Salmonella enterica subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate 017/018_Cm

Catalog No. NR-29418

For research use only. Not for human use.

Contributor:

Helene Andrews-Polymenis, Associate Professor, Department of Microbial Pathogenesis and Immunology, College of Medicine, Texas A&M Health Science Center, Bryan, Texas, USA and Michael McClelland, Professor, Scientific Director, Vaccine Research Institute of San Diego, San Diego, California, USA

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-29418 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 1 day.

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 017/018_Cm, NR-29418."

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 017/018_Cm[1,2]

Well Position	Deleted Region of Chromosome	Deletion Start	Deletion End	Locus Tag	14028S Gene Start	14028S Gene End	14028S Gene Strand	Description
A01	chr_14028S	16118	16402	STM14_0017	16088	16432	-	Putative bacteriophage protein
A02	chr_14028S	1133922	1134542	STM14_1232	1133892	1134572	+	Pathogenicity island-encoded protein A
A03	chr_14028S	1491882	1493033	STM14_1690	1491852	1493063	-	Virulence protein
A04	chr_14028S	1500283	1500972	STM14_1705	1500253	1501002	-	Needle complex inner membrane lipoprotein
A05	chr_14028S	1507359	1507673	STM14_1713	1507329	1507703	-	Type III secretion system apparatus protein
A06	chr_14028S	1966442	1966801	STM14_2249	1966412	1966831	+	Putative acetyltransferase
A07	chr_14028S	3036609	3037727	STM14_3473	3036579	3037757	+	Needle complex inner membrane protein
A08	chr_14028S	3053652	3053852	STM14_3488	3053622	3053882	+	Needle complex export protein
A09	chr_14028S	3061864	3063492	STM14_3497	3061834	3063522	+	Outer membrane secretin precursor
A10	chr_14028S	3068486	3068566					
A11	chr_14028S	3965455	3966432	STM14_4524	3965425	3966462	-	Putative cytoplasmic protein
A12	chr_14028S	3978210	3978845	STM14_4538	3978180	3978875	+	Mg2+ transport protein
B01	chr_14028S	75118	75406	STM14_0076	74964	75239	+	Hypothetical protein
B03	chr_14028S	1493101	1493283	STM14_1691	1493071	1493313	-	Secretion system effector
B04	chr_14028S	1501050	1501538	STM14_1706	1501020	1501568	-	Putative cytoplasmic protein
B05	chr_14028S	1507714	1508622	STM14_1714	1507684	1508652	-	Type III secretion system protein
B06	chr_14028S	2446378	2447271	STM14_2824	2446348	2447301	-	Deubiquitinase
B07	chr_14028S	3041802	3042224	STM14_3476	3041772	3042254	-	Invasion protein precursor
B08	chr_14028S	3053938	3054552	STM14_3489	3053908	3054582	+	Surface presentation of antigens protein SpaP
B09	chr_14028S	3063549	3064139	STM14_3498	3063519	3064268	+	Invasion regulatory protein
B11	chr_14028S	3967791	3968348	STM14_4526	3967761	3968378	-	Putative cytoplasmic protein
B12	chr_14028S	4253706	4253921	STM14_4845	4253676	4253987	+	Putative cytoplasmic protein
C01	chr_14028S	737824	738339	STM14_0783	737794	738369	+	Putative inner membrane protein
C02	chr_14028S	1136247	1136528	STM14_1236	1136217	1136558	+	Pathogenicity island-encoded protein C
C03	chr_14028S	1493607	1493873	STM14_1693	1493577	1493903	-	Secretion system chaperone protein
C04	chr_14028S[3]	1501595	1502209	STM14_1707	1501565	1502239	-	Type III secretion system apparatus protein
C05	chr_14028S	1508750	1509337	STM14_1715	1508720	1509367	-	Type III secretion system protein
C06	chr_14028S	2457676	2459598	STM14_2837	2457646	2459628	-	Bifunctional UDP-glucuronic acid decarboxylase/UDP-4-amino-4-deoxy-L-arabinose formyltransferase
C07	chr_14028S	3043956	3044288	STM14_3478	3043926	3044318	+	Secretion chaperone
C08	chr_14028S	3054602	3055453	STM14_3490	3054572	3055483	+	Surface presentation of antigens protein SpaO
C09	chr_14028S	3064656	3065039	STM14_3499	3064626	3065069	-	Needle complex outer membrane lipoprotein precursor
C10	chr_14028S	3335936	3336286	STM14_3821	3335906	3336316	-	Putative cytoplasmic protein
C11	chr_14028S	3968483	3971290	STM14_4527	3968453	3971320	-	Putative autotransporter
D01	chr_14028S	786601	787674	STM14_0840	786571	787704	-	Putative glycosyl transferase
D02	chr_14028S	1138793	1138843	STM14_1239	1138763	1138873	+	Putative cytoplasmic protein
D03	chr_14028S	1494527	1494940	STM14_1695	1494497	1494970	-	Secretion system chaperone
D04	chr_14028S[4]	1502235	1503191	STM14_1708	1502205	1503221	-	Type III secretion system apparatus protein
D05	chr_14028S	1509394	1509600	STM14_1716	1509364	1509630	-	Type III secretion system apparatus protein
D06	chr_14028S	2900699	2901832	STM14_3305	2900669	2901862	-	Putative cytoplasmic protein

