

***Vibrio cholerae* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 10**

**Catalog No. NR-19688**

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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.

Plate orientation and viability were confirmed for NR-19688.

**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-19688 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 10, NR-19688.”

**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](http://www.cdc.gov/biosafety/publications/bmb15/index.htm).

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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.

ATCC® is a trademark of the American Type Culture Collection.



**Table 1: *Vibrio cholerae* Gateway® Clones, Plate 10**

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
200228	A02	N/A	VCA0885	tdh	threonine 3-dehydrogenase	N/A
200242	A03	N/A	VCA0903		conserved hypothetical protein	N/A
200256	A04	421	VC2554		conserved hypothetical protein	NP_232182.1
200378	A05	282	VC1296	thiD	phosphomethylpyrimidine kinase	NP_230941.1
200400	A06	362	VC2487		conserved hypothetical protein	NP_232116.1
200412	A07	402	VC2475	ubiH	ubiH protein	NP_232104.1
200423	A08	N/A	VCA0854		conserved hypothetical protein	N/A
200438	A09	N/A	VCA0832		methyltransferase, putative	N/A
200009	A10	N/A	VCA0397		hypothetical protein	N/A
200031	A11	N/A	VCA0368		hypothetical protein	N/A
200047	A12	N/A	VCA0370		hypothetical protein	N/A
200217	B01	N/A	VCA0919		conserved hypothetical protein	N/A
200230	B02	N/A	VCA0884		hypothetical protein	N/A
200243	B03	173	VC2555		hypothetical protein	NP_232183.1
200259	B04	N/A	VCA0901		hypothetical protein	N/A
200380	B05	292	VC1311		conserved hypothetical protein	NP_230955.1
200402	B06	363	VC2491	leuB	3-isopropylmalate dehydrogenase	NP_232120.1
200413	B07	409	VC2481	serA	D-3-phosphoglycerate dehydrogenase	NP_232110.1
200424	B08	164	VC2482	ilvH	acetolactate synthase III, small subunit	NP_232111.1
200440	B09	211	VC2489		transcriptional regulator, TetR family	NP_232118.1
200013	B10	N/A	VCA0406	blc-3	lipoprotein Blc	N/A
200033	B11	N/A	VCA0405		hypothetical protein	N/A
200049	B12	N/A	VCA0356		hypothetical protein	N/A
200218	C01	330	VC2539	tbpA	thiamin ABC transporter, periplasmic thiamin-binding protein	NP_232167.1
200232	C02	365	VC1360		amino acid ABC transporter, permease protein	NP_231004.1
200245	C03	N/A	VCA0907	hutZ	heme-binding protein HutZ	N/A
200263	C04	212	VC2549	msrA	peptide methionine sulfoxide reductase	NP_232177.1
200382	C05	N/A	VCA0833		transcriptional regulator, LysR family	N/A
200404	C06	372	VC1628		conserved hypothetical protein	NP_231265.1
200414	C07	135	VC1637		hypothetical protein	NP_231274.1
200425	C08	407	VC1312		alanine racemase, putative	NP_230956.2
200444	C09	218	VC2480	rpiA	ribose-5-phosphate isomerase	NP_232109.1
200019	C10	184	VC0263		galactosyl-transferase, putative	NP_229919.1
200035	C11	N/A	VCA0355		conserved hypothetical protein	N/A
200050	C12	N/A	VCA0386		hypothetical protein	N/A
200220	D01	336	VC2544	fbp	fructose-1,6-bisphosphatase	NP_232172.1
200235	D02	148	VC2531	ptsN	PTS system, nitrogen regulatory IIA component	NP_232159.1
200246	D03	N/A	VCA0905		conserved hypothetical protein	N/A
200266	D04	453	VC2534	mgtE-2	magnesium transporter	NP_232162.1
200384	D05	306	VC1317		conserved hypothetical protein	NP_230961.1
200405	D06	102	VC2478		conserved hypothetical protein	NP_232107.1
200416	D07	157	VC1310		hypothetical protein	NP_230954.1
200427	D08	N/A	VCA0837		hemolysin, putative	N/A
200446	D09	219	VC1638		DNA-binding response regulator	NP_231275.1

