

***Vibrio cholerae* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 6**

Catalog No. NR-19684

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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-19684 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 6, NR-19684.”

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 6

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
197990	A02	N/A	VCA0593		hypothetical protein	N/A
197999	A03	147	VC1713		transcriptional regulator, AsnC family	NP_231349.1
198008	A04	N/A	VCA0629		conserved hypothetical protein	N/A
198019	A05	200	VC1056	recR	recombination protein RecR	NP_230701.1
198032	A06	450	VC0600		hypothetical protein	NP_230250.1
198047	A07	247	VC1715	mukE	mukE protein	NP_231351.1
197976	B01	281	VC2238		conserved hypothetical protein	NP_231869.1
197992	B02	N/A	VCA0623	talB	transaldolase B	N/A
198000	B03	N/A	VCA0129	rbsC	ribose ABC transporter, permease protein	N/A
198009	B04	N/A	VCA0627		rRNA methylase, putative	N/A
198020	B05	N/A	VCA0136	glpQ	glycerophosphoryl diester phosphodiesterase	N/A
198034	B06	452	VC2253		conserved hypothetical protein	NP_231884.1
198049	B07	247	VC1059		oxidoreductase, short-chain dehydrogenase-reductase family	NP_230704.1
197978	C01	282	VC1718		conserved hypothetical protein	NP_231354.1
197993	C02	N/A	VCA0607		regulator of nucleoside diphosphate kinase	N/A
198001	C03	148	VC0596	dksA	dnaK suppressor protein	NP_230246.1
198011	C04	174	VC2240		decarboxylase	NP_231871.1
198021	C05	204	VC1712		conserved hypothetical protein	NP_231348.1
198035	C06	N/A	VCA0616	folE	GTP cyclohydrolase	N/A
197980	D01	286	VC1063	tesB	acyl-CoA thioesterase II	NP_230708.1
197994	D02	N/A	VCA0595		hypothetical protein	N/A
198002	D03	N/A	VCA0132	rbsR	ribose operon repressor	N/A
198014	D04	353	VC1060		sohB protein, peptidase U7 family	NP_230705.2
198024	D05	N/A	VCA0602		ABC transporter, ATP-binding protein	N/A
198037	D06	234	VC1719	torR	DNA-binding response regulator TorR	NP_231355.1
197982	E01	N/A	VCA0130	rbsB	ribose ABC transporter, periplasmic D-ribose-binding protein	N/A
197995	E02	N/A	VCA0612	mscL	large-conductance mechanosensitive channel	N/A
198003	E03	153	VC2249	fabZ	(3R)-hydroxymyristoyl-(acyl-carrier-protein) dehydratase	NP_231880.1
198015	E04	N/A	VCA0592		MutT-nudix family protein	N/A
198025	E05	N/A	VCA0610		sigma cross-reacting protein 27A	N/A
198040	E06	484	VC1726	glgA	glycogen synthase	NP_231362.1
197984	F01	N/A	VCA0599		hypothetical protein	N/A
197996	F02	319	VC2244	accA	acetyl-CoA carboxylase, carboxyl transferase alpha subunit	NP_231875.1
198004	F03	336	VC1721		transcriptional regulator, LacI family	NP_231357.1
198016	F04	N/A	VCA0590		peptide ABC transporter, permease protein, putative	N/A
198027	F05	220	VC1720	torD	torD protein	NP_231356.1
198041	F06	N/A	VCA0134	udp-2	uridine phosphorylase	N/A
197987	G01	114	VC2239	glnB-2	nitrogen regulatory protein P-II	NP_231870.1
197997	G02	145	VC1058		conserved hypothetical protein	NP_230703.1
198005	G03	156	VC2234	rnhA	ribonuclease HI	NP_231865.1

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
198017	G04	195	VC1722		conserved hypothetical protein	NP_231358.1
198029	G05	N/A	VCA0608		conserved hypothetical protein	N/A
198043	G06	N/A	VCA0133	pnuC	pnuC protein, authentic point mutation	N/A
197989	H01	120	VC1055		conserved hypothetical protein	NP_230700.1
197998	H02	N/A	VCA0621		transcriptional regulator, SorC family	N/A
198006	H03	N/A	VCA0603		ABC transporter, periplasmic substrate-binding protein	N/A
198018	H04	N/A	VCA0604	phnW	2-aminoethylphosphonate:pyruvate aminotransferase	N/A
198030	H05	445	VC1716	mukF	mukF protein	NP_231352.1
198045	H06	245	VC2235		conserved hypothetical protein	NP_231866.1