

***Vibrio cholerae* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 17**

Catalog No. NR-19695

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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:

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 does not confirm or validate individual mutants provided by the contributor.

The *Vibrio cholerae* (*V. cholerae*) Gateway® clone set consists of 46 plates which contain 3813 sequence validated clones from *V. cholerae*, strain EI Tor N16961 cloned in *Escherichia coli* (*E. coli*) DH10B-T1 cells. Each open reading frame was constructed in vector [pDONR™221](#) with a native start codon and 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 an *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.

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-19695 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 or agar containing 50 µg/mL kanamycin

Incubation:

Temperature: *E. coli*, strain DH10B-T1 clones should be grown at 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 1 day.

Citation:

Acknowledgment for publications should read “The following reagent was obtained through BEI Resources, NIAID, NIH: *Vibrio cholerae* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 17, NR-19695.”

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. Heidelberg, J. F., et al. "DNA Sequence of both Chromosomes of the Cholera Pathogen *Vibrio cholerae*." *Nature* 406 (2000): 477-483. PubMed: 10952301

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Table 1: *Vibrio cholerae* Gateway® Clones, Plate 17

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
198716	A02	282	VC0443	ksgA	dimethyladenosine transferase	NP_230097.1
198742	A03	128	VC0897		conserved hypothetical protein	NP_230544.1
198758	A04	N/A	VCA0538		cytochrome b561, putative	N/A
198769	A05	431	VC2092	gltA	citrate synthase	NP_231724.1
198781	A06	N/A	VCA0540		formate transporter 1, putative	N/A
198792	A07	257	VC2094		hypothetical protein	NP_231726.1
198530	A08	N/A	VCA0207	nadE	NH(3)-dependent NAD(+) synthetase	N/A
198549	A09	95	VC2149		hypothetical protein	NP_231780.1
198565	A10	118	VC2147		hypothetical protein	NP_231778.1
198586	A11	N/A	VCA0218		thermolabile hemolysin	N/A
198597	A12	214	VC0494		conserved hypothetical protein	NP_230148.1
200552	B01	236	VC1290		DNA polymerase III, epsilon subunit, putative	NP_230935.1
198718	B02	284	VC0447		DnaJ-related protein	NP_230101.1
198745	B03	N/A	VCA0527		conserved hypothetical protein	N/A
198759	B04	372	VC0917	wecB	UDP-N-acetylglucosamine 2-epimerase	NP_230564.1
198771	B05	431	VC0445	surA	survival protein SurA	NP_230099.1
198794	B07	258	VC0904		conserved hypothetical protein	NP_230551.1
198532	B08	N/A	VCA0222		lipase activator protein, putative	N/A
198551	B09	N/A	VCA0236		hypothetical protein	N/A
198567	B10	131	VC2150		conserved hypothetical protein	NP_231781.1
198587	B11	N/A	VCA0208		conserved hypothetical protein	N/A
198600	B12	N/A	VCA0210		response regulator, putative	N/A
200553	C01	111	VC1289		methyl-accepting chemotaxis protein	NP_230934.1
198720	C02	281	VC0902		conserved hypothetical protein	NP_230549.2
198746	C03	143	VC0457		hypothetical protein	NP_230111.1
198761	C04	382	VC0920		exopolysaccharide biosynthesis protein EpsF, putative	NP_230567.1
198772	C05	200	VC0456		HAM1 protein	NP_230110.1
198784	C06	236	VC2088	sdhB	succinate dehydrogenase, iron-sulfur protein	NP_231720.1
198796	C07	260	VC2083	znuB	zinc ABC transporter, permease protein	NP_231715.1
198534	C08	291	VC0959		hemolysin, putative	NP_230606.1
198554	C09	N/A	VCA0221	hlyC	lactonizing lipase	N/A
198572	C10	377	VC2152	dapE	succinyl-diaminopimelate desuccinylase	NP_231783.1
198589	C11	N/A	VCA0237		hypothetical protein	N/A
198602	C12	N/A	VCA0213		conserved hypothetical protein	N/A
200554	D01	237	VC1612		fimbrial biogenesis and twitching motility protein, putative	NP_231252.1
198751	D03	345	VC0462	pilT-1	twitching motility protein PilT	NP_230116.1
198762	D04	185	VC0459		conserved hypothetical protein	NP_230113.1
198773	D05	457	VC0899		conserved hypothetical protein	NP_230546.1
198785	D06	515	VC0894	thiI	thiamin biosynthesis protein ThiI	NP_230541.1
198797	D07	588	VC2089	sdhA	succinate dehydrogenase, flavoprotein subunit	NP_231721.1
198539	D08	N/A	VCA0209		hypothetical protein	N/A
198557	D09	105	VC0952		conserved hypothetical protein	NP_230599.1
198574	D10	377	VC2143	flaD	flagellin FlaD	NP_231774.1
198591	D11	199	VC0496		hypothetical protein	NP_230150.1

