

***Vibrio cholerae* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 18**

Catalog No. NR-19696

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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 El 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-19696 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 18, NR-19696.”

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 18

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
198868	A02	239	VC2064	cheZ	chemotaxis protein CheZ	NP_231696.1
198800	A03	313	VC2067		MinD-related protein	NP_231699.1
198820	A04	377	VC2062	cheB-2	protein-glutamate methyltransferase CheB	NP_231694.1
198836	A05	468	VC0433		arginine-ornithine antiporter	NP_230087.1
198849	A06	N/A	VCA1111		thermostable hemolysin	N/A
198632	A08	318	VC0468	gshB	glutathione synthetase	NP_230122.1
198652	A09	368	VC0463	pilT-2	twitching motility protein PilT	NP_230117.1
198672	A10		VC0922		hypothetical protein	NP_230569.1
198687	A11	187	VC0467		conserved hypothetical protein	NP_230121.2
198701	A12	250	VC0927	cpsF	UDP-N-acetyl-D-mannosamine transferase	NP_230574.1
198608	B01	580	VC2133	fliF	flagellar M-ring protein FliF	NP_231764.1
198870	B02	N/A	VCA0021		hypothetical protein	N/A
198804	B03	430	VC0430		immunogenic protein	NP_230084.1
198821	B04	145	VC0870	tnpA	IS1004 transposase	NP_230517.1
198840	B05	481	VC0422	tldD	tldD protein	NP_230076.1
198850	B06	530	VC0866		transglycosylase, Slt family	NP_230513.1
198616	B07	N/A	VCA0194		potassium channel protein, putative	N/A
198634	B08	N/A	VCA0559		hypothetical protein	N/A
198654	B09	N/A	VCA0200		hypothetical protein	N/A
198674	B10	408	VC0929		hypothetical protein	NP_230576.1
198688	B11	465	VC0934		capsular polysaccharide biosynthesis glycosyltransferase, putative	NP_230581.1
198609	C01	249	VC2153		D,D-carboxypeptidase-related protein	NP_231784.1
198873	C02	244	VC2066	fliA	RNA polymerase sigma factor for flagellar operon FliA	NP_231698.1
198805	C03	103	VC0887		conserved hypothetical protein	NP_230534.1
198827	C04	150	VC0407	mshF	MSHA biogenesis protein MshF	NP_230061.1
198842	C05	489	VC0419	cafA	cytoplasmic axial filament protein	NP_230073.1
198851	C06	203	VC0411	mshD	MSHA pilin protein MshD	NP_230065.1
198618	C07	287	VC0480		conserved hypothetical protein	NP_230134.1
198636	C08	N/A	VCA0192	ldhA	D-lactate dehydrogenase	N/A
198660	C09	341	VC0476	epd	D-erythrose-4-phosphate dehydrogenase	NP_230130.2
198676	C10	N/A	VCA0554		oxalate-formate antiporter	N/A
198690	C11	N/A	VCA0557		GGDEF family protein	N/A
198706	C12	652	VC0475	irgA	enterobactin receptor	NP_230129.1
198612	D01	N/A	VCA0211		sensory box sensor histidine kinase	N/A
198877	D02	256	VC0412		hypothetical protein	NP_230066.1
198806	D03	N/A	VCA0024		conserved hypothetical protein	N/A
198829	D04	157	VC2070	sixA	phosphohistidine phosphatase	NP_231702.1
198844	D05	495	VC2068		flagellar biosynthetic protein FliH, putative	NP_231700.1
198620	D07	N/A	VCA0201		hypothetical protein	N/A
198638	D08	338	VC2107		aspartate-semialdehyde dehydrogenase, putative	NP_231739.1
198662	D09	387	VC2108	pdxB	erythronate-4-phosphate dehydrogenase	NP_231740.1
198678	D10	N/A	VCA0560		GGDEF family protein	N/A
198692	D11	470	VC0485	pykF	pyruvate kinase I	NP_230139.1