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Well Position	Deleted Region of Chromosome	Deletion Start	Deletion End	Locus Tag	14028S Gene Start	14028S Gene End	14028S Gene Strand	Description
D07	chr_14028S	3044375	3044575	STM14_3479	3044345	3044605	+	Putative cytoplasmic protein
D08	chr_14028S	3056523	3056906	STM14_3492	3056493	3056936	+	Needle complex assembly protein
D09	chr_14028S	3065530	3065919	STM14_3500	3065500	3065949	-	Putative cytoplasmic protein
D10	chr_14028S	3822662	3823900	STM14_4369	3822632	3823930	-	Putative transport protein
D11	chr_14028S	3971445	3971849	STM14_4528	3971415	3971879	+	Putative inner membrane protein
D12	chr_14028S	4493436	4494695	STM14_5119	4493406	4494725	-	Putative ABC exporter outer membrane component
E01	chr_14028S	788637	789287	STM14_0842	788607	789317	-	Putative ABC-type polysaccharide/polyol phosphate transport system ATPase component
E02	chr_14028S	1156052	1156354	STM14_1261	1156022	1156384	+	Hypothetical protein
E03	chr_14028S	1497531	1497905	STM14_1699	1497501	1497935	-	Secretion system chaperone
E04	chr_14028S	1503309	1503617	STM14_1709	1503279	1503647	-	Type III secretion system apparatus protein
E05	chr_14028S	1509661	1510380	STM14_1717	1509631	1510410	-	Type III secretion system apparatus protein
E06	chr_14028S[5]	3034055	3034675	STM14_3468	3034025	3034705	+	Needle complex export protein
E07	chr_14028S	3044679	3044867	STM14_3480	3044649	3044897	+	Acyl carrier protein
E08	chr_14028S	3056944	3058179	STM14_3493	3056914	3058209	+	ATP synthase SpaL
E09	chr_14028S	3065964	3066251	STM14_3501	3065934	3066281	-	Putative cytoplasmic protein
E10	chr_14028S	3870858	3871730	STM14_4427	3870828	3871760	-	Putative chemotaxis protein
E11	chr_14028S	3972830	3972883					
E12	chr_14028S	4494752	4495969	STM14_5120	4494722	4495999	-	Cation efflux pump
F02	chr_14028S	1357534	1358067	STM14_1526	1357504	1358097	-	Putative response regulator
F03	chr_14028S	1499543	1499698	STM14_1702	1499513	1499728	-	Type III secretion system apparatus protein
F04	chr_14028S	1503662	1505647	STM14_1710	1503632	1505677	-	Secretion system apparatus protein SsaV
F05	chr_14028S	1510437	1511435	STM14_1718	1510407	1511465	-	Secretion system apparatus protein SsaU
F06	chr_14028S	3035263	3035961	STM14_3470	3035233	3035991	+	Needle complex inner membrane lipoprotein
F07	chr_14028S	3051165	3051602	STM14_3485	3051135	3051632	+	Secretion chaperone
F08	chr_14028S	3058236	3058583	STM14_3494	3058206	3058613	+	Secretion chaperone
F09	chr_14028S	3066584	3066850	STM14_3503	3066554	3066880	+	Putative cytoplasmic protein
F10	chr_14028S	3929405	3929578	STM14_4481	3929375	3929608	+	Putative inner membrane protein
