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SUPPORTING INFECTIOUS DISEASE RESEARCH

Yersinia pestis, Strain KIM, Gateway[®] Clone Set, Recombinant in *Escherichia coli*, Plate 23

Catalog No. NR-19619

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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-19619 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 μ 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 23, NR-19619."

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:

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References:

- 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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Clone	Well Position	Locus ID ¹	Description (Gene name)	ORF Length	Accession Number	Average Depth of Coverage
37632	A01	NTL02YP2738	cytidylate kinase	693	AAM86334.1	4.77353
37641	A02	NTL02YP3821	hypothetical protein	696	AAM87417.1	3.99049
37716	A03	NTL02YP1658	hypothetical protein	723	AAM85254.1	2.80603
37715	A04	NTL02YP1646	ATP-binding component of glutamine high-affinity glutamine ABC	723	AAM85242.1	6.37615
37771	A05	NTL02YP3820	transcriptional regulatory protein	738	AAM87416.1	5.99229
37786	A06	NTL02YP1648	glutamine-binding periplasmic protein for high-affinity glutamine ABC	744	AAM85244.1	4.61224
37783	A07	NTL02YP0555	hypothetical protein	784	AAM84151.1	6.66709
37811	A08	NTL02YP2728	CTP:CMP-3-deoxy-D-manno- octulosonate transferase	753	AAM86324.1	6.69609
37875	A09	NTL02YP1651	putative enzyme	777	AAM85247.1	1.99633
38368	A10	NTL02YP1624	hypothetical protein	951	AAM85220.1	6.28254
38596	A12	NTL02YP2700	3-oxoacyl-[acyl-carrier-protein] synthase II	1029	AAM86296.1	3.37418
36016	B01	NTL02YP2711	hypothetical protein	153	AAM86307.1	-
36029	B02	NTL02YP3803	hypothetical protein	156	AAM87399.1	5.89796
36073	B03	NTL02YP2705	hypothetical protein	168	AAM86301.1	4.96154
36120	B04	NTL02YP1628	hypothetical protein	183	AAM85224.1	2
36141	B05	NTL02YP2725	hypothetical protein	189	AAM86321.1	-
36164	B06	NTL02YP3789	hypothetical protein	198	AAM87386.1	4.37395
36245	B07	NTL02YP1617	hypothetical protein	228	AAM85213.1	5.77239
36269	B08	NTL02YP1621	hypothetical protein	237	AAM85217.1	5.787
38708	B09	NTL02YP0543	hypothetical protein	1080	AAM84139.1	3.49732
38915	B10	NTL02YP0530	cystathionine beta-lyase (beta- cystathionase)	1182	AAM84126.1	5.68494
38973	B11	NTL02YP2716	aspartate aminotransferase	1206	AAM86312.1	3.2801
36454	B12	NTL02YP1612	NADH dehydrogenase I chain K	303	AAM85208.1	5.77551
36551	C01	NTL02YP3788	hypothetical protein	336	AAM87384.1	5.77128
36622	C02	NTL02YP3809	hypothetical protein	363	AAM87405.1	5.49628
36884	C03	NTL02YP3808	universal stress protein	447	AAM87404.1	5.79055
37024	C04	NTL02YP0541	hypothetical protein	489	AAM84137.1	4.90926
37052	C05	NTI 02YP3796	hypothetical protein	498	AAM87392 1	2

 Table 1: Yersinia pestis, Strain KIM, Gateway[®] Clone Set, Recombinant in Escherichia coli, Plate 23 (UYPVW)

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Product Information Sheet for NR-19619

