

***Vibrio cholerae* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 1**

**Catalog No. NR-19679**

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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.

Plate orientation and viability were confirmed for NR-19679.

**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-19679 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 1, NR-19679.”

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

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



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

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
174071	A02	367	VC2271	ribD	riboflavin-specific deaminase	NP_231902.1
174346	A03	336	VC1877	lpxK	tetraacyldisaccharide 4'-kinase	NP_231511.1
174354	A04	342	VC0953	holA	DNA polymerase III, delta subunit	NP_230600.1
174115	A05	388	VC2085	sucC	succinyl-CoA synthase, beta subunit	NP_231717.1
174310	A06	506	VC2400	murC	UDP-N-acetylmuramate--alanine ligase	NP_232030.1
174523	A07	132	VC0644	rbfA	ribosome-binding factor A	NP_230293.2
174632	A08	322	VC0681	ribF	riboflavin kinase-FMN adenylyltransferase	NP_230330.1
174930	A09	433	VC0720	phoR	histidine protein kinase PhoR	NP_230369.1
174953	A10	206	VC1178		conserved hypothetical protein	NP_230823.1
174976	A11	213	VC2358		hypothetical protein	NP_231988.1
174898	A12	369	VC0154	trmA	tRNA (uracil-5-)-methyltransferase	NP_229811.1
174059	B01	73	VC2098		hypothetical protein	NP_231730.1
174075	B02	82	VC0561	rpsP	ribosomal protein S16	NP_230212.1
174087	B03	378	VC1843	cydB-1	cytochrome d ubiquinol oxidase, subunit II	NP_231477.1
174099	B04	383	VC1798	eha	eha protein	NP_231433.1
174294	B05	494	VC0763		GTP-binding protein	NP_230412.1
174311	B06	314	VC2183	prsA	ribose-phosphate pyrophosphokinase	NP_231814.1
174603	B07	108	VC0675	thyA	thymidylate synthase	NP_230324.1
174474	B08	466	VC1297	asnS	asparaginyl-tRNA synthetase	NP_230942.2
174933	B09	198	VC0166		transcriptional regulator, TetR family	NP_229823.1
174773	B10	302	VC0195	rarD	rarD protein	NP_229852.1
175070	B11	268	VC1169	trpA	tryptophan synthase, alpha subunit	NP_230814.1
174902	B12	156	VC0181		conserved hypothetical protein	NP_229838.1
174143	C01	398	VC2397	ftsZ	cell division protein FtsZ	NP_232027.1
174160	C02	142	VC0324	rplK	ribosomal protein L11	NP_229978.1
174172	C03	414	VC1882		conserved hypothetical protein	NP_231516.1
174192	C04	162	VC0326	rplJ	ribosomal protein L10	NP_229980.1
174298	C05	294	VC0890	ispA	geranyltranstransferase	NP_230537.1
174390	C06	362	VC2179	prfA	peptide chain release factor 1	NP_231810.1
174527	C07	545	VC2448	pyrG	CTP synthase	NP_232077.1
174478	C08	105	VC2585	rplX	ribosomal protein L24	NP_232213.1
174847	C09	107	VC0714		hypothetical protein	NP_230363.1
174867	C10	332	VC1172	trpD	anthranilate phosphoribosyltransferase	NP_230817.2
174796	C11	315	VC0174		hypothetical protein	NP_229831.1
175086	C12	279	VC0187		conserved hypothetical protein	NP_229844.1
174322	D01	321	VC0943	lipA	lipoic acid synthetase	NP_230590.1
174250	D02	242	VC2260	rpsB	ribosomal protein S2	NP_231891.1
174176	D03	150	VC2106	fur	ferric uptake regulation protein	NP_231738.1
174278	D04	468	VC0371	dnaB	replicative DNA helicase	NP_230025.1
174378	D05	N/A	VCA0589		peptide ABC transporter, permease protein, putative	N/A
174315	D06	509	VC2214	gltX	glutamyl-tRNA synthetase	NP_231845.1
174547	D07	166	VC2724	epsM	cholerae toxin secretion protein EpsM	NP_232351.1
174724	D08	372	VC1128	trmU	tRNA (5-methylaminomethyl-2-thiouridylate)-methyltransferase	NP_230773.1
174941	D09	199	VC0146		conserved hypothetical protein	NP_229804.1

Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
174870	D10	125	VC0198		hypothetical protein	NP_229855.1
174890	D11	368	VC0165		conserved hypothetical protein	NP_229822.1
174815	D12	85	VC0143		hypothetical protein	NP_229801.1
174063	E01	366	VC0013	dnaN	DNA polymerase III, beta chain	NP_062597.1
174079	E02	N/A	VCA0921		hypothetical protein	N/A
174266	E03	265	VC2670	tpiA	triosephosphate isomerase	NP_232298.1
174362	E04	347	VC0415	mreB	rod shape-determining protein MreB	NP_230069.1
174123	E05	394	VC0362	tufA	elongation factor TU	NP_230016.1
174574	E06	206	VC2572	rpsD	ribosomal protein S4	NP_232200.1
174701	E07	366	VC2500		conserved hypothetical protein	NP_232129.1
175006	E08	233	VC0148	ftsE	cell division ATP-binding protein FtsE	NP_229806.1
174851	E09	330	VC0149	ftsX	cell division protein FtsX	NP_229807.1
174957	E10	466	VC0151	udhA	pyridine nucleotide-disulfide oxidoreductase, class I	NP_229808.1
174894	E11	152	VC0159		RNA-binding protein	NP_229816.1
174910	E12	183	VC0702		conserved hypothetical protein	NP_230351.1
174148	F01	399	VC0215	dfp	DNA-pantothenate metabolism flavoprotein	NP_229872.1
174164	F02	404	VC2086	sucB	2-oxoglutarate dehydrogenase, E2 component, dihydrolipoamide succinyltransferase	NP_231718.1
174095	F03	N/A	VCA0323		conserved hypothetical protein	NA
174286	F04	472	VC0012	dnaA	chromosomal DNA replication initiator DnaA	NP_062596.1
174382	F05	359	VC0332	hemE	uroporphyrinogen decarboxylase	NP_229986.1
174402	F06	385	VC0472	metK	S-adenosylmethionine synthase	NP_230126.1
174466	F07	475	VC2412	lpdA	pyruvate dehydrogenase, E3 component, lipoamide dehydrogenase	NP_232042.1
174831	F08	99	VC0197		gene 3 protein-related protein	NP_229854.1
174855	F09	108	VC0706		sigma-54 modulation protein, putative	NP_230355.1
174874	F10	355	VC0178		patatin family protein	NP_229835.1
175078	F11	270	VC0192		transcriptional regulator, AraC-XylS family	NP_229849.1
175184	F12	315	VC0744	secF-1	protein-export membrane protein SecF	NP_230393.1
174242	G01	212	VC2016	tmk	thymidylate kinase	NP_231650.1
174168	G02	142	VC0756	ndk	nucleoside diphosphate kinase	NP_230405.1
174184	G03	156	VC2768	atpF	ATP synthase F0, B subunit	NP_232394.1
174290	G04	280	VC2255	cdsA	phosphatidate cytidyltransferase	NP_231886.1
174131	G05	394	VC0321	tufB	elongation factor Tu	NP_229975.1
174410	G06	377	VC1255	nrdB	ribonucleoside-diphosphate reductase, beta subunit	NP_230900.2
174470	G07	103	VC2597	rpsJ	ribosomal protein S10	NP_232225.1
174922	G08	404	VC0185		transposase, putative	NP_229842.1
174949	G09	203	VC1173	trpG	anthranilate synthase component II	NP_230818.1
174964	G10	466	VC0717		protease, putative	NP_230366.1
174803	G11	318	VC1182	trxB	thioredoxin reductase	NP_230827.1
175277	G12	379	VC2390	carA	carbamoyl-phosphate synthase, small subunit	NP_232020.1
174330	H01	326	VC2523		conserved hypothetical protein	NP_232152.1
174258	H02	247	VC2233	dnaQ	DNA polymerase III, epsilon subunit	NP_231864.1
174270	H03	467	VC2764	atpD	ATP synthase F1, beta subunit	NP_232390.1
174370	H04	348	VC1014		RnfD-related protein	NP_230660.1
174224	H05	175	VC2099	fldA	flavodoxin 1	NP_231731.1
174515	H06	129	VC2573	rpsK	ribosomal protein S11	NP_232201.1
174560	H07	169	VC0046	def-1	polypeptide deformylase	NP_229705.1
174835	H08	324	VC1907	cysB	cys regulon transcriptional activator	NP_231541.1
174863	H09	123	VC0153		conserved hypothetical protein	NP_229810.1
174886	H10	143	VC0182		hypothetical protein	NP_229839.1
174807	H11	84	VC0716		ferredoxin	NP_230365.1