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SUPPORTING INFECTIOUS DISEASE RESEARCH

Staphylococcus aureus (MRSA), Strain COL Gateway[®] Clone Set, Recombinant in *Escherichia coli*, Plate 2

Catalog No. NR-19498

This reagent is the tangible property of the U.S. Government.

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 InvitrogenTM. Recombination was facilitated through an *attB* substrate (*attB*-PCR product or a linearized *attB* expression clone) with an *attP* substrate (pDONRTM221) to create an *attL*-containing entry clone. The entry clone contains recombinational cloning sites, *attL*1 and *attL*2 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[®]</u> <u>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-19498 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.
- 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 2, NR-19498."

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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its derivatives must contact the contributor to determine if a license is required. U.S. Government contractors may need a license before first commercial sale.

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." J. Bacteriol. 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 2 (ZSAJB)

Clone	Well	ORF	Locus ID	Description (Gene name)	Accession	Average Depth
	Position	Length			Number	of Coverage
229	A01	160	SACOL2604	hypothetical protein	N/A	3
232	A02	163	SACOL0056	hypothetical protein	N/A	1.969325153
233	A03	163	SACOL0493	hypothetical protein	N/A	2
235	A04	163	SACOL1684	hypothetical protein	N/A	2
238	A05	163	SACOL2351	hypothetical protein	YP_187157.1	2
241	A06	166	SACOL1046	hypothetical protein	YP_185911.1	-
246	A07	166	SACOL1469	hypothetical protein	YP_186317.1	2
249	A08	166	SACOL2247	hypothetical protein	YP_187056.1	2.759036145
252	A09	166	SACOL2480	hypothetical protein	YP_187277.1	1.915662651
253	A10	169	SACOL0231	hypothetical protein	YP_185127.1	2
256	A11	169	SACOL0965	hypothetical protein	YP_185834.1	-
257	A12	169	SACOL1087	hypothetical protein	YP_185951.1	2
259	B01	169	SACOL1186	antibacterial protein (phenol soluble modulin)	YP_186048.1	3
261	B02	169	SACOL1187	antibacterial protein (phenol soluble modulin)	YP_186049.1	3
263	B03	169	SACOL1344	hypothetical protein	YP_186197.1	2
265	B04	169	SACOL1863	hypothetical protein	YP_186691.1	-
267	B05		SACOL2022	CLONE IS NOT AVAILABLE ¹	YP_186841.1	
269	B06	169	SACOL2558	hypothetical protein	N/A	2
271	B07	172	SACOL0471	hypothetical protein	YP 185361.1	1.843023256
273	B08	172	SACOL0919	hypothetical protein	YP 185790.1	-
275	B09	172	SACOL1343	hypothetical protein	 N/A	-
277	B10	172	SACOL2076	conserved hypothetical protein	YP 186892.1	3
280	B11	172	SACOL2740	ribosomal protein L34	YP 187526.1	1.970930233
281	B12	175	SACOL0210	hypothetical protein	YP 185109.1	-
283	C01	175	SACOL1766	hypothetical protein	YP_186600.1	3
285	C02	175	SACOL2024	accessory gene regulator protein D	YP_186843.1	2
287	C03	175	SACOL2455	hypothetical protein	N/A	2
290	C04	175	SACOL2490	hypothetical protein	N/A	1.931428571
291	C05	178	SACOL0492	hypothetical protein	N/A	2
293	C06	178	SACOL1755	hypothetical protein	N/A	2
295	C07	178	SACOL1878	lantibiotic epidermin precursor EpiA	YP_186705.1	-
297	C08	178	SACOL2005	hypothetical protein	N/A	2
301	C09	178	SACOL2429	hypothetical protein	YP_187231.1	3
303	C10	181	SACOL0234	conserved hypothetical protein	YP_185130.1	2
307	C11	181	SACOL0850	hypothetical protein	YP_185724.1	2
309	C12	181	SACOL0867	hypothetical protein	N/A	2
311	D01	181	SACOL0893	pathogenicity island protein	YP_185764.1	3
313	D02	181	SACOL1174	hypothetical protein	YP_186037.1	3
315	D03	181	SACOL1679	conserved hypothetical protein	YP_186519.1	3
317	D04	181	SACOL1959	hypothetical protein	N/A	2
325	D06	187	SACOL0324	hypothetical protein	YP_185216.1	2
327	D07	187	SACOL0361	conserved hypothetical protein	YP_185253.1	3
329	D08	187	SACOL0465	hypothetical protein	YP_185355.1	2
331	D09	187	SACOL0909	hypothetical protein	YP_185780.1	3