SUPPORTING INFECTIOUS DISEASE RESEARCH

Clone	Well Position	Locus ID ¹	Description (Gene name)	ORF Length	Accession Number	Average Depth of Coverage
37077	C06	NTL02YP3797	hypothetical protein	507	AAM87393.1	2
37105	C08	NTL02YP1626	hypothetical protein	516	AAM85222.1	5.78777
37157	C09	NTL02YP1619	gluconokinase 1	537	AAM85215.1	5.58579
39038	C10	NTL02YP2713	nicotinate phosphoribosyltransferase	1251	AAM86309.1	3.27653
39063	C11	NTL02YP3799	conserved hypothetical protein	1269	AAM87395.1	3.92131
39142	C12	NTL02YP0539	hypothetical protein	1314	AAM84135.1	3.85746
39167	D01	NTL02YP2723	mukF protein (killing factor KICB)	1323	AAM86319.1	3.16654
39189	D02	NTL02YP3806	putative secreted metalloprotease	1338	AAM87402.1	3.89042
39205	D03	NTL02YP3807	NADP-specific glutamate dehydrogenase	1347	AAM87403.1	3.81471
37173	D04	NTL02YP1610	NADH dehydrogenase I chain I	543	AAM85206.1	2
37185	D05	NTL02YP0538	orfX protein in hemin uptake locus	546	AAM84134.1	2.74573
37192	D06	NTL02YP1611	NADH dehydrogenase I chain J	546	AAM85207.1	7
37202	D07	NTL02YP2718	hypothetical protein	549	AAM86314.1	2.73005
37210	D08	NTL02YP2709	hypothetical protein	552	AAM86305.1	2.80574
37290	D09	NTL02YP2702	hypothetical protein	579	AAM86298.1	6.66882
37439	D10	NTL02YP0547	component in transcription antitermination	667	AAM84068.1	2.81109
37486	D11	NTL02YP1631	hypothetical protein	645	AAM85227.1	2.76788
37503	D12	NTL02YP2717	hypothetical protein	648	AAM86313.1	2.75872
37507	E01	NTL02YP0537	orfY protein in hemin uptake locus	651	AAM84133.1	7.589
37611	E02	NTL02YP0531	ATP-binding protein of ABC transporter	687	AAM84127.1	2
37729	E03	NTL02YP1618	putative oxidoreductase	726	AAM85214.1	5.15274
39268	E04	NTL02YP3801	glutathione oxidoreductase	1383	AAM87397.1	3.31342
39370	E05	NTL02YP1615	NADH dehydrogenase I chain N	1464	AAM85211.1	2.9262
39443	E06	NTL02YP1614	NADH dehydrogenase I chain M	1533	AAM85210.1	3.74698
39547	E07	NTL02YP2703	paraquat-inducible protein B	1680	AAM86299.1	3.56221
37816	E08	NTL02YP2722	hypothetical protein	756	AAM86318.1	6.75879
37861	E09	NTL02YP3805	hypothetical protein	771	AAM87401.1	9.37731
37921	E11	NTL02YP2724	S-adenosylmethionine- dependent methyltransferase	792	AAM86320.1	6.09615
37945	E12	NTL02YP3792	CDP-diacylglycerol phosphotidylhydrolase	798	AAM87388.1	3.63842
37946	F01	NTL02YP0540	hypothetical protein	801	AAM84136.1	3.08086
38061	F02	NTL02YP0533	periplasmic binding protein	840	AAM84129.1	3.59318
38085	F03	NTL02YP3790	hypothetical protein	846	AAM87385.1	4.27991
38111	F04	NTL02YP0544	hypothetical protein	858	AAM84140.1	3.63252
37651	F05	NTL02YP0493	hypothetical protein	702	AAM84089.1	6.96765
37672	F06	NTL02YP3782	hypothetical protein	705	AAM87378.1	2.63758
37678	F07	NTL02YP0522	putative type III secretion system component	711	AAM84118.1	2.80559
37695	F08	NTL02YP1582	histidine ABC transporter, inner membrane permease	717	AAM85178.1	2
37774	F09	NTL02YP0490	endonuclease V (deoxyinosine 3endoduclease)	741	AAM84086.1	6.85275
37899	F11	NTL02YP0488	hypothetical protein	786	AAM84083.1	6.49516
37926	F12	NTL02YP0524	putative type III secretion system component	795	AAM84120.1	7.44551

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Clone	Well Position	Locus ID ¹	Description (Gene name)	ORF Length	Accession Number	Average Depth of
						Coverage
37938	G01	NTL02YP1583	ATP-binding component of histidine ABC transport	798	AAM85179.1	2
37936	G02	NTL02YP0486	hypothetical protein	798	AAM84082.1	5.84845
36149	G03	NTL02YP0513	hypothetical protein	195	AAM84109.1	-
36211	G04	NTL02YP1590	hypothetical protein	213	AAM85186.1	4.74308
36221	G05	NTL02YP0511	putative type III secretion system component	219	AAM84107.1	2.95753
36231	G06	NTL02YP0509	putative type III secretion system component	222	AAM84105.1	4.72137
36295	G07	NTL02YP0512	hypothetical protein	249	AAM84108.1	4.87543
36379	G08	NTL02YP0492	DNA-binding protein HU-alpha (HU-2)	276	AAM84088.1	2
36393	G09	NTL02YP0523	putative type III secretion system component	282	AAM84119.1	1.98758
37972	G10	NTL02YP0497	putative transposase	810	AAM84093.1	2
38274	G11	NTL02YP1584	putative sugar nucleotide epimerase	915	AAM85180.1	5.22094
38290	G12	NTL02YP3780	putative endoglucanase	921	AAM87376.1	5.24246
38320	H01	NTL02YP1602	NADH dehydrogenase transcriptional regulator, LysR family	933	AAM85198.1	5.98458
38357	H02	NTL02YP3787	ketodeoxygluconokinase	945	AAM87383.1	6.03756
38406	H03	NTL02YP0521	putative type III secretion system component	963	AAM84117.1	5.37488
38446	H04	NTL02YP1609	NADH dehydrogenase I chain H	978	AAM85205.1	6.03438
38488	H05	NTL02YP0500	hypothetical protein	993	AAM84096.1	3.55082
38590	H07	NTL02YP3778	putative ATP-binding component of dipeptide ABC transport	1026	AAM87374.1	5.23171
38626	H08	NTL02YP3779	putative ATP-binding component of dipeptide ABC trans	1041	AAM87375.1	3.605
38660	H09	NTL02YP1580	histidine-binding periplasmic protein of high-affinity histidine transport	1056	AAM85176.1	5.78467
36404	H10	NTL02YP2680	hypothetical protein	285	AAM86276.1	2
36478	H11	NTL02YP0502	hypothetical protein	312	AAM84098.1	5.9375
36530	H12	NTL02YP2679	putative sulfite reductase	330	AAM86275.1	2

¹The locus ID numbers given in this table were obtained from JCVI at the time of deposition. Please note that the locus tags have since been updated by JCVI and the corrected locus ID numbers can be obtained from the <u>JCVI-CMR</u> webpage using the locus search option and inputting the above locus IDs.