Salmonella enterica subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate 003/004_Cm

Catalog No. NR-29411

For research use only. Not for human use.

Contributor:

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Manufacturer:

BEI Resources

Product Description:

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 does not confirm or validate individual mutants provided by the contributor.

The *Salmonella enterica* (*S. enterica*) subsp. *enterica*, strain 14028s (serovar Typhimurium) targeted single-gene deletion (SGD) mutant library contains a total of 3,773 individual genes deleted simultaneously across two collections of mutants differentiated by kanamycin or chloramphenicol resistance.[1,2] The chloramphenicol-resistant mutant collection contains 3,376 mutants distributed among eleven 96-well plates. In these mutants, a single gene is replaced by a cassette conferring the chloramphenicol resistance gene, and includes 4 double mutants that contain both kanamycin and chloramphenicol cassettes. Deletions were confirmed by the depositor.[1,2] The parent strain *S. enterica* subsp. *enterica*, strain 14028s is available from BEI Resources as NR-12154.

Genes were targeted for deletion by primers designed to preserve the first and last 30 bases of each deleted gene.[2] Gene replacement followed a modified Lambda-Red technique, with an added T7 RNA polymerase promoter positioned in plasmid pCLF3 to generate a gene-specific transcript from the *Salmonella* genome directly downstream of each mutant.[2-4] Detailed information about each mutant is shown in Table 1.

Material Provided:

Each inoculated well of the 96-well plate contains approximately 50 µL of culture in Luria Bertani (LB) broth containing 20 µg/mL chloramphenicol supplemented with 10% glycerol.

Packaging/Storage:

NR-29411 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 20 µg/mL chloramphenicol
Incubation:
Temperature: 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 24 hours.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Salmonella enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate 003/004_Cm, NR-29411."

Biosafety Level: 2

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

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References:

1. Andrews-Polymenis, H. and M. McClelland, Personal Communication.
2. Porwollik, S., et al. "Defined Single-Gene and Multi-Gene Deletion Mutant Collections in *Salmonella enterica* sv Typhimurium." PLoS One 9 (2014): e99820. PubMed: 25007190.
3. Santiviago, C. A., et al. "Analysis of Pools of Targeted *Salmonella* Deletion Mutants Identifies Novel Genes Affecting Fitness during Competitive Infection in Mice." PLoS Pathog. 5 (2009): e1000477. PubMed: 19578432.
4. Datsenko, K. A. and B. L. Wanner. "One-step Inactivation of Chromosomal Genes in *Escherichia coli* K-13 Using PCR Products." Proc. Natl. Acad. Sci. USA 97 (2000): 6640-6645. PubMed: 10829079.

Support Provided by NIAID

Table 1: *S. enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate 003/004_Cm[1,2]

