

***Salmonella enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD\_059/060\_Kan**

**Catalog No. NR-42840**

**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.<sup>1,2</sup> 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.<sup>1,2</sup>

Genes were targeted for deletion by primers designed to preserve the first and last 30 bases of each deleted gene.<sup>2</sup> 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.<sup>2-4</sup> 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.<sup>5</sup> 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-42840 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\_059/060\_Kan, NR-42840."

**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](http://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\_059/060\_Kan<sup>1,2</sup>**

Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region Start	Deleted Region End	Gene Strand	Description
A01	CDS	1525050	1525946	STM14_1737	1525080	1525915	-	Putative aldo/keto reductase
A02	CDS	1531083	1532204	STM14_1744	1531113	1532173	-	Anhydro-N-acetylmuramic acid kinase
A03	CDS	1524760	1524999	STM14_1736	1524790	1524968	-	Putative oxidoreductase
A04	CDS	1524058	1524657	STM14_1735	1524088	1524626	+	Putative transcriptional repressor
A05	CDS	1436576	1437409	STM14_1637	1436606	1437378	+	Hypothetical protein
A06	CDS	1455097	1456215	STM14_1657	1455127	1456184	+	Putative inner membrane protein
A07	CDS	1433549	1434991	STM14_1633	1433579	1434960	-	Hypothetical protein
A08	CDS	1416287	1417045	STM14_1611	1416317	1417014	-	Putative outer membrane protein
A09	CDS	1384916	1385467	STM14_1574	1384946	1385436	-	Hypothetical protein
A11	CDS	1410051	1411442	STM14_1604	1410081	1411411	+	Kinase/transporter-like protein
B01	CDS	1933939	1934517	STM14_2207	1933969	1934486	-	Hypothetical protein
B02	CDS	1380462	1380740	STM14_1568	1380492	1380709	-	Putative cytoplasmic protein
B03	CDS	1377451	1378335	STM14_1563	1377481	1378304	+	Aldose 1-epimerase
B04	CDS	1371804	1373738	STM14_1558	1371834	1373707	+	Putative serine protein kinase
B05	CDS	1370395	1371681	STM14_1556	1370425	1371650	+	Hypothetical protein
B07	CDS	1364878	1366062	STM14_1547	1364908	1366031	+	Putative amino acid/amine transport protein
B09	CDS	1996485	1997804	STM14_2299	1996515	1997773	+	Hypothetical protein
B10	CDS	2005190	2005930	STM14_2310	2005220	2005899	+	Hypothetical protein
B11	CDS	1992889	1993758	STM14_2295	1992919	1993727	-	DNA-binding transcriptional regulator HexR
B12	CDS	1943495	1944115	STM14_2216	1943525	1944084	-	Hypothetical protein
C01	CDS	1955391	1955975	STM14_2234	1955421	1955944	+	Putative nucleotide-binding protein
C02	CDS	1956017	1957300	STM14_2235	1956047	1957269	-	Putative inner membrane protein
C03	CDS	1959980	1961419	STM14_2237	1960055	1961388	-	rRNA (cytosine-C(5)-)-methyltransferase RsmF
C04	CDS	2052017	2052769	STM14_2369	2052047	2052738	+	Putative amino-acid ABC transporter ATP-binding protein YecC
C05	CDS	2008612	2009178	STM14_2314	2008642	2009147	-	Hypothetical protein
C06	CDS	2050772	2050996	STM14_2367	2050802	2050965	-	Hypothetical protein
C07	CDS	2013195	2013764	STM14_2320	2013225	2013733	+	Hypothetical protein
C08	CDS	2010482	2011225	STM14_2317	2010512	2011194	-	Putative SAM-dependent methyltransferase
C09	CDS	2011222	2012193	STM14_2318	2011252	2012162	-	Putative enzyme
C10	CDS	2052766	2053434	STM14_2370	2052796	2053403	+	Putative ABC-type amino acid transporter permease component

Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region Start	Deleted Region End	Gene Strand	Description
C11	CDS	2065147	2066412	STM14_2385	2065177	2066381	-	Putative inner membrane protein
C12	CDS	2083287	2084198	STM14_2410	2083317	2084167	+	Hypothetical protein
D01	CDS	2053455	2054441	STM14_2371	2053485	2054410	+	D-cysteine desulfhydrase
D02	CDS	2185780	2186838	STM14_2550	2185810	2186807	+	Putative inner membrane protein
D03	CDS	2197379	2198743	STM14_2560	2197409	2198712	+	Putative amino acid transport protein
D04	CDS	2133902	2134699	STM14_2485	2133932	2134668	-	Putative inner membrane protein
D05	CDS	2185200	2185535	STM14_2548	2185230	2185504	+	Hypothetical protein
D06	CDS	2199983	2200807	STM14_2564	2200013	2200776	+	Putative dehydratase
D07	CDS	2273278	2274690	STM14_2627	2273308	2274659	-	Multidrug efflux system protein MdtE
D08	CDS	2264044	2265396	STM14_2622	2264122	2265365	-	Putative chaperone
D09	CDS	2254878	2256458	STM14_2615	2254908	2256427	-	Putative inner membrane protein
D10	CDS	2267078	2270200	STM14_2625	2267108	2270169	-	Multidrug efflux system subunit MdtB
D11	CDS	2270201	2273281	STM14_2626	2270231	2273250	-	Multidrug efflux system subunit MdtC
D12	CDS	2281237	2282598	STM14_2634	2281267	2282567	-	Putative protease
E01	CDS	2285566	2286465	STM14_2641	2285596	2286434	-	Lipid kinase
E02	CDS	2305297	2306016	STM14_2662	2305327	2305985	+	Putative two-component response-regulatory protein YehT
E03	CDS	2306013	2307800	STM14_2663	2306043	2307667	+	Putative sensor kinase
E04	CDS	2308800	2309531	STM14_2666	2308830	2309500	+	Putative ABC-type proline/glycine betaine transport system permease component
E05	CDS	2309515	2310462	STM14_2667	2309545	2310431	+	Putative ABC-type proline/glycine betaine transport system ATPase component
E06	CDS	2311628	2312545	STM14_2669	2311658	2312514	+	Putative transport protein
E07	CDS	2352100	2352963	STM14_2717	2352130	2352932	+	Putative DNA-binding transcriptional regulator
E08	CDS	2342995	2343852	STM14_2707	2343025	2343701	-	S-formylglutathione hydrolase
E09	CDS	2353084	2354133	STM14_2719	2353114	2354102	-	Putative inner membrane protein
E10	CDS	2362139	2362849	STM14_2733	2362076	2362818	-	Elongation factor P
E11	CDS	2363062	2364048	STM14_2734	2363092	2364017	-	Putative cobalamin biosynthetic protein
F01	CDS	2369587	2370681	STM14_2741	2369617	2370650	-	Putative ABC-type dipeptide/oligopeptide/nickel transport system permease
F02	CDS	2370681	2371706	STM14_2742	2370711	2371675	-	Putative ABC-type dipeptide/oligopeptide/nickel transport system permease component
F03	CDS	2371708	2373297	STM14_2743	2371738	2373266	-	Putative ABC-type transport system ATPase component
F04	CDS	2373301	2373645	STM14_2744	2373331	2373614	+	Hypothetical protein
F05	CDS	2376101	2377861	STM14_2748	2376131	2377830	-	Putative ATP-dependent helicase
F06	CDS	2379027	2380034	STM14_2752	2379057	2380003	+	Nucleoid-associated protein NdpA
F07	CDS	2380214	2380441	STM14_2753	2380244	2380410	-	Hypothetical protein
F08	CDS	2454150	2454575	STM14_2832	2454180	2454544	-	Putative NTP pyrophosphohydrolase
F09	CDS	2453329	2453976	STM14_2831	2453359	2453945	+	Putative inner membrane protein
F10	CDS	2493547	2494761	STM14_2874	2493577	2494730	-	Aminotransferase AlaT
F11	CDS	2495546	2497372	STM14_2876	2495576	2497341	+	Putative response regulator
F12	CDS	2498119	2498613	STM14_2878	2498149	2498582	+	Hypothetical protein
G01	CDS	2544456	2545265	STM14_2931	2544486	2545234	+	Hypothetical protein
G02	CDS	2547214	2548146	STM14_2934	2547244	2548061	+	N5-glutamine S-adenosyl-L-methionine-dependent methyltransferase
G03	CDS	2510571	2511122	STM14_2893	2510601	2511091	+	Phosphodiesterase
G04	CDS	2511178	2511822	STM14_2894	2511208	2511791	+	Putative glutathione S-transferase
G05	CDS	2511965	2512612	STM14_2895	2511995	2512581	-	Putative glutathione S-transferase
G06	CDS	2512738	2513631	STM14_2896	2512768	2513600	-	Putative sugar nucleotide epimerase
G07	CDS	2543600	2543875	STM14_2929	2543630	2543844	+	Putative cytoplasmic protein
G08	CDS	2543908	2544456	STM14_2930	2543938	2544425	+	Putative cytoplasmic protein
G09	CDS	2548312	2548863	STM14_2935	2548342	2548832	-	Hypothetical protein
G10	CDS	2549657	2551804	STM14_2937	2549687	2551773	+	Multifunctional fatty acid oxidation complex subunit alpha
H01	CDS	2567557	2568795	STM14_2954	2567587	2568764	+	Aminotransferase
H02	CDS	2589338	2590336	STM14_2980	2589368	2590305	-	Putative Na <sup>+</sup> -dependent transporter
H03	CDS	2648886	2649242	STM14_3045	2648916	2649211	-	Hypothetical protein
H04	CDS	2638641	2639216	STM14_3038	2638671	2639185	+	Putative pyrophosphohydrolase
H05	CDS	2710249	2711415	STM14_3097	2710279	2711384	+	Hypothetical protein
H06	CDS	2661026	2661385	STM14_3059	2661056	2661354	-	Putative arsenate reductase

Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region Start	Deleted Region End	Gene Strand	Description
H07	CDS	2661412	2662137	STM14_3060	2661442	2662106	+	DNA replication initiation factor
H08	CDS	2704600	2705778	STM14_3091	2704630	2705747	+	Outer membrane protein assembly complex subunit YfgL
H09	CDS	2705789	2706409	STM14_3092	2705819	2706378	+	Putative inner membrane protein
H10	CDS	2755627	2756964	STM14_3141	2755657	2756933	+	Putative transcriptional regulator
H11	CDS	2766121	2766756	STM14_3149	2766151	2766725	+	Hypothetical protein
H12	CDS	2732323	2732646	STM14_3116	2732353	2732615	+	Iron-sulfur cluster assembly protein

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

<sup>2</sup>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.