

Product Information Sheet for NR-42826

***Salmonella enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_031/032_Kan**

Catalog No. NR-42826

For research use only. Not for human use.

Contributor:

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 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-42826 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_031/032_Kan, NR-42826."

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.

Disclaimers:

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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_031/032_Kan^{1,2}

Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region Start	Deleted Region End	Gene Strand	Description
A01	CDS	3707639	3708973	STM14_4250	3707669	3708942	-	
A02	CDS	4232846	4233580	STM14_4823	4232876	4233549	+	Putative hydrolase
A03	CDS	2784866	2785444	STM14_3168	2784896	2785413	+	Phage tail assembly-like protein
A04	CDS	3075786	3077282	STM14_3515	3075816	3077251	+	Putative permease
A05	CDS	3989635	3991017	STM14_4554	3989665	3990986	-	Putative glycosyl hydrolase
A06	CDS	2136957	2137655	STM14_2490	2136987	2137624	-	Putative DNA-binding protein
A07	CDS	1099352	1099800	STM14_1194	1099382	1099769	-	
A08	CDS	3831669	3832793	STM14_4377	3831699	3832762	+	Hypothetical protein
A10	CDS	4319606	4320268	STM14_4925	4319636	4320237	+	Putative outer membrane lipoprotein
A11	CDS	720843	721539	STM14_0765	720873	721508	-	
A12	CDS	3830286	3831611	STM14_4376	3830316	3831580	+	Putative xanthine permease
B01	CDS	3262486	3263502	STM14_3725	3262516	3263471	+	Putative zinc-binding dehydrogenase
B02	CDS	2673033	2673269	STM14_3072	2673063	2673238	+	Putative inner membrane protein
B03	CDS	3350874	3351356	STM14_3842	3350904	3351325	-	Putative inner membrane protein
B06	CDS	393112	394569	STM14_0408	393142	394571	+	Outer membrane efflux-like protein
B07	CDS	4290533	4291492	STM14_4890	4290563	4291461	-	Aminoimidazole riboside kinase
B08	CDS	1557794	1559500	STM14_1775	1557824	1559469	+	Putative periplasmic protein
B09	CDS	4420332	4420739	STM14_5033	4420362	4420708	+	Putative cytoplasmic protein
B10	CDS	4628403	4629473	STM14_5254	4628433	4629442	-	Putative potassium channel
B12	CDS	376400	377314	STM14_0388	376430	377283	-	Putative hydrolase/acyltransferase
C02	CDS	946126	946389	STM14_1023	946156	946358	+	Glutaredoxin 1
C04	CDS	4474899	4475012	STM14_5096	4474929	4474981	-	Putative cytoplasmic protein
C05	CDS	867104	867277	STM14_0927	867134	867246	-	Putative cytoplasmic protein
C06	CDS	2672447	2672584	STM14_3069	2672477	2672553	+	Putative cytoplasmic protein
C09	CDS	1659521	1660405	STM14_1893	1659551	1660374	+	Formate dehydrogenase-N beta subunit
C12	CDS	4280813	4281502	STM14_4879	4280780	4281471	-	Hypothetical protein
D02	CDS	4635030	4635779	STM14_5262	4635060	4635748	-	Esterase
D03	CDS	3765532	3766656	STM14_4313	3765562	3766625	+	Putative ABC transport protein
D04	CDS	1568776	1569810	STM14_1789	1568806	1569779	+	Putative transport protein
D05	CDS	1145850	1146290	STM14_1248	1145880	1146259	+	4-hydroxyphenylacetate catabolism
D06	CDS	2323324	2324517	STM14_2682	2323354	2324486	+	Salicylate hydroxylase
D07	CDS	3267173	3267904	STM14_3728	3267203	3267873	+	Putative outer membrane lipoprotein