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
198604	D12	N/A	VCA0220	hylB	hemolysin secretion protein HylB	N/A
200555	E01	637	VC1291		conserved hypothetical protein	NP_230936.1
198725	E02	290	VC2084	sucD	succinyl-CoA synthase, alpha subunit	NP_231716.1
198754	E03	N/A	VCA0543		conserved hypothetical protein	N/A
198776	E05	225	VC0906		ABC transporter, permease protein	NP_230553.1
198786	E06	239	VC0453		conserved hypothetical protein	NP_230107.1
198799	E07	N/A	VCA0529	kup	potassium uptake protein, Kup system	N/A
198540	E08	N/A	VCA0241		hexulose-6-phosphate isomerase SgbU, putative	N/A
198558	E09	N/A	VCA0212		hypothetical protein	N/A
198576	E10	378	VC2144	flaE	flagellin FlaE	NP_231775.1
198605	E12	224	VC0495		conserved hypothetical protein	NP_230149.1
198710	F01	269	VC0441	apaH	bis(5' -nucleosyl)-tetraphosphatase	NP_230095.1
198735	F02	316	VC0454		glutaminase family protein	NP_230108.1
198755	F03	353	VC0452	mutY	A-G-specific adenine glycosylase	NP_230106.1
198766	F04	N/A	VCA0539		conserved hypothetical protein	N/A
198777	F05	469	VC0921		polysaccharide export protein, putative	NP_230568.1
198522	F07	263	VC0948		rare lipoprotein A, putative	NP_230595.1
198542	F08	302	VC2145		tyrA protein	NP_231776.1
198560	F09	351	VC2136	flrB	flagellar regulatory protein B	NP_231767.1
198580	F10	391	VC0947	dacA-1	D-alanyl-D-alanine carboxypeptidase	NP_230594.1
198593	F11	N/A	VCA0240		hypothetical protein	N/A
198606	F12	N/A	VCA0238		sensor histidine kinase	N/A
198712	G01	272	VC0460	proC	pyrroline-5-carboxylate reductase	NP_230114.1
198738	G02	N/A	VCA0535		hypothetical protein	N/A
198756	G03	177	VC2096	seqA	seqA protein	NP_231728.1
198767	G04	413	VC0918	epsD	UDP-N-acetyl-D-mannosaminuronic acid dehydrogenase	NP_230565.1
198778	G05	229	VC2100		conserved hypothetical protein	NP_231732.1
198790	G06	257	VC2097		esterase-lipase YbfF, putative	NP_231729.1
198526	G07	N/A	VCA0243		conserved hypothetical protein	N/A
198544	G08	N/A	VC0501		transposase, authentic frameshift	N/A
198562	G09	N/A	VCA0231		transcriptional regulator, AraC-XylS family	N/A
198581	G10	156	VC0951		conserved hypothetical protein	NP_230598.1
198594	G11	474	VC0962		conserved hypothetical protein	NP_230609.1
198607	G12	N/A	VCA0244		sugar isomerase SgaE, AraD-FucA family	N/A
198714	H01	269	VC0905	yaeC	lipoprotein YaeC	NP_230552.2
198739	H02	N/A	VCA0546		conserved hypothetical protein	N/A
198757	H03	N/A	VCA0550		hypothetical protein	N/A
198768	H04	N/A	VCA0549	phnA	phnA protein	N/A
198779	H05	478	VC0910	treB	PTS system, trehalose-specific IIBC component	NP_230557.1
198791	H06	548	VC2095	pgm	phosphoglucomutase	NP_231727.1
198528	H07	N/A	VC0499	orfB	transposase OrfAB, subunit B authentic point mutation	N/A
198546	H08	N/A	VCA0228	vctD	vibriobactin and enterobactin ABC transporter, permease protein	N/A
198564	H09	N/A	VCA0217		GGDEF family protein	N/A
198596	H11	479	VC2135	flrC	flagellar regulatory protein C	NP_231766.1