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SUPPORTING INFECTIOUS DISEASE RESEARCH

Staphylococcus aureus (MRSA), Strain COL Gateway[®] Clone Set, Recombinant in *Escherichia coli*, Plate 17

Catalog No. NR-19513

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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 methicillin-resistant *Staphylococcus aureus* (*S. aureus*), strain COL Gateway[®] clone set consists of 25 plates which contain 2343 sequence validated clones from *S. aureus* strain COL cloned in *Escherichia coli* (*E. coli*) DH10B-T1 cells. Each open reading frame was constructed in vector <u>pDONRTM221</u> (<u>InvitrogenTM</u>) with a native start codon and no 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 <u>Gateway[®] Technology Manual</u> for additional details.

Material Provided:

Every 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-19513 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: *Staphylococcus aureus* (MRSA), Strain COL Gateway[®] Clone Set, Recombinant in *Escherichia coli*, Plate 17, NR-19513."

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:

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

Use of this product is subject to the terms and conditions of the BEI Resources Material Transfer Agreement (MTA). The MTA is available on our Web site at <u>www.beiresources.org</u>.

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References:

 Gill, S. R., et al. "Insights on Evolution of Virulence and Resistance from the Complete Genome Analysis of an Early Methicillin-Resistant *Staphylococcus aureus* Strain and a Biofilm-Producing Methicillin-Resistant *Staphylococcus epidermidis* Strain." <u>J. Bacteriol.</u> 187 (2005): 2426-2438. PubMed: 15774886.

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



Table 1: Staphylococcus aureus, Strain COL Gateway® Clones, Plate 17 (ZSAJQ)¹

Clone	Well	ORF	Locus ID	Description (Gene name)	Accession	Average Depth
	Position	Length			Number	of Coverage
3343	A01	955	SACOL1548	ribonuclease Z	YP_186389.1	5.106806283
3345	A02	955	SACOL1628	PhoH family protein	YP 186468.1	5.036649215
3347	A03	955	SACOL1731	primosomal protein Dnal	YP 186568.1	4.210471204
3349	A04	958	SACOL0013	conserved hypothetical protein	YP 184924.1	4.221294363
3351	A05	958	SACOL0182	hypothetical protein	YP 185081.1	5.086638831
3353	A06	958	SACOL0309	conserved hypothetical protein	YP 185201.1	5.057411273
3355	A07	958	SACOL0715	conserved hypothetical protein	YP_185597.1	5.04697286
3357	A08	958	SACOL0801	UDP-N-acetylenolpyruvoylglucosamine reductase	YP_185675.1	5.001043841
3359	A09	958	SACOL1668	peptidase, U32 family	YP_186508.1	4.659707724
3365	A12	961	SACOL0526	DNA polymerase III, delta prime subunit, putative	YP_185414.1	4.237252862
3367	B01	961	SACOL0747	cobalamin synthesis protein/P47K family protein	YP_185626.1	5.092611863
3370	B02	961	SACOL1130	glycerophosphoryl diester phosphodiesterase, putative	YP_185994.1	5.182101977
3371	B03	961	SACOL1244	malonyl CoA-acyl carrier protein transacylase	YP_186104.1	4.32049948
3373	B04	961	SACOL1423	phosphate ABC transporter, permease protein	YP_186275.1	5.089490114
3375	B05	961	SACOL1717	porphobilinogen deaminase	YP_186555.1	5.097814776
3377	B06	964	SACOL0688	ABC transporter, substrate-binding protein	YP_185570.1	5.044605809
3379	B07	964	SACOL0962	glycerophosphoryl diester phosphodiesterase GlpQ, putative	YP_185831.1	5.039419087
3381	B08	964	SACOL1533	lipoprotein, putative	YP_186375.1	4.985477178
3383	B09	964	SACOL1723	conserved hypothetical protein	YP_186561.1	4.411825726
3385	B10	964	SACOL1982	manganese-dependent inorganic pyrophosphatase	YP_186806.1	5.02593361
3390	B12	967	SACOL0067	conserved hypothetical protein	YP_184972.1	4.613236815
3391	C01	967	SACOL0557	cysteine synthase	YP_185445.1	5.087900724
3393	C02	967	SACOL0825	HPr kinase/phosphatase	YP_185699.1	5.003102378
3395	C03	967	SACOL1182	carbamate kinase	YP_186045.1	4.778697001
3398	C04	967	SACOL2152	conserved hypothetical protein	YP_186965.1	5.127197518
3399	C05	967	SACOL2184	tagatose-6-phosphate kinase	YP_186995.1	5.06204757
3401	C06	970	SACOL0225	inosine-uridine preferring nucleoside hydrolase	YP_185123.1	5.022680412
3403	C07	970	SACOL0829	thioredoxin-disulfide reductase	YP_185703.1	5.024742268
3405	C08	970	SACOL1192	S-adenosyl-methyltransferase MraW	YP_186054.1	5.015463918
3407	C09	970	SACOL1228	methionyl-tRNA formyltransferase	YP_186091.1	4.919587629
3409	C10	970	SACOL1323	tRNA delta(2)-isopentenylpyrophosphate transferase	YP_186178.1	4.977319588
3413	C12	970	SACOL2327	formiminoglutamase	YP_187134.1	5.046391753
3415	D01	970	SACOL2448	2-dehydropantoate 2-reductase, putative	YP_187247.1	5.053608247
3417	D02	970	SACOL2475	peptide ABC transporter, permease protein, putative	YP_187272.1	5.04742268
3419	D03	973	SACOL0440	Abi-like protein	YP_185331.1	4.74717369
3421	D04	973	SACOL0655	oxidoreductase, aldo/keto reductase family	YP_185540.1	5.021582734