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D08	CDS	1976912	1977445	STM14_2272	1976942	1977414	+	Phage-tail assembly-like protein
D09	CDS	587393	588541	STM14_0615	587423	588510	-	Glycerate kinase II
D10	CDS	589417	590652	STM14_0617	589447	590621	+	Allantoate amidohydrolase
D11	CDS	92890	94230	STM14_0094	92920	94199	-	Putative transport protein
D12	CDS	4854291	4854806	STM14_5505	4854321	4854775	+	NTPase
E01	CDS	2628108	2628911	STM14_3025	2628138	2628880	+	Putative cobalamin adenosyltransferase
E03	CDS	3098713	3099633	STM14_3542	3098743	3099554	+	Putative cytoplasmic protein
E04	CDS	10092	10805	STM14_0010	10122	10774	+	Hypothetical protein
E05	CDS	4112640	4113344	STM14_4685	4112670	4113313	+	Putative regulatory protein
E06	CDS	1980947	1981321	STM14_2280	1980977	1981290	+	Hypothetical protein
E07	CDS	1597580	1597783	STM14_1823	1597610	1597752	+	Putative cytoplasmic protein
E08	CDS	1442186	1443472	STM14_1642	1442216	1443441	+	Hypothetical protein
E09	CDS	1191175	1191714	STM14_1313	1191205	1191683	-	Hypothetical protein
E10	CDS	1555377	1556306	STM14_1773	1555407	1556275	+	DNA replication terminus site-binding protein
E11	CDS	2405236	2405838	STM14_2784	2405266	2405807	+	Cytochrome c-type protein NapC
E12	CDS	2174875	2175357	STM14_2535	2174905	2175326	-	Polyhedral body protein
F01	CDS	957281	958768	STM14_1037	957311	958737	-	Putative sulfatase
F02	CDS	1727650	1728768	STM14_1968	1727680	1728737	+	Alcohol dehydrogenase class III
F03	CDS	2163973	2165352	STM14_2523	2164003	2165321	+	Cobyrinic acid a,c-diamide synthase
F06	CDS	81732	82328	STM14_0082	81762	82297	+	Carnitine operon protein CaiE
F07	CDS	3408478	3409170	STM14_3902	3408508	3409139	-	Putative integral membrane protein
F08	CDS	1962094	1962762	STM14_2241	1962124	1962713	+	Serine/threonine protein phosphatase 1
F09	CDS	2740886	2741872	STM14_3129	2740916	2741841	+	Putative inner membrane protein
F10	CDS	4266425	4266991	STM14_4861	4266473	4266960	-	Hypothetical protein
G01	CDS	3284603	3285649	STM14_3750	3284633	3285618	+	L-asparaginase II
G02	CDS	1711019	1711558	STM14_1949	1711049	1711527	+	Ribosomal-protein-L7/L12-serine acetyltransferase
G03	CDS	2238572	2239942	STM14_2598	2238602	2239911	+	Phosphomannomutase
G04	CDS	852181	853404	STM14_0913	852211	853373	-	Imidazolonepropionase
G05	CDS	2176481	2176756	STM14_2538	2176511	2176725	-	Polyhedral body protein
G06	CDS	481716	482825	STM14_0506	481746	482794	+	2-aminoethylphosphonate transporter
G07	CDS	2410454	2410717	STM14_2789	2410484	2410686	+	Assembly protein for periplasmic nitrate reductase
G08	CDS	3543129	3543395	STM14_4056	3543159	3543364	-	Putative periplasmic protein
G09	CDS	1225307	1226260	STM14_1355	1225337	1226229	-	Flagellar hook-associated protein FigL
G10	CDS	4787291	4788661	STM14_5430	4787321	4788630	-	Putative NAD-dependent aldehyde dehydrogenase
G11	CDS	1714509	1715315	STM14_1954	1714539	1715284	-	Putative nucleoside triphosphatase
G12	CDS	4700418	4701536	STM14_5337	4700448	4701505	-	Putative selenocysteine synthase
H01	CDS	3079050	3079688	STM14_3518	3079080	3079657	+	Putative aldolase
H02	CDS	2024094	2025143	STM14_2332	2024124	2025112	+	Chemotaxis-specific methyltransferase
H03	CDS	1713182	1714495	STM14_1953	1713212	1714464	-	Putative PTS system enzyme IIC component
H04	CDS	3398411	3399058	STM14_3892	3398441	3399027	-	Putative transcriptional regulator
H05	CDS	588621	589406	STM14_0616	588651	589375	+	Hypothetical protein
H07	CDS	2038645	2039148	STM14_2350	2038675	2039117	-	Ferritin-like protein
H09	CDS	2563682	2565073	STM14_2950	2563712	2565042	-	Transporter
H10	CDS	4811388	4812398	STM14_5454	4811418	4812367	-	Putative glucosamine-fructose-6-phosphate aminotransferase
H11	CDS	975844	976812	STM14_1057	975874	976781	+	VirK-like protein
H12	CDS	419680	420849	STM14_0432	419710	420818	-	Methylcitrate synthase

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.