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
198708	D12	653	VC0490		conserved hypothetical protein	NP_230144.1
198860	E01	214	VC0873		conserved hypothetical protein	NP_230520.1
198881	E02	265	VC0425		hypothetical protein	NP_230079.1
198808	E03	338	VC2060		conserved hypothetical protein	NP_231692.1
198830	E04	407	VC0406	mshG	MSHA biogenesis protein MshG	NP_230060.1
198845	E05	196	VC0408	mshB	MSHA pilin protein MshB	NP_230062.1
198853	E06	205	VC0418	maf	maf protein	NP_230072.1
198622	E07	292	VC2103		transcriptional regulator, LysR family	NP_231735.1
198640	E08	N/A	VCA0197	guaC	GMP reductase	N/A
198664	E09	390	VC0926		hypothetical protein	NP_230573.1
198681	E10	N/A	VCA0556		hypothetical protein	N/A
198694	E11	N/A	VCA0191		conserved hypothetical protein	N/A
200104	E12	227	VC0258	rfbT	rfbT protein	NP_229914.1
198862	F01	N/A	VCA1105		DNA-binding response regulator	N/A
198883	F02	N/A	VCA1108		oxidoreductase, short-chain dehydrogenase-reductase family	N/A
198812	F03	345	VC0886		hypothetical protein	NP_230533.1
198833	F04	162	VC0410	mshC	MSHA pilin protein MshC	NP_230064.1
198846	F05	N/A	VCA1110		long-chain-fatty-acid--CoA ligase, putative	N/A
198854	F06	559	VC0402	mshL	MSHA biogenesis protein MshL	NP_230056.1
198626	F07	298	VC0474	irgB	iron-regulated virulence regulatory protein IrgB	NP_230128.1
198642	F08	349	VC2126	fliM	flagellar motor switch protein FliM	NP_231757.1
198665	F09	N/A	VCA0190		hypothetical protein	N/A
198682	F10	406	VC0924		capK protein, putative	NP_230571.1
198695	F11	235	VC0938		hypothetical protein	NP_230585.1
200106	F12	247	VC0230		hypothetical protein	NP_229887.1
198864	G01	231	VC0876		conserved hypothetical protein	NP_230523.1
198889	G02	N/A	VCA0019		hypothetical protein	N/A
198813	G03	N/A	VCA0022		glutathione S-transferase-related protein	N/A
198834	G04	N/A	VCA0025		transporter, NadC family	N/A
198847	G05	198	VC2073		conserved hypothetical protein	NP_231705.1
198855	G06	207	VC0884		acetyltransferase-related protein	NP_230531.1
198628	G07	299	VC2123	fliP	flagellar biosynthetic protein FliP	NP_231754.1
198644	G08	358	VC0478	fbaA	fructose-bisphosphate aldolase, class II	NP_230132.1
198667	G09	162	VC0471		sprT protein, putative	NP_230125.1
198684	G10	N/A	VCA0193		Na ⁺ -H ⁺ antiporter, putative	N/A
198697	G11	238	VC0483		conserved hypothetical protein	NP_230137.1
200108	G12	251	VC0227		conserved hypothetical protein	NP_229884.1
198866	H01	N/A	VCA1109		hypothetical protein	N/A
198891	H02	296	VC0416	mreC	rod shape-determining protein MreC	NP_230070.1
198818	H03	311	VC0432	mdh	malate dehydrogenase	NP_230086.2
198835	H04	167	VC2058		hypothetical protein	NP_231690.1
198856	H06	575	VC0405	mshE	MSHA biogenesis protein MshE	NP_230059.1
198630	H07	314	VC2118		adenine-specific methylase, putative	NP_231749.1
198648	H08	361	VC2116	aroC	chorismate synthase	NP_231747.1
198668	H09	398	VC0935		hypothetical protein	NP_230582.1
198686	H10	N/A	VCA0564	pntB	NAD(P) transhydrogenase, beta subunit	N/A