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Clone	Well Position	ORF Length	Locus ID	Description (Gene name)	Accession Number	Average Depth of Coverage
3423	D05	973	SACOL1049	1,4-dihydroxy-2-naphthoate octaprenyltransferase	YP_185914.1	5.014388489
3425	D06	973	SACOL1150	ribonuclease HIII	YP_186013.1	5.046248715
3427	D07	973	SACOL1530	lipoprotein, putative	YP_186372.1	5.059609455
3429	D08	973	SACOL1635	ribosomal protein L11 methyltransferase	YP_186475.1	4.976361768
3432	D09	973	SACOL2135	mannose-6-phosphate isomerase, class I	YP_186950.1	4.353545735
3433	D10	973	SACOL2664	mannose-6-phosphate isomerase, class I	YP_187452.1	4.391572456
3435	D11	976	SACOL0987	3-oxoacyl-(acyl-carrier-protein) synthase III	YP_185855.1	4.967213115
3438	D12	976	SACOL0994	oligopeptide ABC transporter, ATP-binding protein	YP_185862.1	5.088114754
3439	E01	976	SACOL1751	DHH subfamily 1 protein	YP_186587.1	5.024590164
3441	E02	976	SACOL1898	cmp-binding-factor 1	YP 186724.1	5.047131148
3443	E03	976	SACOL2276	inosine-uridine preferring nucleoside hydrolase	YP_187083.1	4.994877049
3445	E04	976	SACOL2451	amino acid ABC transporter, amino acid- binding protein	YP_187250.1	4.983606557
3451	E05	979	SACOL0832	conserved hypothetical protein	YP_185706.1	5.009193054
3453	E06	979	SACOL1747	acetyl-CoA carboxylase, carboxyl transferase, alpha subunit	YP_186583.1	4.072522983
3455	E07	979	SACOL2213	DNA-directed RNA polymerase, alpha subunit	YP 187023.1	4.879468846
3457	E08	979	SACOL2375	transporter, CorA family	YP 187179.1	4.995914198
3459	E09	979	SACOLA0001	replication initiation protein	_	5
3461	E10	982	SACOL0100	cysteine synthase/cystathionine beta-synthase family protein	YP_185005.1	5.018329939
3465	E12	982	SACOL1761	conserved hypothetical protein	YP 186595.1	3.263747454
3467	F01	982	SACOL1958	conserved hypothetical protein TIGR00147	YP 186783.1	5.00101833
3470	F02	982	SACOL2342	magnesium and cobalt transport protein CorA, putative	YP_187148.1	5.151731161
3473	F04	985	SACOL0666	iron compound ABC transporter, permease protein	YP_185550.1	4.972588832
3475	F05	985	SACOL1920	D-isomer specific 2-hydroxyacid dehydrogenase family protein	YP_186745.1	4.968527919
3477	F06	985	SACOL2030	sucrose operon repressor	YP_186848.1	4.841624365
3479	F07	988	SACOL0222	L-lactate dehydrogenase	YP_185120.1	4.893724696
3481	F08	988	SACOL1225	lipoprotein, putative	YP_186088.1	4.204453441
3483	F09	988	SACOL1524	conserved hypothetical protein	YP 186367.1	5.003036437
3485	F10	988	SACOL1810	conserved hypothetical protein TIGR01212	YP_186643.1	5.037449393
3487	F11	988	SACOL2296	D-isomer specific 2-hydroxyacid dehydrogenase family protein	YP_187103.1	5.020242915
3489	F12	988	SACOL2302	transcriptional regulator, putative	YP_187109.1	4.245951417
3494	G01	991	SACOL0455	conserved hypothetical protein	YP_185345.1	5.120080727
3495	G02	991	SACOL0797	iron compound ABC transporter, permease protein	YP_185671.1	4.988900101
3498	G03	991	SACOL1462	thymidylate synthase	YP_186311.1	5.143289606
3499	G04	991	SACOL2713	rhodanese-like domain protein	YP 187499.1	4.98284561
3501	G05	994	SACOL0155	cation efflux family protein	YP 185055.1	4.932595573
3503	G06	994	SACOL0932	D-isomer specific 2-hydroxyacid dehydrogenase family protein	YP_185802.1	4.870221328
3505	G07	994	SACOL1045	iron compound ABC transporter, iron compound-binding protein	YP_185910.1	4.973843058
3509	G09	994	SACOL1510	polyprenyl synthetase	YP_186354.1	5.021126761
3511	G10	994	SACOL2028	fructokinase, putative	YP 186846.1	4.838028169
3513	G11	994	SACOL2618	L-lactate dehydrogenase	YP_187407.1	5.017102616
3515	G12	994	SACOL2672	accessory secretory protein Asp3, putative	YP_187460.1	5.019114688
3517	H01	997	SACOL0999	oligopeptide ABC transporter, permease protein	YP_185867.1	5.006018054

