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SUPPORTING INFECTIOUS DISEASE RESEARCH

Yersinia pestis, Strain KIM, Gateway[®] Clone Set, Recombinant in *Escherichia coli*, Plate 19

Catalog No. NR-19615

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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:

The Yersinia pestis (Y. pestis), strain KIM, Gateway[®] clone set consists of 43 plates (plate 2 of this clone set has been discontinued) which contain more than 3600 sequence validated clones from Y. pestis, strain KIM cloned in *Escherichia coli* (*E. coli*) DH10B-T1 cells. Each open reading frame was constructed in vector <u>pDONRTM221</u> (InvitrogenTM) with an ATG start codon and a TAG stop codon. The sequence was validated by full length sequencing of each clone with greater than 1X coverage and a mutation rate of less than 0.2%. 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>TM. 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.

<u>Note:</u> 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 cannot confirm or validate any clone not identified on the plate information table.

Packaging/Storage:

NR-19615 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 $\mu\text{g/mL}$ 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 18 to 24 hours.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Yersinia pestis*, Strain KIM, Gateway[®] Clone Set, Recombinant in *Escherichia coli*, Plate 19, NR-19615."

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

Disclaimers:

You are authorized to use this product for research use only. It is not intended for human use.

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References:

- 1. Deng, W., et al. "Genome Sequence of *Yersinia pestis* KIM." <u>J. Bacteriol.</u> 184 (2002): 4601-4611. PubMed: 12142430.
- Lindler, L. E., et al. "Complete DNA Sequence and Detailed Analysis of the *Yersinia pestis* KIM5 Plasmid Encoding Murine Toxin and Capsular Antigen." <u>Infect.</u> <u>Immun.</u> 66 (1998): 5731-5742. PubMed: 9826348.

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Table 1: Yersinia pestis, Strain KIM, Gateway[®] Clone Set, Recombinant in *Escherichia coli*, Plate 19 (UYPVS)

Clone	Well	Locus ID	Description (Gene name)	ORF	Accession	Average
	Position			Length	Number	Depth of
						Coverage
37281	A01	NTL02YP1744	putative response regulator	616	AAM85043.1	5.03409091
37327	A02	NTL02YP3923	glycine decarboxylase	628	AAM86842.1	4.61624204
37467	A03	NTL02YP0650	hypothetical protein	679	AAM84154.1	5.32253314
37475	A04	NTL02YP0673	hypothetical protein	682	AAM84176.1	2.63489736
37492	A05	NTL02YP2839	putative regulator	685	AAM86435.1	2
37547	A06	NTL02YP0669	putative oxidoreductase	703	AAM84172.1	2.69274538
37670	A07	NTL02YP1758	putative kinase sensor protein	745	AAM85054.1	2.77852349
37849	A08	NTL02YP3912	putative oxidoreductase	808	AAM86835.1	4.63613861
37968	A10	NTL02YP1734	hypothetical protein	847	AAM85033.1	1.3175915
37970	A11	NTL02YP3890	hypothetical protein	847	AAM86812.1	8.6635183
37993	A12	NTL02YP2813	ABC transporter ATP-binding protein	856	AAM85922.1	2
38025	B01	NTL02YP2822	putative lipoprotein	865	AAM86418.1	7.1132948
38108	B02	NTL02YP3913	taurine ABC transport system inner	905		2 4706057
30100	DU2	NILUZIP3913	membrane permease protein	895	AAM87509.1	3.4726257
38144	B03	NTL02YP3914	taurine dioxygenase	907	AAM87510.1	2
35944	B04	NTL02YP2818	putative integral membrane protein	157	AAM85928.1	2
36048	B05	NTL02YP0632	hypothetical protein	202	AAM84140.1	-
36244	B06	NTL02YP0638	putative tellurium resistance protein	268	AAM84146.1	3.97761194
36274	B07	NTL02YP3908	hypothetical protein	277	AAM86831.1	2.70036101
38258	B08	NTL02YP3907	catabolic regulation response regulator	946	AAM86830.1	5.77484144
38312	B09	NTL02YP2823	putative DEOR-type transcriptional regulator	970	AAM85931.1	3.06907216
38311	B10	NTL02YP1729	hypothetical protein	970	AAM85028.1	3.61134021
38370	B11	NTL02YP1728	3-oxoacyl-[acyl-carrier-protein] synthase	991	AAM85324.1	5.9283552
38420	B12	NTL02YP2803	hypothetical protein	1006	AAM85912.1	2
38451	C01	NTL02YP3915	hypothetical protein	1021	AAM86836.1	4.20763957
38465	C02	NTL02YP2821	ATP-binding component of ATP transporter	1027	AAM86417.1	4.59493671
38584	C04	NTL02YP1722	hypothetical protein	1066	AAM85318.1	5.76360225
36487	C05	NTL02YP2809	outer membrane protein	355	AAM85918.1	4.74366197
36697	C06	NTL02YP0637	putative tellurium resistance protein	433	AAM84145.1	3.99076212
36759	C07	NTL02YP3905	inner membrane protein	448	AAM86828.1	3.97767857
36834	C08	NTL02YP3906	catabolite repression sensor kinase for PhoB	472	AAM86829.1	3.98305085