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Product Information Sheet for NR-19498

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Clone	Well	ORF	Locus ID	Description (Gene name)	Accession	Average Depth
	Position	Length			Number	of Coverage
333	D10	187	SACOL0934	conserved hypothetical protein	YP 185804.1	-
335	D11	187	SACOL1911	conserved hypothetical protein	YP 186736.1	2
337	D12	190	SACOL0532	hypothetical protein	YP 185420.1	3
339	E01	190	SACOL0795	hypothetical protein	YP 185668.1	2
345	E02	193	SACOL0378	hypothetical protein	YP 185270.1	2
347	E03	196	SACOL0334	conserved hypothetical protein	YP 185226.1	2
351	E04	196	SACOL1318	hypothetical protein	N/A	3
353	E05	196	SACOL1934	conserved hypothetical protein	YP 186759.1	2
357	E06	199	SACOL0386	conserved hypothetical protein TIGR01669	YP 185278.1	-
359	E07	199	SACOL1487	hypothetical protein		1.924623116
361	E08	199	SACOL2250	conserved hypothetical protein	YP 187058.1	2
363	F09	202	SACOI 0029	HMG-CoA synthase, truncation	YP 184940.1	-
366	E10	202	SACOL 0625	conserved hypothetical protein	YP 185510.1	1 975247525
367	F11	202	SACOI 1119	conserved hypothetical protein	YP 185983 1	2
369	E12	202	SACOI 1394	bypothetical protein	YP 186246 1	3
371	E12	202	SACOL 2568	hypothetical protein	YP 187360 1	2
373	F02	205	SACOL 2059	conserved hypothetical protein	YP 186875 1	2
375	F03	205	SACOL 2259	hypothetical protein	YP 187066 1	2
377	F04	205	SACOL 2457	hypothetical protein	YP 187255 1	2
379	F05	205	SACOL 2542	conserved hypothetical protein	YP 187334 1	2
381	F06	200		bypothetical protein	VP 185118 1	3
383	F07	200	SACOL0213	hypothetical protein	<u>Ν/Δ</u>	1 047115385
385	F08	200	SACOL0504	hypothetical protein	VD 195/197 1	2
200	F00	200	SACOL0001		VD 196706 1	2
309	F09	200	SACOL 1972		TF_100790.1	<u> </u>
391		200	SACOL 1963		<u>TP_100007.1</u>	2
393	F11	208	SACOL 1987	conserved hypothetical protein	<u>YP_180811.1</u>	3
395		211	SACOL 1035		TP_100900.1	4 007000704
400	GUI	211	SACUL1853	nypothetical protein	<u>YP_180083.1</u>	1.08/203/91
402	GUZ	211	SACOL 1996		TP_100022.1	1.970303310
403	G03	214	SACOL0258	nypotnetical protein	<u>YP_185153.1</u>	3
407	G04	214	SACOL1556	nypotnetical protein	YP_186397.1	3
409	G05	214	SACOL2221	ribosomal protein L30p/L7e	<u>YP_187031.1</u>	2
411	G06	217	SACOL0581		YP_185467.1	3
413	G07	217	SACOL1680	conserved hypothetical protein	YP_186520.1	2
416	G08	220	SACOL0348	conserved hypothetical protein	YP_185240.1	-
417	G09	220	SACOL0986	hypothetical protein	YP_185854.1	2
419	G10	220	SACOL1171	hypothetical protein	YP_186034.1	2
421	G11	220	SACOL1335	hypothetical protein	YP_186189.1	2
425	G12	220	SACOL1852	hypothetical protein	N/A	3
427	H01	220	SACOL2226	ribosomal protein S14	YP_187036.1	-
429	H02	220	SACOL2311	hypothetical protein	YP_187118.1	3
432	H03	223	SACOL1185	hypothetical protein	YP_186047.1	2
435	H04	226	SACOL0331	hypothetical protein	YP_185223.1	2
437	H05	226	SACOL0436	conserved hypothetical protein	YP_185327.1	2
440	H06	226	SACOL1117	conserved hypothetical protein	YP_185981.1	-
441	H07	226	SACOL1348	conserved hypothetical protein	YP_186201.1	2
443	H08	226	SACOL2312	conserved hypothetical protein	YP_187119.1	3
445	H09	226	SACOL2491	conserved hypothetical protein	YP_187286.1	2
447	H10	226	SACOL2571	conserved hypothetical protein	YP_187363.1	2
449	H11	226	SACOL2726	hypothetical protein	YP_187512.1	2
451	H12	226	SACOL2730	hypothetical protein	YP_187516.1	3

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).

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