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SUPPORTING INFECTIOUS DISEASE RESEARCH

Vibrio cholerae Gateway[®] Clone Set, Recombinant in *Escherichia coli*, Plate 11

Catalog No. NR-19689

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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 crosscontamination 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 <u>pDONR™221</u> 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 <u>Invitrogen</u>[™]. Recombination was facilitated through an *att*B substrate (*att*B-PCR product or a linearized *att*B expression clone) with an *att*P substrate (pDONR[™]221) to create an *att*L-containing entry clone. The entry clone contains recombinational cloning sites, *att*L1 and *att*L2 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[™] <u>Gateway[®] Technology Manual</u> for additional details.

Material Provided:

Each inoculated well of the 96-well plate contains approximately 60 μ L of *E. coli* culture (strain DH10B-T1) in Luria Bertani (LB) broth containing 50 μ g/mL kanamycin supplemented with 15% glycerol.

Packaging/Storage:

NR-19689 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 11, NR-19689."

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. <u>Biosafety in Microbiological and Biomedical Laboratories</u>. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

Disclaimers:

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References:

 Heidelberg, J. F., et al. "DNA Sequence of both Chromosomes of the Cholera Pathogen Vibrio cholera." <u>Nature</u> 406 (2000): 477-483. PubMed. 10952301.

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



Clone	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
200074	A02	N/A	VCA0391		killer protein, putative	N/A
200091	A03	N/A	VCA0358		hypothetical protein	N/A
199556	A04	277	VC1992	purU	formyltetrahydrofolate deformylase	NP_231626.1
199578	A05	337	VC1995	ansA	L-asparaginase I	NP_231629.1
199594	A06	404	VC1977		aspartate aminotransferase, putative	NP_231611.1
199607	A07	N/A	VCA0442		acetyltransferase, putative	N/A
198988	A08	259	VC1509	cobB	nicotinate mononucleotide:5,6-dimethylbenzimidazole phosphoribosyltransferase	NP_231150.1
199007	A09	N/A	VCA1058		transcriptional regulator, LysR family	N/A
199017	A10	310	VC2740		conserved hypothetical protein	NP_232366.1
199033	A11	N/A	VCA1046	mtlD	mannitol-1-phosphate 5-dehydrogenase	N/A
199053	A12	N/A	VCA1054		conserved hypothetical protein	N/A
200057	B01	N/A	VCA0371	orfB	transposase OrfAB, subunit B	N/A
200075	B02	N/A	VCA0374		hypothetical protein	N/A
200095	B03	N/A	VCA0400		acetyltransferase, putative	N/A
199557	B04	68	VC0351		conserved hypothetical protein	NP_230005.1
199580	B05	357	VC0318	murB	UDP-N-acetylenolpyruvoylglucosamine reductase	NP_229972.1
199596	B06	426	VC1921	clpX	ATP-dependent Clp protease, ATP-binding subunit ClpX	NP_231555.1
199609	B07	164	VC0330	rsd	regulator of sigma D	NP_229984.1
198990	B08	N/A	VCA1038		amino acid ABC transporter, permease protein	N/A
199008	B09	107	VC2745		hypothetical protein	NP_232371.1
199018	B10	140	VC1510		hypothetical protein	NP_231151.1
199034	B11	N/A	VCA1053		conserved hypothetical protein	N/A
199060	B12	221	VC2727	epsJ	general secretion pathway protein J	NP_232354.1
200063	C01	307	VC0267		D-isomer specific 2-hydroxyacid dehydrogenase family protein	NP_229923.1
200076	C02	352	VC0271		conserved hypothetical protein	NP_229927.1
199540	C03	232	VC1928		C4-dicarboxylate transport protein DctQ, putative	NP_231562.1
199558	C04	284	VC1986		conserved hypothetical protein	NP_231620.1
199581	C05	118	VC0357		conserved hypothetical protein	NP_230011.1
199597	C06	N/A	VCA0432		hypothetical protein	N/A
199611	C07	N/A	VCA0443	blc-4	lipoprotein Blc	N/A
198994	C08	N/A	VCA1039		amino acid ABC transporter, periplasmic amino acid- binding protein	N/A
199009	C09	301	VC2722	cysQ	cysQ protein	NP_232349.1
199020	C10	N/A	VCA1051		conserved hypothetical protein	N/A
199036	C11	184	VC2743		hypothetical protein	NP_232369.1
199061	C12	232	VC1524		ABC transporter, permease protein	NP_231164.1
200064	D01	N/A	VCA0392		antidote protein, putative	N/A
200078	D02	399	VC0269	manA-1	mannose-6-phosphate isomerase	NP_229925.1
199542	D03	233	VC1978		conserved hypothetical protein	NP_231612.1
199561	D04	N/A	VCA0426		conserved hypothetical protein	N/A
199583	D05	N/A	VCA0423		conserved hypothetical protein	N/A
199598	D06	429	VC0348	hflX	GTP-binding protein HfIX	NP_230002.1
199613	D07	N/A	VCA0436		acetyltransferase, putative	N/A

