

***Vibrio cholerae* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 14**

Catalog No. NR-19692

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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-19692 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 14, NR-19692.”

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 cholera*." *Nature* 406 (2000): 477-483. PubMed. 10952301.

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Table 1: *Vibrio cholerae* Gateway® Clones, Plate 14

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
200588	A04	N/A	VCA0686		iron(III) ABC transporter, permease protein	N/A
200599	A05	881	VC2262	glnD	protein-P-II uridylyltransferase	NP_231893.1
200607	A06	N/A	VCA0574		conserved hypothetical protein	N/A
200617	A07	948	VC2438	glnE	glutamate-ammonia-ligase adenyltransferase	NP_232067.1
200635	A08	760	VC1740		oxidoreductase, acyl-CoA dehydrogenase family	NP_231376.1
198146	A09	235	VC2198	flgD	basal-body rod modification protein FlgD	NP_231829.1
198164	A10	264	VC0999	truA	tRNA pseudouridine synthase A	NP_230645.1
198181	A11	N/A	VCA0277	gcvH	glycine cleavage system H protein	N/A
198190	A12	367	VC0568		conserved hypothetical protein	NP_230219.1
200558	B01	672	VC1898		methyl-accepting chemotaxis protein	NP_231532.1
200569	B02	793	VC1769		DNA methylase HsdM, putative	NP_231404.1
200578	B03	691	VC1888		hemolysin-related protein	NP_231522.1
200590	B04	701	VC0723	ppk	polyphosphate kinase	NP_230372.1
200608	B06	714	VC1097	pta	phosphate acetyltransferase	NP_230742.1
200619	B07	708	VC1047	yfcX	fatty oxidation complex, alpha subunit	NP_230692.2
200636	B08	764	VC0603		conserved hypothetical protein	NP_230253.1
198148	B09	N/A	VCA0274	cah	carbonic anhydrase	N/A
198166	B10	267	VC0559		conserved hypothetical protein	NP_230210.1
198182	B11	312	VC2192	flgJ	flagellar protein FlgJ	NP_231823.1
198191	B12	138	VC2199	flgC	flagellar basal-body rod protein FlgC	NP_231830.1
200559	C01	787	VC1212	polB	DNA polymerase II	NP_230857.1
200570	C02	N/A	VCA0141		C4-dicarboxylate transport sensor protein, putative	N/A
200581	C03	820	VC0601	hrpB	ATP-dependent helicase HrpB	NP_230251.1
200601	C05	883	VC2217	chb-2	beta-N-acetylhexosaminidase	NP_231848.1
200609	C06	915	VC2215		cation transport ATPase, E1-E2 family	NP_231846.1
200622	C07	1021	VC1198		conserved hypothetical protein	NP_230843.1
200637	C08	768	VC1033		cation transport ATPase, E1-E2 family	NP_230678.1
198150	C09	243	VC1006	rnt	ribonuclease T	NP_230652.1
198168	C10	275	VC2201	cheR-2	chemotaxis protein methyltransferase CheR	NP_231832.1
198183	C11	N/A	VCA0265		hypothetical protein	N/A
198192	C12	376	VC0551	oadB-1	oxaloacetate decarboxylase, beta subunit	NP_230202.1
200571	D02	801	VC0612		cellobiose-cellobextrin-phosphorylase, putative	NP_230261.1
200593	D04	N/A	VCA0736	luxQ	sensor histidine kinase LuxQ	N/A
200602	D05	706	VC2319	recD	exodeoxyribonuclease V, 67 kDa subunit	NP_231950.1
200610	D06	718	VC0200		iron(III) compound receptor	NP_229857.1
200628	D07	738	VC2451	relA	GTP pyrophosphokinase	NP_232080.1
200638	D08	1155	VC1886	mfd	transcription-repair coupling factor	NP_231520.1
198152	D09	247	VC0563	trmD	tRNA (guanine-N1)-methyltransferase	NP_230214.1
198172	D10	294	VC1008	motY	sodium-type flagellar protein MotY	NP_230654.1
198184	D11	337	VC0552		quinone oxidoreductase	NP_230203.1
198194	D12	377	VC2187	flaC	flagellin FlaC	NP_231818.1
200562	E01	N/A	VCA0805	rnb	exoribonuclease II	N/A
200572	E02	685	VC0658		c-di-GMP phosphodiesterase A-related protein	NP_230307.1

