_003997)	NP_843585.1	242852
50059	F08	BA0651	DNA-binding response regulator	Chromosome (NC_003997)	NP_843182.1	245620
49807	F09	BA2530	transcriptional regulator, TetR family	Chromosome (NC_003997)	NP_844898.1	245605
51557	F10	BA2537	hypothetical protein	Chromosome (NC_003997)	NP_844905.1	245593

Clone	Well Position	Locus ID	Description	Source	Accession Number	FLEX Clone ID
49082	F11	BA4895	hypothetical protein	Chromosome (NC_003997)	NP_847095.1	245584
48950	F12	BA2536	spore coat protein, putative	Chromosome (NC_003997)	NP_844904.1	245576
48898	G01	BA1145	spore germination protein GerPE	Chromosome (NC_003997)	NP_843629.1	242920
51099	G02	BA1089	lipoate-protein ligase A, putative	Chromosome (NC_003997)	NP_843582.1	242907
50834	G03	BA1114	transcriptional regulator, AraC family	Chromosome (NC_003997)	NP_843601.1	242897
50443	G04	BA3531	iron compound ABC transporter, iron compound-binding protein, putative	Chromosome (NC_003997)	NP_845808.1	242887
50322	G05	BA3492	ABC transporter, efflux permease protein	Chromosome (NC_003997)	NP_845772.1	242877
50010	G06	BA3524	hypothetical protein	Chromosome (NC_003997)	NP_845802.1	242865
47728	G07	BA1097	hypothetical protein	Chromosome (NC_003997)	NP_843590.1	242850
49982	G08	BA0641	amino acid ABC transporter, permease protein	Chromosome (NC_003997)	NP_843172.1	245616
49620	G09	BA2556	hypothetical protein	Chromosome (NC_003997)	NP_844924.1	245604
49212	G10	BA4875	universal stress protein family	Chromosome (NC_003997)	NP_847076.1	245592
51448	G11	BA0681	membrane protein, putative	Chromosome (NC_003997)	NP_843208.1	245583
48940	G12	BA4891	hypothetical protein	Chromosome (NC_003997)	NP_847091.1	245575
51460	H01	BA5397	transposase, IS605 family	Chromosome (NC_003997)	NP_847570.1	242919
51066	H02	BA1086	sugar-binding transcriptional regulator, LacI family	Chromosome (NC_003997)	NP_843579.1	242905
48522	H03	BA1122	hypothetical protein	Chromosome (NC_003997)	NP_843608.1	242894
48392	H04	BA1109	hypothetical protein	Chromosome (NC_003997)	NP_843597.1	242886
48191	H05	BA1117	hypothetical protein	Chromosome (NC_003997)	NP_843603.1	242876
47883	H06	BA1087	hypothetical protein	Chromosome (NC_003997)	NP_843580.1	242864
49586	H07	BA1156	hypothetical protein	Chromosome (NC_003997)	NP_843638.1	242849
49975	H08	BA2543	transcriptional regulator, TetR family	Chromosome (NC_003997)	NP_844911.1	245614
51788	H09	BA2548	acetyl-CoA carboxylase, biotin carboxylase, putative	Chromosome (NC_003997)	NP_844916.1	245603
51545	H10	BA4888	acetate kinase	Chromosome (NC_003997)	NP_847088.1	245591
49061	H11	BA4872	hypothetical protein	Chromosome (NC_003997)	NP_847073.1	245582

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