Table 1: Vibrio cholerae Gateway® Clones, Plate 11

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Product Information Sheet for NR-19689

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Clone ID	Well Position	ORF Length	Locus ID	Symbol	Product	Accession Number
198996	D08	286	VC1505		hypothetical protein	NP_231146.1
199010	D09	117	VC2728	epsl	general secretion pathway protein I	NP_232355.1
199024	D10	158	VC1518		hypothetical protein	NP_231158.1
199037	D11	395	VC1507	aroH	phospho-2-dehydro-3-deoxyheptonate aldolase, trp- sensitive	NP_231148.1
199063	D12	238	VC1495		hypothetical protein	NP_231136.1
200065	E01	N/A	VCA0366		hypothetical protein	N/A
200080	E01	412	VC0265		conserved hypothetical protein	NP_229921.1
199546	E03	237	VC1989		conserved hypothetical protein	NP 231623.1
199562	E04	285	VC0339	psd	phosphatidylserine decarboxylase	NP_229993.1
199587	E05	N/A	VCA0431	peu	hypothetical protein	N/A
199599	E06	145	VC1991		hypothetical protein	NP_231625.1
199621	E07	200	VC1980		conserved hypothetical protein	NP_231614.1
199000	E08	296	VC2736		conserved hypothetical protein	NP_232362.1
199012	E09	127	VC2735		conserved hypothetical protein	NP_232361.1
199025	E10	336	VC2726	epsK	general secretion pathway protein K	NP_232353.1
199040	E10	N/A	VCA1047	mtlR	mannitol operon repressor	N/A
199067	E12	N/A	VCA1037		amino acid ABC transporter, ATP-binding protein	N/A
200069	F01	331	VC0291		NifR3-Smm1 family protein	NP_229946.1
200082	F02	429	VC0275	purD	phosphoribosylamineglycine ligase	NP 229931.1
199552	F03	259	VC0354	fkpA	peptidyl-prolyl cis-trans isomerase, FKBP-type	NP_230008.1
199564	F04	N/A	VCA0441		hypothetical protein	N/A
199588	F05	382	VC0335		NlpD-related protein	NP_229989.1
199604	F06	441	VC1979	dgt	deoxyguanosinetriphosphate triphosphohydrolase	NP_231613.1
199625	F07	202	VC1987		outer membrane lipoprotein Slp, putative	NP_231621.1
199002	F08	297	VC2742	rbn	ribonuclease BN	NP_232368.1
199014	F09	N/A	VCA1042		Ccm2-related protein	N/A
199026	F10	158	VC1506		hypothetical protein	NP_231147.1
199044	F11	N/A	VCA1062	potE	putrescine-ornithine antiporter	N/A
199070	F12	N/A	VCA1052		conserved hypothetical protein	N/A
200071	G01	332	VC0289	gntR	gluconate utilization system gnt-I transcriptional repressor	NP_229944.1
200084	G02	445	VC0280		cadaverine-lysine antiporter CadB, putative	NP_229936.1
199553	G03	N/A	VCA0435		hypothetical protein	N/A
199565	G04	87	VC0347		host factor-I, putative	NP_230001.1
199590	G05	389	VC1912		conserved hypothetical protein	NP_231546.1
199605	G06	156	VC0360	rpsG	ribosomal protein S7	NP_230014.1
199627	G07	222	VC1981		hypothetical protein	NP_231615.1
199004	G08	298	VC1503		conserved hypothetical protein	NP_231144.1
199015	G09	N/A	VCA1050		hypothetical protein	N/A
199027	G10	337	VC1519	fdhD	formate dehydrogenase accessory protein	NP_231159.1
199046	G11	N/A	VCA1033		extracellular solute-binding protein, putative	N/A
199915	G12	69	VC1375		hypothetical protein	NP_231019.1
200072	H01	N/A	VCA0408		hypothetical protein	N/A
200089	H02	N/A	VCA0382		conserved hypothetical protein	N/A
199554	H03	269	VC0331	<u> </u>	MutT-nudix family protein	NP_229985.1
199567	H04	90	VC1919	hupB	DNA-binding protein HU-beta	NP_231553.1
199592	H05	395	VC0349	hflK	hflK protein	NP_230003.1
199606	H06	446	VC1926	dctD-1	C4-dicarboxylate transport transcriptional regulatory protein	NP_231560.1
199629	H07	229	VC1916	cmk	cytidylate kinase	NP_231550.1
199006	H08	N/A	VCA1035		hypothetical protein	N/A
199016	H09	N/A	VCA1032		hypothetical protein	N/A
199031	H10	357	VC1511	fdnl	formate dehydrogenase, cytochrome B556 subunit	NP_231152.1
199047	H11	202	VC1512	fdhB	formate dehydrogenase, iron-sulfur subunit	NP_231153.1

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