Well Position	Deleted Region of Chromosome or Plasmid	Deletion Start	Deletion End	Locus Tag	14028S Gene Start	14028S Gene End	14028S Gene Strand	Description
A01	plasmid_14028S	10277	10900	STM14_5541	10247	10939	+	Plasmid-encoded fimbriae; chaperone
A02	chr_14028S	205814	208300	STM14_0207	205784	208330	+	Putative fimbrial usher
A03	chr_14028S	668767	669609	STM14_0705	668737	669639	+	Putative transcriptional regulator
A04	chr_14028S	1138622	1138732	STM14_1238	1138592	1138762	-	Putative cytoplasmic protein
A05	chr_14028S	1401171	1401452	STM14_1592	1401141	1401482	-	DNA-binding transcriptional activator OsmE
A06	chr_14028S	1498760	1499389	STM14_1701	1498730	1499419	-	Secreted effector protein
A07	chr_14028S	1963447	1964109	STM14_2244	1963417	1964139	+	Type III-secreted effector protein
A08	chr_14028S	2287854	2289065	STM14_2643	2287824	2289095	-	Putative transport protein
A09	chr_14028S	2604212	2605249	STM14_2999	2604182	2605279	+	Sulfate/thiosulfate transporter subunit
A10	chr_14028S	3049381	3051102	STM14_3484	3049351	3051132	+	Translocation machinery component
A11	chr_14028S	3711708	3712403	STM14_4253	3711678	3712433	+	Putative transcriptional regulator
A12	chr_14028S	4388961	4389911	STM14_4996	4388931	4389941	-	Putative cytoplasmic protein
B01	plasmid_14028S	10962	13310	STM14_5542	10932	13340	+	Plasmid-encoded fimbriae; usher protein
B02	chr_14028S	318919	319191	STM14_0326	318838	319221	-	Putative periplasmic protein
B03	chr_14028S[3]	690183	690803	STM14_0728	690153	690833	-	Two-component response regulator DpiA
B04	chr_14028S	1139012	1140421	STM14_1240	1138982	1140544	+	Pathogenicity island-encoded protein D
B05	chr_14028S	1403983	1404765	STM14_1597	1403953	1404795	-	DNA-binding transcriptional regulator ChbR
B06	chr_14028S	1687103	1688110	STM14_1924	1687073	1688143	+	Putative NADP-dependent oxidoreductase
B07	chr_14028S	2026190	2027791	STM14_2334	2026160	2028052	+	Methyl accepting chemotaxis protein II
B08	chr_14028S	2289122	2290066	STM14_2644	2289092	2290096	-	Putative glycohydrolase
B09	chr_14028S	2932888	2933367	STM14_3337	2932858	2933397	+	Phase-1 flagellin repressor
B10	chr_14028S	3055513	3056463	STM14_3491	3055483	3056493	+	Needle length control protein
B11	chr_14028S	3837836	3840304	STM14_4385	3837806	3840334	+	Long polar fimbrial outer membrane usher protein
B12	chr_14028S	4429048	4429857	STM14_5040	4429018	4429887	-	23S rRNA pseudouridine synthase F
C01	plasmid_14028S	26296	26886	STM14_5560	26266	26916	+	Hydrophilic protein
C02	chr_14028S	328445	329173	STM14_0334	328415	329203	-	Putative cytoplasmic protein
C03	chr_14028S	733763	734788	STM14_0779	733733	734818	+	Putative phosphate starvation-inducible protein
C04	chr_14028S	1176649	1177269	STM14_1290	1176619	1177299	+	N-acetylmannosamine-6-phosphate 2-epimerase
C05	chr_14028S	1432804	1433457	STM14_1632	1432774	1433487	-	Hypothetical protein
C06	chr_14028S	1702188	1703078	STM14_1940	1702158	1703108	-	Secreted effector protein
C07	chr_14028S	2046215	2046487	STM14_2359	2046185	2046517	+	Putative inner membrane protein
C08	chr_14028S	2366139	2367635	STM14_2739	2366109	2367665	-	Hypothetical protein
C09	chr_14028S	2947067	2948059	STM14_3350	2947037	2948089	+	Secreted effector protein
C10	chr_14028S	3074863	3075714	STM14_3514	3074812	3075744	-	Putative transcriptional regulator
C11	chr_14028S	3868330	3869070	STM14_4423	3868300	3869100	+	Transcriptional repressor
C12	chr_14028S	4562749	4563408	STM14_5176	4562719	4563438	+	DNA-binding transcriptional activator DcuR
D01	plasmid_14028S	28213	29928	STM14_5562	28183	29958	+	Hydrophilic protein
D02	chr_14028S	329260	329586	STM14_0335	329200	329616	-	Putative periplasmic protein
D03	chr_14028S	785695	786528	STM14_0839	785662	786558	-	Putative glycosyl transferase

