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SUPPORTING INFECTIOUS DISEASE RESEARCH

Vibrio cholerae Gateway[®] Clone Set, Recombinant in *Escherichia coli*, Plate 16

Catalog No. NR-19694

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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 InvitrogenTM. Recombination was facilitated through an *att*B substrate (*att*B-PCR product or a linearized *att*B expression clone) with an *att*P substrate (pDONRTM221) 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 InvitrogenTM Gateway[®] Technology Manual 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-19694 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 16, NR-19694."

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 cholerae.*" <u>Nature</u> 406 (2000): 477-483. PubMed: 10952301

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



| Clone ID | Well Position | ORF Length | Locus ID | Symbol | Product | Accession Number |
|-------------|------------------|---------------|----------|--------|---|---------------------|
| 198915 | A02 | 85 | VC2769 | atpE | ATP synthase F0, C subunit | NP_232395.1 |
| 198927 | A03 | N/A | VCA1096 | cheY-4 | chemotaxis protein CheY | N/A |
| 198938 | A04 | N/A | VCA1088 | | methyl-accepting chemotaxis protein | N/A |
| 198959 | A05 | 203 | VC1526 | mobA | molybdopterin-guanine dinucleotide biosynthesis protein | NP_231166.1 |
| 198968 | A06 | N/A | VCA1098 | | ABC transporter, periplasmic substrate-binding protein | N/A |
| 198984 | A07 | 631 | VC2775 | gidA | glucose inhibited division protein A | NP_232401.1 |
| 200483 | A08 | 319 | VC1611 | metA | homoserine O-succinyltransferase | NP_231251.1 |
| 200505 | A09 | 387 | VC1608 | | conserved hypothetical protein | NP_231248.1 |
| 200520 | A10 | 171 | VC0810 | | hypothetical protein | NP_230459.1 |
| 200531 | A11 | 464 | VC0003 | thdF | thiophene and furan oxidation protein ThdF | NP_062587.1 |
| 200542 | A12 | 225 | VC2461 | rnc | ribonuclease III | NP_232090.1 |
| 198904 | B01 | 282 | VC1542 | ligA-2 | DNA ligase | NP_231182.1 |
| 198916 | B02 | | VC2760 | | transcriptional regulator, LysR family | NP_232386.1 |
| 198928 | B03 | N/A | VCA1078 | | transcriptional regulator, LuxR family | N/A |
| 198939 | B04 | N/A | VCA0010 | | conserved hypothetical protein | N/A |
| 198960 | B05 | N/A | VCA1080 | | secretion protein, HlyD family | N/A |
| 198970 | B06 | 530 | VC1535 | | methyl-accepting chemotaxis protein | NP_231175.1 |
| 198985 | B07 | 252 | VC1548 | | hypothetical protein | NP_231188.1 |
| 200485 | B08 | 321 | VC2465 | rseB | sigma-E factor regulatory protein RseB | NP_232094.1 |
| 200511 | B09 | 408 | VC1609 | | conserved hypothetical protein | NP_231249.1 |
| 200521 | B10 | 440 | VC1284 | celF | 6-phospho-beta-glucosidase | NP_230929.1 |
| 200532 | B11 | 212 | VC2466 | rseA | sigma-E factor negative regulatory protein RseA | NP_232095.1 |
| 200543 | B12 | 541 | VC0819 | aldA-1 | aldehyde dehydrogenase | NP_230467.1 |
| 198905 | C01 | 53 | VC1530 | | hypothetical protein | NP_231170.1 |
| 198917 | C02 | 91 | VC1531 | | hypothetical protein | NP_231171.1 |
| 198929 | C03 | 126 | VC1528 | | hypothetical protein | NP_231168.1 |
| 198942 | C04 | 387 | VC2759 | fadA | fatty oxidation complex, beta subunit | NP_232385.1 |
| 198961 | C05 | N/A | VCA0003 | | hypothetical protein | N/A |
| 198986 | C07 | N/A | VCA0002 | | hypothetical protein | N/A |
| 200495 | C08 | 335 | VC1266 | | hypothetical protein | NP_230911.1 |
| 200512 | C09 | 156 | VC2464 | rseC | sigma-E factor regulatory protein RseC | NP_232093.1 |
| 200522 | C10 | 177 | VC1274 | | conserved hypothetical protein | NP_230919.1 |
| 200534 | C11 | 216 | VC1277 | | transcriptional regulator, LuxR family | NP_230922.1 |
| 198908 | D01 | 288 | VC2765 | atpG | ATP synthase F1, gamma subunit | NP_232391.1 |
| 198919 | D02 | N/A | VCA0005 | | hypothetical protein | N/A |
| 198930 | D03 | 327 | VC1541 | | hypothetical protein | NP_231181.1 |
| 198944 | D04 | 390 | VC1562 | | beta-lactamase-related protein | NP_231202.1 |
| 200469 | D07 | 238 | VC1271 | | hypothetical protein | NP_230916.1 |
| 200497 | D08 | 340 | VC1286 | | transcriptional regulator, Lacl family | NP_230931.1 |
| 200514 | D09 | 159 | VC1278 | | transcriptional regulator, MarR family | NP_230923.1 |
| 200523 | D10 | 446 | VC1282 | celB | PTS system, cellobiose-specific IIC component | NP_230927.1 |
| 200535 | D11 | 524 | VC1268 | | conserved hypothetical protein | NP_230913.1 |
| 200546 | D12 | 227 | VC0807 | | hypothetical protein | NP_230456.1 |

