

***Vibrio cholerae* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 22**

Catalog No. NR-19700

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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-19700 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 22, NR-19700.”

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 22

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Description	Accession Number
200457	A02	516	VC2490	leuA	2-isopropylmalate synthase	NP_232119.1
200067	A04	326	VC0270		ROK family protein	NP_229926.1
200093	A05	150	VC0297	aroQ	3-dehydroquinate dehydratase	NP_229952.1
199946	A06	N/A	VCA0422		conserved hypothetical protein	N/A
199981	A07	450	VC1386		heat shock protein 70 family protein	NP_231030.1
200002	A08	226	VC2624	gph	phosphoglycolate phosphatase	NP_232252.1
200789	A09	428	VC1111	bioA	adenosylmethionine-8-amino-7-oxononanoate aminotransferase	NP_230756.1
200805	A10	449	VC1108		conserved hypothetical protein	NP_230753.1
199019	A11	N/A	VCA1048		oxidoreductase, Gfo-Idh-MocA family	N/A
199051	A12	208	VC1497		conserved hypothetical protein	NP_231138.1
200442	B01	212	VC1622		outer membrane protein, putative	NP_231261.1
200458	B02	N/A	VCA0853		hypothetical protein	N/A
200040	B03	N/A	VCA0381		hypothetical protein	N/A
200068	B04	N/A	VCA0385		conserved hypothetical protein	N/A
200094	B05	541	VC0278		transcriptional activator CadC, putative	NP_229934.1
199950	B06	116	VC1380		hypothetical protein	NP_231024.1
199983	B07	N/A	VCA0931		conserved hypothetical protein	N/A
200003	B08	631	VC1374		DnaK-related protein	NP_231018.1
200791	B09	430	VC2278		conserved hypothetical protein	NP_231909.1
200811	B10	489	VC1104		GGDEF family protein	NP_230749.1
200443	C01	467	VC2492	leuC	3-isopropylmalate dehydratase, large subunit	NP_232121.1
200459	C02	543	VC1313		methyl-accepting chemotaxis protein	NP_230957.1
200046	C03	N/A	VCA0353		hypothetical protein	N/A
200070	C04	N/A	VCA0372	orfA	transposase OrfAB, subunit A	N/A
199951	C06	316	VC1390		transcriptional regulator, LysR family	NP_231034.1
200004	C08	226	VC1371		hypothetical protein	NP_231015.1
200813	C10	505	VC2285		GGDEF family protein	NP_231916.1
199056	C12	530	VC1520		ABC transporter, ATP-binding protein	NP_231160.1
200445	D01	478	VC1632		conserved hypothetical protein	NP_231269.1
200461	D02	543	VC1298		methyl-accepting chemotaxis protein	NP_230943.1
200077	D04	N/A	VCA0369		hypothetical protein	N/A
200097	D05	N/A	VCA0409		hypothetical protein	N/A
199952	D06	N/A	VCA0935		hypothetical protein	N/A
199991	D07	476	VC2559	cysN	sulfate adenylate transferase, subunit 1	NP_232187.1
200815	D10	534	VC2279	pepD	aminoacyl-histidine dipeptidase	NP_231910.1
199042	D11	194	VC2729	epsH	general secretion pathway protein H	NP_232356.1
199057	D12	542	VC2738	pckA	phosphoenolpyruvate carboxykinase	NP_232364.1
200449	E01	492	VC1295		conserved hypothetical protein	NP_230940.1
200463	E02	573	VC2483	ilvI	acetolactate synthase III, large subunit	NP_232112.1
200059	E03	N/A	VCA0399		hypothetical protein	N/A
200083	E04	133	VC0277		transcriptional regulator, MerR family	NP_229933.1
200098	E05	607	VC0300		conserved hypothetical protein	NP_229955.1

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Description	Accession Number
199995	E07	485	VC2616	aruD	succinylglutamate 5-semialdehyde dehydrogenase	NP_232244.1
200006	E08	231	VC1366	exsB	exsB protein	NP_231010.1
200797	E09	439	VC1084		sensory box sensor histidine kinase	NP_230729.1
200817	E10	534	VC1088		sensor histidine kinase	NP_230733.1
199043	E11	419	VC1500		PqiA family protein	NP_231141.1
199062	E12	N/A	VCA1041		phosphomannomutase, putative	N/A
200451	F01	505	VC1304		fumarate hydratase, class I, putative	NP_230949.1
200465	F02	601	VC2484		long-chain-fatty-acid--CoA ligase, putative	NP_232113.1
200060	F03	N/A	VCA0359		plasmid stabilization element ParE, putative	N/A
200086	F04	447	VC0295	accC	acetyl-CoA carboxylase, biotin carboxylase	NP_229950.1
200099	F05	N/A	VCA0360		hypothetical protein	N/A
199959	F06	332	VC1972	menC	o-succinylbenzoate-CoA synthase	NP_231606.1
199996	F07	210	VC2614		cyclic AMP receptor protein	NP_232242.1
200007	F08	664	VC2608		ABC transporter, ATP-binding protein	NP_232236.1
200802	F09	198	VC1107	lolA	outer membrane lipoproteins carrier protein	NP_230752.1
200819	F10	536	VC1086		response regulator	NP_230731.1
199048	F11	468	VC2749	ntrC	nitrogen regulation protein NR(I)	NP_232375.1
199064	F12	609	VC2744	typA	GTP-binding protein TypA	NP_232370.1
200453	G01	N/A	VCA0843	gapA-2	glyceraldehyde 3-phosphate dehydrogenase	N/A
200467	G02	657	VC1636		helicase-related protein	NP_231273.1
200061	G03	295	VC0293	prmA	ribosomal protein L11 methyltransferase	NP_229948.1
200088	G04	455	VC0264		conserved hypothetical protein	NP_229920.1
200100	G05	598	VC0288	edd	phosphogluconate dehydratase	NP_229943.2
199970	G06	161	VC2609		hypothetical protein	NP_232237.1
199997	G07	504	VC1392		deoxyribodipyrimidine photolyase, putative	NP_231036.1
200767	G08	360	VC2287	dinP	DNA-damage-inducible protein P	NP_231918.1
200803	G09	447	VC1087		response regulator	NP_230732.1
200821	G10	543	VC1091	oppA	oligopeptide ABC transporter, periplasmic oligopeptide-binding protein	NP_230736.1
199049	G11	182	VC2721		MutT-nudix family protein	NP_232348.2
199066	G12	610	VC1499		ABC transporter, ATP-binding protein	NP_231140.1
200455	H01	513	VC1308	tyrR	transcriptional regulator TyrR	NP_230952.1
200017	H02	N/A	VCA0402		acetyltransferase, putative	N/A
200062	H03	N/A	VCA0362		hypothetical protein	N/A
200092	H04	530	VC0276	purH	phosphoribosylaminoimidazolecarboxamide formyltransferase-IMP cyclohydrolase	NP_229932.1
199919	H05	257	VC2622		hypothetical protein	NP_232250.1
199976	H06	176	VC1377		conserved hypothetical protein	NP_231021.1
200787	H08	417	VC2273	proA	gamma-glutamyl phosphate reductase	NP_231904.1
200824	H10	N/A	VCA0646		conserved hypothetical protein-hemolysin, putative	N/A
199050	H11	469	VC2746	glnA	glutamate--ammonia ligase	NP_232372.1