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Well Position	Deleted Region of Chromosome or Plasmid	Deletion Start	Deletion End	Locus Tag	14028S Gene Start	14028S Gene End	14028S Gene Strand	Description
D04	chr_14028S	1187077	1187433	STM14_1304	1187047	1187463	+	Curli assembly protein CsgF
D05	chr_14028S	1490005	1490346	STM14_1688	1489975	1490376	-	Secreted effector protein
D06	chr_14028S	1706313	1707431	STM14_1944	1706283	1707461	-	Putative benzoate membrane transport protein
D07	chr_14028S	2070637	2071284	STM14_2392	2070607	2071314	-	Flagellar assembly protein H
D08	chr_14028S	2390767	2391072	STM14_2766	2390737	2391105	+	Putative phage antiterminator
D09	chr_14028S	3030166	3031013	STM14_3462	3030207	3031043	+	Secreted effector protein
D10	chr_14028S	3107354	3108247	STM14_3550	3107324	3108277	-	Secreted effector protein
D11	chr_14028S	3883401	3884222	STM14_4439	3883371	3884252	-	Putative transcriptional regulator
E01	plasmid_14028S	92483	93163	STM14_5627	92453	93193	-	Conjugative transfer: fimbrial acetylation
E02	chr_14028S[4]	377378	378283	STM14_0389	377348	378313	-	Putative transcriptional regulator
E03	chr_14028S	867315	869552	STM14_928	867285	869582	-	Leucine-rich repeat-containing protein
E04	chr_14028S	1189325	1189720	STM14_1309	1189295	1189750	-	Curlin minor subunit
E05	chr_14028S	1493940	1494470	STM14_1694	1493910	1494500	-	Translocation machinery component
E06	chr_14028S	1707545	1708153	STM14_1945	1707515	1708183	+	Putative outer membrane lipoprotein
E07	chr_14028S	2074970	2075914	STM14_2397	2074940	2075944	-	Flagellar motor switch protein FliM
E08	chr_14028S	2392468	2394774	STM14_2769	2392438	2394804	+	Leucine-rich repeat-containing protein
E09	chr_14028S	3033606	3033998	STM14_3467	3033576	3034028	+	Putative cytoplasmic protein
E10	chr_14028S	3297009	3297791	STM14_3769	3296979	3297821	+	Putative transcriptional regulator
E11	chr_14028S	4038389	4039021	STM14_4616	4038359	4039051	-	DNA-binding transcriptional regulator TorR
E12	chr_14028S	4729876	4732671	STM14_5371	4729846	4732701	+	Valyl-tRNA synthetase
F01	chr_14028S[5]	24499	24981	STM14_0028	24469	25011	-	Fimbrial subunit
F02	chr_14028S	384301	385002	STM14_0395	384271	385032	+	Putative fimbrial chaperone
F03	chr_14028S	921087	922565	STM14_992	921057	922595	-	Putative ABC transporter periplasmic binding protein
F04	chr_14028S	1196138	1198621	STM14_1318	1196108	1198651	-	Glucosyltransferase MdoH
F05	chr_14028S	1495003	1496397	STM14_1696	1494973	1496427	-	Translocation machinery component
F06	chr_14028S	1731300	1732466	STM14_1974	1731270	1732496	-	Secreted effector protein
F07	chr_14028S[6]	2193152	2195440	STM14_2557	2193122	2195470	-	Secreted effector protein
F08	chr_14028S	2443437	2444636	STM14_2820	2443407	2444666	-	Anaerobic glycerol-3-phosphate dehydrogenase subunit B
F09	chr_14028S	3042338	3043909	STM14_3477	3042308	3043939	+	Protein tyrosine phosphatase/GTPase activating protein
F10	chr_14028S	3414671	3414979	STM14_3910	3414641	3415009	-	Putative periplasmic protein
F11	chr_14028S	4055021	4056034	STM14_4629	4054991	4056064	+	Recombination protein F
F12	chr_14028S	4748438	4749367	STM14_5387	4748408	4749397	-	Putative restriction endonuclease
G01	chr_14028S	194139	194846	STM14_0196	194109	194876	-	Putative transcriptional regulator
G02	chr_14028S	484072	484731	STM14_0508	484042	484761	+	2-aminoethylphosphonate transport protein
G03	chr_14028S	1012450	1013349	STM14_1098	1012420	1013379	-	SopD-like protein
G04	chr_14028S	1218001	1219152	STM14_1348	1217971	1219182	-	Flagellar hook protein FlgE
G05	chr_14028S	1496473	1497000	STM14_1697	1496443	1497030	-	Translocation machinery component
G06	chr_14028S	1739122	1739499	STM14_1982	1738867	1739529	-	Putative SAM-dependent methyltransferase
G07	chr_14028S	2256574	2258370	STM14_2616	2256544	2258400	+	Putative assembly protein
G08	chr_14028S	2460551	2462137	STM14_2839	2460521	2462167	-	4-amino-4-deoxy-L-arabinose transferase
G09	chr_14028S	3044946	3046943	STM14_3481	3044916	3046973	+	Secreted effector protein
G10	chr_14028S	3524479	3525057	STM14_4033	3524449	3525087	+	Stringent starvation protein A
G11	chr_14028S	4191777	4192322	STM14_4777	4191747	4192352	-	Putative inner membrane protein
G12	chr_14028S	4839330	4841678	STM14_5492	4839300	4841708	+	Putative fimbrial usher protein
H01	chr_14028S	204726	205745	STM14_0206	204696	205775	+	Putative fimbrial protein precursor
H02	chr_14028S	609399	610346	STM14_0639	609369	610376	-	Minor fimbrial subunit
H03	chr_14028S	1098346	1099254	STM14_1193	1098316	1099284	-	Secreted effector protein
H04	chr_14028S	1268037	1268987	STM14_1400	1268007	1269017	+	Secreted effector protein
H07	chr_14028S	2283079	2284065	STM14_2636	2283049	2284095	-	Pseudogene
H08	chr_14028S	2588352	2589218	STM14_2979	2588322	2589248	+	Putative transcriptional regulator
H09	chr_14028S	3047022	3047993	STM14_3482	3046992	3048023	+	Translocation machinery component
H10	chr_14028S	3680866	3681720	STM14_4225	3680836	3681750	-	Putative cytoplasmic protein
H11	chr_14028S	4195034	4195753	STM14_4781	4195004	4195783	-	TatABCE protein translocation system subunit
H12	chr_14028S	4865712	4866197	STM14_5516	4865682	4866227	+	Putative fimbrial chaperone

[1]All information in this table was provided by the depositor at the time of deposition.
[2]Construction of each listed mutant has been confirmed either by PCR or by an array indicating a functional T7 promoter in the correct location and orientation. Mutants that did not produce such a signal on the array, or did not yield the expected mutant product during PCR, are not listed.
[3]Deleted region also overlaps STM14_0727 (0.1%)
[4]Deleted region also overlaps STM14_0390 (9.5%)
[5]Deleted region also overlaps STM14_0029 (1.3%)
[6]Deleted region also overlaps STM14_2556 (25.2%)

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