Table 1: Vibrio cholerae Gateway® Clones, Plate 16

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Product Information Sheet for NR-19694

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| Clone | Well | ORF | Locus ID | Symbol | Product | Accession |
|--------------|----------|--------|------------------|--------|--|----------------------------|
| ID 100010 | Position | Length | 1/04500 | - | | Number |
| 198910 | E01 | 228 | VC1539 | | conserved hypothetical protein | NP_231179.1 |
| 198920 | E02 | 304 | VC1534 | | conserved hypothetical protein | NP_231174.1 |
| 198932 | E03 | 344 | VC1557 | | transcriptional regulator, LacI family | NP_231197.1 |
| 198945 | E04 | 166 | VC1537 | nlpC | lipoprotein NIpC | NP_231177.1 |
| 198963 | E05 | 206 | VC1544 | tonB2 | tonB2 protein | NP_231184.1 |
| 200471 | E07 | 241 | VC2459 | recO | DNA repair protein RecO | NP_232088.1 |
| 200499 | E08 | 360 | VC1267 | | hypothetical protein | NP_230912.1 |
| 200515 | E09 | 427 | VC1275 | | conserved hypothetical protein | NP_230920.1 |
| 200524 | E10 | 182 | VC1619 | | hypothetical protein | NP_231259.1 |
| 200536 | E11 | 216 | VC1263 | ribA | GTP cyclohydrolase II | NP_230908.1 |
| 200548 | E12 | 228 | VC1604 | | response regulator | NP_231244.1 |
| 198911 | F01 | N/A | VCA1070 | | hypothetical protein | N/A |
| 198924 | F02 | N/A | VCA0007 | | 3-hydroxyisobutyrate dehydrogenase, putative | N/A |
| 198935 | F03 | 141 | VC1529 | | hypothetical protein | NP_231169.1 |
| 198954 | F04 | 457 | VC1547 | | biopolymer transport protein ExbB-related protein | NP_231187.1 |
| 198964 | F05 | N/A | VCA1101 | | ABC transporter, ATP-binding protein | N/A |
| 198981 | F06 | N/A | VCA0006 | | conserved hypothetical protein | N/A |
| 200475 | F07 | 245 | VC0008 | | amino acid ABC transporter, ATP-binding protein | NP_062592.1 |
| 200500 | F08 | 126 | VC2457 | acpS | holo-(acyl-carrier-protein) synthase | NP_232086.1 |
| 200516 | F09 | 159 | VC0814 | • | transcriptional regulator, putative | NP_230463.1 |
| 200526 | F10 | 182 | VC1269 | | conserved hypothetical protein | NP_230914.1 |
| 200537 | F11 | 538 | VC1605 | | sensor kinase citA, putative | NP_231245.1 |
| 198912 | G01 | 293 | VC1550 | ugpA | glycerol-3-phosphate ABC transporter, permease protein | NP_231190.1 |
| 198925 | G02 | 120 | VC1556 | | conserved hypothetical protein | NP_231196.1 |
| 198936 | G03 | N/A | VCA1075 | | hypothetical protein | N/A |
| 198957 | G04 | 180 | VC1546 | exbB2 | TonB system transport protein ExbB2 | NP_231186.1 |
| 198965 | G05 | 210 | VC2774 | gidB | glucose inhibited division protein B | NP 232400.1 |
| 200477 | G07 | 252 | VC1285 | | conserved hypothetical protein | NP_230930.1 |
| 200501 | G08 | 362 | VC1614 | | conserved hypothetical protein | NP 231254.1 |
| 200517 | G09 | 431 | VC1280 | | hypothetical protein | NP_230925.1 |
| 200527 | G10 | 456 | VC0815 | | hypothetical protein | NP_230464.1 |
| 200539 | G11 | 538 | VC0806 | | conserved hypothetical protein | NP_230455.1 |
| 200551 | G12 | 589 | VC0809 | | hypothetical protein | NP_230458.1 |
| 198914 | H01 | 293 | VC2772 | | ParB family protein | NP_232398.1 |
| 198926 | H02 | N/A | VCA1099 | | ABC transporter, permease protein | N/A |
| 198937 | H03 | 143 | VC1538 | | hypothetical protein | NP_231178.1 |
| 198958 | H04 | 478 | VC1558 | bglA | 6-phospho-beta-glucosidase | NP_231198.1 |
| 198966 | H05 | N/A | VCA1071 | putP | sodium-proline symporter | N/A |
| 198983 | H06 | 245 | VC1533 | put | conserved hypothetical protein | NP_231173.1 |
| 200481 | H07 | 245 | VC1617 | | transcriptional regulator, LysR family | NP_231257.1 |
| 200481 | H08 | 379 | VC1017 VC0821 | | hypothetical protein | NP_230469.1 |
| 200503 | H08 | 170 | VC0621 VC1615 | | conserved hypothetical protein | NP_231255.1 |
| 200518 | H10 | 192 | VC1615 VC0813 | | tellurite resistance protein-related protein | NP_231255.1 NP_230462.1 |
| | | | | | | |
| 200541 | H11 | 540 | VC1279 | | transporter, BCCT family | NP_230924 |