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Product Information Sheet for NR-19615

SUPPORTING INFECTIOUS DISEASE RESEARCH

Clone	Well Position	Locus ID	Description (Gene name)	ORF Length	Accession Number	Average Depth of Coverage
36828	C09	NTL02YP1717	hypothetical protein	472	AAM85019.1	5.29449153
36895	C10	NTL02YP0639	putative tellurium resistance protein	493	AAM84147.1	3.98377282
36901	C11	NTL02YP0641	hypothetical protein	496	AAM84149.1	5.24395161
36940	C12	NTL02YP2801	sodium-calcium/proton antiporter	505	AAM85910.1	4.68118812
38627	D01	NTL02YP3888	hypothetical protein	1081	AAM86810.1	2.84181314
38636	D02	NTL02YP1718	putative multimodular enzyme	1087	AAM85020.1	6.33854646
38742	D03	NTL02YP1735	putative transcriptional regulator	1132	AAM85035.1	5.37102473
38827	D04	NTL02YP2800	nitrate/nitrite sensor	1171	AAM85909.1	4.96242528
38831	D05	NTL02YP2804	hypothetical protein	1174	AAM85913.1	4.66950596
38934	D06	NTL02YP3895	permease (major facilitator superfamily)	1225	AAM87491.1	4.68734694
39002	D08	NTL02YP2806	hypothetical protein	1267	AAM85915.1	2.20599842
39007	D09	NTL02YP3902	protein disulfide isomerase II	1270	AAM86825.1	2.91968504
36985	D10	NTL02YP0628	hypothetical protein	517	AAM84136.1	5.56673114
37017	D11	NTL02YP2807	hypothetical protein	526	AAM85916.1	4.39543726
37126	D12	NTL02YP1724	putative membrane protein	565	AAM85025.1	6.18584071
37287	E01	NTL02YP3901	ssDNA exonuclease	616	AAM86824.1	5.60876623
37358	E02	NTL02YP3899	lysine tRNA synthetase	640	AAM86822.1	2
37379	E02	NTL02YP1723	hypothetical protein	649	AAM85024.1	2.55007704
37413	E04	NTL02YP0643	hypothetical protein	661	AAM84151.1	2.75189107
37457	E05	NTL02YP3904	flavodoxin 2	673	AAM86827.1	2.78008915
39028	E05	NTL02YP1732	putative membrane protein	1282	AAM85031.1	3.17550702
39117	E07	NTL02YP2819	putative transport protein	1339	AAM85929.1	1.42419716
39141	E07 E08	NTL02YP3894	hypothetical	1359	AAM86817.1	6.08068098
39141	E08 E09	NTL02YP3898	putative phage integrase	1354	AAM86821.1	5.23190547
39452	E09 E10	NTL02YP3900	peptide chain release factor RF-2	1582	AAM86823.1	2.517067
39541	E10 E11	NTL02YP0631	hypothetical protein	1714	AAM84139.1	4.59043174
39553	E11 E12	NTL02YP2812	putative thioredoxin-family protein	1714	AAM85921.1	6.70098323
39555	F02	NTL02YP2012 NTL02YP1736	sialic acid transporter	679	AAM85036.1	2
37470	F02 F03	NTL02YP1736	hypothetical protein	679	AAM84243.1	6.65243004
37509	F03	NTL02YP2825	L-asparagine permease	691	AAM85932.1	2.78147612
37560	F04 F05	NTL02YP2625	hypothetical protein	706	AAM84152.1	2.70147012
37578	F05 F06	NTL02YP2820		708		6.37921348
37578	F06 F07	NTL02YP2820	putative glutaminase putative ABC inner membrane permease	712	AAM85930.1 AAM85924.1	5.2008427
	F07 F08			763		
37719	F08 F09	NTL02YP2826	putative aminotransferase		AAM85933.1	6.1965924
37758	F09 F10	NTL02YP1730	putative acetylneuraminate lyase	775	AAM85029.1	6.49677419
37776		NTL02YP0642 NTL02YP0630	hypothetical protein	781 781	AAM84150.1	4.75928297 4.7631242
37775	F11		hypothetical protein		AAM84138.1	
38186 38302	F12 G01	NTL02YP0612 NTL02YP2798	multi modular putative transcriptional	922 967	AAM84122.1 AAM85908.1	5.23861171 6.13547053
20240	<u> </u>		regulator; also putative	070		6 20026500
38340	G02	NTL02YP1688	hypothetical protein	979	AAM84997.1	6.39836568
38347	G03	NTL02YP0621	hemin uptake system component	982	AAM84130.1	5.50203666
38448	G04	NTL02YP1703	hypothetical protein	1021	AAM85299.1	5.73163565
38510	G05	NTL02YP0610	putative type III secretion system component	1039	AAM84120.1	5.78344562
38530	G06	NTL02YP0626	orfX protein in hemin uptake locus	1045	AAM84134.1	2.72822967
36008	G07	NTL02YP0624	hypothetical protein	193	AAM84220.1	5.93782383
36307	G08	NTL02YP2795	hypothetical protein	292	AAM85906.1	3.9760274
36322	G09	NTL02YP1693	hypothetical protein	298	AAM85001.1	6.77852349
38533	G10	NTL02YP1698	putative enzyme	1045	AAM85007.1	3.24976077
38569	G11	NTL02YP2778	hypothetical protein	1060	AAM85893.1	3.30849057

