

***Salmonella enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_053/054_Kan**

Catalog No. NR-42837

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 kanamycin-resistant mutant collection contains 3517 mutants distributed among 11 96-well plates, in which a single gene is replaced by a cassette conferring the kanamycin resistance gene, and includes 9 double mutants that contain both kanamycin and chloramphenicol cassettes. Deletions were confirmed by the depositor.^{1,2}

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.

Note: The strain designation on the plate, strain CDC 6516-60, is incorrect. The correct strain designation is strain 14028s. *S. enterica* subsp. *enterica*, strain 14028s was originally known as strain 14028. A variant of the original strain with a rough colony morphology was designated 14028r and the original smooth strain was renamed 14028s. Strain 14028 is a descendent of strain CDC 6516-60 which was isolated from pools of hearts and livers of 4-week-old chickens.⁵ The complete genome of *S. enterica* subsp. *enterica*, strain 14028s (GenBank: [CP001363.1](#)) and plasmid (GenBank: [CP001362.1](#)) sequences are available.

Material Provided:

Each inoculated well of the 96-well plate contains approximately 50 µL of culture in Luria Bertani (LB) broth containing 60 µg/mL kanamycin supplemented with 10% glycerol.

Packaging/Storage:

NR-42837 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 60 µg/mL kanamycin

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 SGD_053/054_Kan, NR-42837."

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/bmb15/index.htm.

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References:

1. McClelland, M., 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.

5. Jarvik, T., et al. "Short-Term Signatures of Evolutionary Change in the *Salmonella enterica* Serovar Typhimurium 14028 Genome." *J. Bacteriol.* 192 (2010): 560-567. PubMed: 19897643.

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Table 1: *S. enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_053/054_Kan^{1,2}

Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region Start	Deleted Region End	Gene Strand	Description
A02	CDS	3184950	3185168	STM14_3630	3184980	3185137	-	Putative peptide transport protein
A04	CDS	2665662	2666300	STM14_3065	2665692	2666269	-	Phosphoribosylglycinamide formyltransferase
A05	CDS	1562124	1562459	STM14_1780	1562154	1562428	-	Putative inner membrane protein
A07	CDS	4817461	4818372	STM14_5461	4817491	4818284	+	Hypothetical protein
A08	CDS	1829100	1829144	STM14_2085	1829106	1829137	-	Trp operon leader peptide
A09	CDS	3817164	3817916	STM14_4359	3817194	3817885	+	Cell division protein
A10	CDS	2414687	2415748	STM14_2795	2414717	2415717	+	O6-methylguanine-DNA methyltransferase/transcription activator/repressor
A12	CDS	4621626	4621823	STM14_5246	4621656	4621792	-	Putative inner membrane protein
B03	CDS	1877411	1878835	STM14_2138	1877441	1878804	+	Hypothetical protein
B04	CDS	1861748	1862206	STM14_2121	1861778	1862175	-	Hypothetical protein
B08	CDS	1561266	1561997	STM14_1779	1561296	1561966	+	DNA-binding transcriptional regulator RstA
B09	CDS	899345	900232	STM14_0968	899375	900201	+	Threonine and homoserine efflux system
B12	CDS	4717774	4717875	STM14_5354	4717804	4717844	+	PyrBI operon leader peptide
C01	CDS	3010358	3010903	STM14_3436	3010388	3010872	+	Electron transport protein HydN
C02	CDS	3519673	3520362	STM14_4028	3519703	3520331	+	N-acetylmannosamine-6-phosphate 2-epimerase
C06	CDS	2861349	2861548	STM14_3265	2861403	2861517	-	
C07	CDS	4067770	4068957	STM14_4642	4067800	4068926	-	Multidrug efflux system protein MdtL
C08	CDS	3761921	3763138	STM14_4309	3761951	3763149	+	Hypothetical protein
C09	CDS	3217737	3218282	STM14_3671	3217767	3218251	-	Isopentenyl-diphosphate delta-isomerase
D01	CDS	2494855	2495454	STM14_2875	2494885	2495423	-	Hypothetical protein
D04	CDS	884739	885734	STM14_0949	884769	885703	+	Hypothetical protein
D05	CDS	2498696	2499151	STM14_2879	2498726	2499120	+	Hypothetical protein
D11	CDS	4059858	4060115	STM14_4636	4059888	4060084	-	Hypothetical protein
D12	CDS	2735138	2735869	STM14_3121	2735168	2735838	+	Putative rRNA methylase
E01	CDS	3134886	3135335	STM14_3572	3134916	3135304	+	Flavodoxin
E02	CDS	2672763	2672954	STM14_3071	2672811	2672923	-	Putative inner membrane protein
E09	CDS	4149733	4150812	STM14_4725	4149763	4150781	-	4-alpha-L-fucosyltransferase
E12	CDS	2652662	2653525	STM14_3049	2652692	2653494	+	Putative inner membrane protein
F04	CDS	2719315	2724249	STM14_3105	2719345	2724218	+	Putative inner membrane lipoprotein
F07	CDS	2509960	2510514	STM14_2892	2509990	2510483	+	Putative NTP pyrophosphohydrolase
F09	CDS	1950043	1951416	STM14_2230	1950073	1951385	-	Putative transport protein
G02	CDS	1033980	1034873	STM14_1119	1034010	1034842	-	Hypothetical protein

Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region Start	Deleted Region End	Gene Strand	Description
G03	CDS	4230571	4230990	STM14_4821	4230601	4230968	+	Putative cytoplasmic protein
G04	CDS	1961534	1961773	STM14_2239	1961564	1961742	-	Putative cytoplasmic protein
G06	CDS	2609180	2610079	STM14_3004	2609210	2610048	+	Putative iron-dependent peroxidase
G07	CDS	2611247	2611807	STM14_3007	2611277	2611752	+	Putative acetyltransferase
G08	CDS	317578	318063	STM14_0324			-	Putative cytoplasmic protein
G11	CDS	20058	23054	STM14_0023	20088	23023	-	Putative hydroxymethyltransferase
G12	CDS	3034662	3035261	STM14_3469	3034692	3035230	+	Needle complex assembly protein
H03	CDS (LT2) ³				490670	490743		
H04	CDS	3822374	3822514	STM14_4368	3822404	3822483	-	Cystathionine gamma-synthase
H06	CDS	4012345	4013178	STM14_4585	4012348	4013147	-	Integral membrane protein
H09	CDS	3696823	3698376	STM14_4239	3696853	3698345	+	Putative ribonucleoprotein related-protein
H10	CDS	2394973	2395176	STM14_2770	2395003	2395145	+	Putative phage tail fiber protein
H11	CDS	5114	5887	STM14_0005	5144	5856	+	Hypothetical protein
H12	CDS	9376	9942	STM14_0009	9406	9911	+	Hypothetical protein

¹All information in this table was provided by the depositor at the time of deposition.

²Construction 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.

³Of the targeted genes, 22 CDSs and 22 sRNA were annotated in strain LT2 but not annotated in strain 14028s.