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
200023	D10	190	VC0266		conserved hypothetical protein	NP_229922.1
200037	D11	N/A	VCA0388		hypothetical protein	N/A
200051	D12	244	VC0274		hypothetical protein	NP_229930.1
200221	E01	102	VC1957		conserved hypothetical protein	NP_231294.1
200237	E02	157	VC1350		antioxidant, putative	NP_230994.1
200247	E03	176	VC2545	ppa	inorganic pyrophosphatase	NP_232173.1
200267	E04	220	VC1959	minC	septum site-determining protein MinC	NP_231296.1
200388	E05	N/A	VCA0834		hypothetical protein	N/A
200406	E06	382	VC1627	nhaA	Na <sup>+</sup> -H <sup>+</sup> antiporter protein	NP_231264.1
200418	E07	157	VC2476		conserved hypothetical protein	NP_232105.1
200430	E08	414	VC1621		agglutination protein	NP_231260.1
200448	E09	N/A	VCA0856		hypothetical protein	N/A
200025	E10	N/A	VCA0361		hypothetical protein	N/A
200039	E11	222	VC0268		conserved hypothetical protein	NP_229924.1
200053	E12	N/A	VCA0396		hypothetical protein	N/A
200222	F01	339	VC1346		conserved hypothetical protein	NP_230990.1
200238	F02	369	VC1344	hppD	4-hydroxyphenylpyruvate dioxygenase	NP_230988.1
200248	F03	N/A	VCA0886	kbl	2-amino-3-ketobutyrate coenzyme A ligase	N/A
200273	F04	N/A	VCA0915	hutD	hemin ABC transporter, ATP-binding protein HutD	N/A
200390	F05	321	VC2485		transcriptional regulator, LysR family	NP_232114.1
200409	F06	122	VC1299		6-pyruvoyl tetrahydrobiopterin synthase, putative	NP_230944.1
200419	F07	414	VC1624		conserved hypothetical protein	NP_231263.1
200431	F08	194	VC2479		conserved hypothetical protein	NP_232108.1
200454	F09	231	VC1635		ribosomal small subunit pseudouridine synthase A	NP_231272.1
200026	F10	42	VC0279		hypothetical protein	NP_229935.1
200041	F11	223	VC0299		DNA polymerase III, epsilon subunit, putative	NP_229944.1
200055	F12	N/A	VCA0395		conserved hypothetical protein	N/A
200224	G01	342	VC1362		amino acid ABC transporter, periplasmic amino acid-binding protein	NP_231006.1
200239	G02	N/A	VCA0917		transcriptional regulator, TetR family	N/A
200249	G03	179	VC2527		conserved hypothetical protein	NP_232155.1
200274	G04	487	VC2529	rpoN	RNA polymerase sigma-54 factor	NP_232157.1
200410	G06	401	VC1634		multidrug resistance protein	NP_231271.1
200420	G07	158	VC1316		chemotaxis protein CheY, putative	NP_230960.1
200433	G08	200	VC2493	leuD	3-isopropylmalate dehydratase, small subunit	NP_232122.1
200462	G09	257	VC1641		conserved hypothetical protein	NP_231278.1
200027	G10	N/A	VCA0380		hypothetical protein	N/A
200043	G11	225	VC0283		hypothetical protein	NP_229939.1
200056	G12	90	VC0273	hupA	DNA-binding protein HU	NP_229929.1
200227	H01	N/A	VCA0890		glyoxylase I family protein	N/A
200240	H02	N/A	VCA0883		hypothetical protein	N/A
200252	H03	405	VC1953		NupC family protein	NP_231290.1
200283	H04	244	VC2537		thiamine ABC transporter, ATP-binding protein, putative	NP_232165.1
200398	H05	N/A	VCA0851		conserved hypothetical protein	N/A
200411	H06	133	VC1642		hypothetical protein	NP_231279.1
200421	H07	417	VC1301	sdaC-1	serine transporter	NP_230946.1
200435	H08	N/A	VCA0846		conserved hypothetical protein	N/A
200464	H09	N/A	VCA0850		response regulator	N/A
200029	H10	201	VC0285	eda	4-hydroxy-2-oxoglutarate aldolase-2-dehydro-3-deoxyphosphogluconate aldolase	NP_229940.1
200045	H11	N/A	VCA0363		hypothetical protein	N/A