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Product Information Sheet for NR-19615

SUPPORTING INFECTIOUS DISEASE RESEARCH

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38588	G12	NTL02YP2780	hypothetical protein	1066	AAM85895.1	3.53377111
38592	H01	NTL02YP0603	putative type III secretion system component	1069	AAM84113.1	6.04583723
38622	H02	NTL02YP1683	putative transposase	1081	AAM84992.1	2.97132285
38661	H03	NTL02YP1687	hypothetical protein	1096	AAM84996.1	2.99817518
38726	H04	NTL02YP1711	putative transport protein	1126	AAM85307.1	2.34369449
38740	H05	NTL02YP3882	hypothetical protein	1129	AAM86805.1	5.26660762
38794	H06	NTL02YP1714	cytochrome c-type protein	1156	AAM85017.1	5.76816609
36427	H07	NTL02YP0600	hypothetical protein	334	AAM84110.1	3.9760479
36460	H08	NTL02YP0622	TonB-dependent outer membrane receptor	346	AAM84131.1	4.62716763
36599	H09	NTL02YP0606	hypothetical protein	397	AAM84116.1	3.98992443
36699	H10	NTL02YP2794	putative aldehyde dehydrogenase	433	AAM85905.1	1.98383372
36696	H11	NTL02YP0607	putative type III secretion system component	433	AAM84117.1	4.84526559
36742	H12	NTL02YP1709	ferredoxin-type protein	445	AAM85014.1	5.16404494