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
200583	E03	819	VC2364	thrA	aspartokinase I-homoserine dehydrogenase, threonine-sensitive	NP_231994.2
200594	E04	691	VC1855		ATP-dependent helicase, DinG family	NP_231489.2
200603	E05	886	VC2414	aceE	pyruvate dehydrogenase, E1 component	NP_232044.1
200612	E06	N/A	VCA0146		conserved hypothetical protein	N/A
200630	E07	741	VC1141	icd	isocitrate dehydrogenase, NADP-dependent, monomeric type	NP_230786.1
200639	E08	777	VC0602	mrcB	penicillin-binding protein 1B	NP_230252.1
198154	E09	249	VC2196	flgF	flagellar basal-body rod protein FlgF	NP_231827.1
198174	E10	295	VC2182		kinase, GHMP family	NP_231813.1
198186	E11	352	VC0565	degS	protease DegS	NP_230216.1
198195	E12	143	VC2205		hypothetical protein	NP_231836.1
200574	F02	687	VC1770		hypothetical protein	NP_231405.1
200584	F03	N/A	VCA0576	hutA	heme transport protein HutA	N/A
200595	F04	865	VC0604	acnB	aconitate hydratase 2	NP_230254.1
200604	F05	706	VC1764		hypothetical protein	NP_231399.1
200614	F06	723	VC0190	uvrD	DNA helicase II	NP_229847.1
200632	F07	748	VC0672	ptsP	phosphoenolpyruvate-protein phosphotransferase	NP_230321.1
200640	F08	1177	VC0622		sensory box sensor histidine kinase-response regulator	NP_230271.1
198156	F09	255	VC2203		flagellar protein, putative	NP_231834.1
198177	F10	N/A	VCA0271		hypothetical protein	N/A
198187	F11	131	VC2200	flgB	flagellar basal-body rod protein FlgB	NP_231831.1
198196	F12	N/A	VCA0267	emrD-3	multidrug resistance protein D	N/A
200565	G01	789	VC1711		conserved hypothetical protein	NP_231347.1
200585	G03	829	VC0130		GGDEF family protein	NP_229788.1
200605	G05	N/A	VCA0157		NADH dehydrogenase, putative	N/A
200615	G06	940	VC1760		helicase, putative	NP_231395.1
200633	G07	1073	VC0629		multidrug resistance protein, putative	NP_230278.1
200641	G08	779	VC2344		hypothetical protein	NP_231974.1
198158	G09	N/A	VCA0266		transcriptional regulator, AraC-XylS family	N/A
198178	G10	308	VC2202	cheV-3	chemotaxis protein CheV	NP_231833.1
198188	G11	361	VC2193	flgI	flagellar P-ring protein FlgI	NP_231824.1
198197	G12	145	VC2206		conserved hypothetical protein	NP_231837.1
200566	H01	N/A	VCA0624	tktA-2	transketolase 1	N/A
200576	H02	687	VC2211	viuA	ferric vibriobactin receptor	NP_231842.1
200586	H03	N/A	VCA0717	helD	helicase IV	N/A
200598	H04	704	VC1211		conserved hypothetical protein	NP_230856.1
200606	H05	N/A	VCA0165		GGDEF family protein	N/A
200616	H06	N/A	VCA0728		hypothetical protein	N/A
200634	H07	756	VC1144	clpA	ATP-dependent Clp protease, ATP-binding subunit ClpA	NP_230789.1
200642	H08	1208	VC2320	recB	exodeoxyribonuclease V, 135 kDa subunit	NP_231951.1
198162	H09	262	VC2195	flgG	flagellar basal-body rod protein FlgG	NP_231826.1
198179	H10	117	VC0564	rplS	ribosomal protein L19	NP_230215.1
198189	H11	N/A	VCA0262		hypothetical protein	N/A