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Clone	Well	ORF	Locus ID	Description (Gene name)	Accession	Average Depth
	Position	Length			Number	of Coverage
3519	H02	997	SACOL1897	protein export protein PrsA, putative	YP_186723.1	4.974924774
3521	H03	997	SACOL2548	conserved hypothetical protein	YP_187340.1	4.977933801
3523	H04	1000	SACOL0544	ribose-phosphate pyrophosphokinase	YP_185432.1	4.97
3525	H05	1000	SACOL0599	conserved hypothetical protein	YP_185485.1	4.795
3528	H06	1000	SACOL0707	dihydroxyacetone kinase family protein	YP_185589.1	4.584
3529	H07	1000	SACOL1144	iron compound ABC transporter, permease protein, putative	YP_186007.1	4.954
3531	H08	1000	SACOL1195	phospho-N-acetylmuramoyl-pentapeptide- transferase	YP_186057.1	4.237
3533	H09	1003	SACOL0198	conserved hypothetical protein	YP_185097.1	5.004985045
3535	H10	1003	SACOL0203	iron compound ABC transporter, iron compound-binding protein, putative	YP_185102.1	4.157527418
3537	H11	1003	SACOL0263	peptidoglycan hydrolase	YP_185158.1	4.058823529
3539	H12	1003	SACOL1519	L-asparaginase	YP_186362.1	4.808574277

¹25 clones in the *Staphylococcus aureus* (MRSA), Strain COL Gateway[®] Clone Set (Plates 1-25), Recombinant in *Escherichia coli*, have been physically removed from the clone set due to international distribution limitations set by U.S. Department of Commerce restrictions (Commerce Control List).