F11	chr_14028S	3973561	3974181	STM14_4532	3973531	3974211	-	Putative inner membrane protein
F12	chr_14028S	4496046	4512665	STM14_5121	4496016	4512695	-	Putative inner membrane protein
G01	chr_14028S	792348	794138	STM14_0845	792273	794168	-	Putative glycosyl transferase
G02	chr_14028S	1485266	1485934	STM14_1684	1485236	1485964	+	Putative regulatory protein
G03	chr_14028S	1499799	1499966	STM14_1703	1499769	1499996	-	Type III secretion system apparatus protein
G04	chr_14028S	1505697	1506938	STM14_1711	1505667	1506968	-	Type III secretion system ATPase
G05	chr_14028S	1770356	1771120	STM14_2015	1770326	1771156	+	Hypothetical protein
G06	chr_14028S	3036018	3036263	STM14_3471	3035988	3036293	+	Needle complex minor subunit
G07	chr_14028S	3051800	3052810	STM14_3486	3051770	3052840	+	Surface presentation of antigens protein SpaS
G08	chr_14028S	3058667	3060664	STM14_3495	3058637	3060694	+	Needle complex export protein
G09	chr_14028S	3067599	3067829	STM14_3505	3067569	3067859	-	Putative ABC-type transporter
G10	chr_14028S	3962709	3962993	STM14_4519	3962679	3963023	+	Putative cytoplasmic protein
G11	chr_14028S	3974470	3974889	STM14_4534	3974440	3974919	+	Putative inner membrane protein
G12	chr_14028S	4512765	4514771	STM14_5122	4512735	4514801	-	Putative ABC-type bacteriocin/lantibiotic exporter
H01	chr_14028S[6]	1062657	1062896	STM14_1145	1062627	1062926	+	Hypothetical protein
H02	chr_14028S	1490408	1491841	STM14_1689	1490378	1491871	-	Outer membrane secretin precursor
H03	chr_14028S	1500038	1500226	STM14_1704	1500008	1500256	-	Type III secretion system apparatus protein
H04	chr_14028S	1507001	1507318	STM14_1712	1506971	1507348	-	Type III secretion system apparatus protein
H05	chr_14028S	1964853	1965188	STM14_2247	1964823	1965218	-	Putative cytoplasmic protein
H06	chr_14028S	3036342	3036524	STM14_3472	3036312	3036554	+	Needle complex major subunit
H07	chr_14028S	3052857	3053588	STM14_3487	3052827	3053618	+	Needle complex export protein
H08	chr_14028S	3060749	3061807	STM14_3496	3060719	3061837	+	Invasion protein
H09	chr_14028S	3067886	3068353	STM14_3506	3067856	3068383	-	Putative acetyltransferase
H10	chr_14028S	3963731	3964861	STM14_4522	3963701	3964891	-	ATP binding protein
H11	chr_14028S	3975264	3977930	STM14_4536	3975234	3977960	+	Mg2+ transporter
H12	chr_14028S	4678797	4679510	STM14_5314	4678716	4679540	-	Putative DNA-binding protein

[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_1708 (0.5%)
[4]Deleted region also overlaps STM14_1707 (0.7%)
[5]Deleted region also overlaps STM14_3469 (2.3%)
[6]Alternative deleted region: 2824342 – 2824632