

Product Information Sheet for NR-42832

***Salmonella enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_043/044_Kan**

Catalog No. NR-42832

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-42832 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_043/044_Kan, NR-42832."

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_043/044_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	1614130	1615077	STM14_1845	1614160	1615046	-	Putative inner membrane protein
A03	CDS	81260	81655	STM14_0081	81290	81624	-	DNA-binding transcriptional activator CaiF
A04	CDS	1361381	1361743	STM14_1534	1361411	1361712	-	Putative cytoplasmic protein
A05	CDS	954165	955295	STM14_1033	954195	955264	-	23S rRNA methyluridine methyltransferase
A06	CDS	4531194	4531760	STM14_5144	4531137	4531729	-	Cytochrome c-type protein NrfB
A07	CDS	3305762	3307063	STM14_3781	3305792	3307032	+	Putative oxidoreductase
A08	CDS	3315674	3317146	STM14_3796	3315704	3317115	-	Putative D-mannonate oxidoreductase
A09	CDS	1367693	1368211	STM14_1552	1367723	1368180	+	Putative cytoplasmic protein
A10	CDS	2028058	2028561	STM14_2335	2028088	2028530	+	Purine-binding chemotaxis protein
A11	CDS	4524322	4525011	STM14_5138	4524352	4524980	-	Putative inner membrane protein
A12	CDS	1147851	1149317	STM14_1251	1147881	1149286	-	4-hydroxyphenylacetate catabolism
B01	CDS	446934	450074	STM14_0467	446964	450043	+	Exonuclease subunit SbcC
B02	CDS	855309	856994	STM14_0916	855339	856963	-	
B03	CDS	1896632	1898425	STM14_2162	1896662	1898394	-	Hydrogenase 1 large subunit
B04	CDS	3327038	3328741	STM14_3811	3327068	3328710	+	Hydrogenase 2 large subunit
B06	CDS	2240053	2241489	STM14_2600	2240083	2241464	+	Mannose-1-phosphate guanylyltransferase
B06	CDS	4427535	4428224	STM14_5037	4427565	4428193	+	Peptidase E
B07	CDS	402582	403835	STM14_0416	402612	403804	-	Putative inner membrane protein
B08	CDS	458766	459368	STM14_0476	458796	459337	+	Putative thiol-alkyl hydroperoxide reductase
B09	CDS	2602140	2602520	STM14_2996	2602170	2602489	-	Hypothetical protein
B10	CDS	4298615	4299658	STM14_4900	4298645	4299627	-	Putative sugar transport protein
B11	CDS	1208513	1209160	STM14_1334	1208543	1209081	+	Glutaredoxin 2
B12	CDS	2160636	2161241	STM14_2519	2160666	2161210	+	Cobalt-precorrin-6Y C(5)-methyltransferase
C01	CDS	2410707	2411198	STM14_2790	2410737	2411167	+	Ferredoxin-type protein
C02	CDS	751868	753172	STM14_0804	751898	753141	+	Citrate-proton symporter
C03	CDS	3658261	3658662	STM14_4201	3658291	3658631	+	Putative inner membrane protein
C04	CDS	1723236	1724579	STM14_1964	1723266	1724548	+	Putative cytoplasmic protein
C05	CDS	682902	683798	STM14_0720	682932	683767	+	Triphosphoribosyl-dephospho-CoA synthase
C06	CDS	1899758	1900162	STM14_2165	1899788	1900131	-	Hydrogenase-1 operon protein HyaE
C08	CDS	2149951	2150694	STM14_2505	2149981	2150663	+	Cobalamin synthase
C09	CDS	2583946	2584830	STM14_2974	2583976	2584799	+	DNA-binding transcriptional activator XapR
C10	CDS	3537695	3538963	STM14_4048	3537725	3538932	+	Putative cation transporter
C11	CDS	621022	622347	STM14_0658	621052	622316	-	Pyridine nucleotide-disulfide oxidoreductase

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C12	CDS	415505	416137	STM14_0427	415535	416106	+	Putative transport protein
D01	CDS	172523	173929	STM14_0178	172553	173898	+	Na ⁺ /galactoside symporter
D02	CDS	4532425	4533381	STM14_5146	4532455	4533350	-	Putative formate-dependent nitrate reductase
D03	CDS	2310455	2311624	STM14_2668	2310485	2311593	+	Putative ABC-type proline/glycine betaine transport system permease component
D04	CDS	3992829	3993152	STM14_4557	3992859	3993121	-	Putative DNA-binding protein
D05	CDS	2181646	2182200	STM14_2543	2181676	2182169	-	Polyhedral body protein
D06	CDS	1151517	1152308	STM14_1256	1151547	1152277	-	4-hydroxyphenylacetate catabolism
D07	CDS	4042905	4044242	STM14_4619	4042935	4044211	+	D-galactonate transport protein
D08	CDS	3741999	3742226	STM14_4285	3742029	3742195	+	Putative cytoplasmic protein
D09	CDS	945717	946094	STM14_1022	945747	946063	-	Putative inner membrane protein
D10	CDS	2324532	2325176	STM14_2683	2324562	2325145	+	Putative glutathione S-transferase
D11	CDS	2023032	2023676	STM14_2330	2023062	2023645	+	Chemotaxis regulator CheZ
D12	CDS	4361695	4363539	STM14_4965	4361725	4363508	-	Vitamin B12/cobalamin outer membrane transporter
E01	CDS	2176766	2177776	STM14_2539	2176796	2177745	-	Propanediol utilization protein
E02	CDS	619952	620806	STM14_0657	619982	620775	+	Putative transcriptional regulator
E04	CDS	1340807	1341019	STM14_1495	1340837	1340976	+	Cold shock-like protein
E06	CDS	3873664	3874650	STM14_4430	3873694	3874619	-	Putative periplasmic dicarboxylate-binding protein
E07	CDS	2470331	2470642	STM14_2849	2470361	2470611	+	Hypothetical protein
E08	CDS	594999	595808	STM14_0621	595029	595777	-	Putative cytoplasmic protein
E09	CDS	1898444	1899175	STM14_2163	1898474	1899144	-	Hydrogenase 1 b-type cytochrome subunit
E10	CDS	4267391	4268644	STM14_4863	4267421	4268508	+	Putative alcohol dehydrogenase
E11	CDS	1751106	1751516	STM14_1992	1751136	1751485	-	Heat-inducible protein
E12	CDS	4518673	4519341	STM14_5130	4518703	4519310	-	Putative glutathione S-transferase
F01	CDS (LT2) ³				1594554	1595470		
F02	CDS	2748184	2749728	STM14_3136	2748214	2749697	-	DNA-binding transcriptional activator CadC
F03	CDS	1845428	1846264	STM14_2105	1845458	1846233	+	Putative voltage-gated potassium channel
F04	CDS	2208384	2209367	STM14_2573	2208414	2209336	+	Lipopolysaccharide O-antigen chain length regulator
F05	CDS	3082844	3083248	STM14_3522	3082874	3083217	+	Putative transcriptional regulator
F06	CDS	1435026	1435217	STM14_1634	1435056	1435186	+	Hypothetical protein
F07	CDS	2810493	2810966	STM14_3194	2810523	2810914	+	Endopeptidase-like protein
F08	CDS	1074039	1074518	STM14_1169	1074069	1074487	-	Hypothetical protein
F09	CDS	1967603	1967776	STM14_2252	1967666	1967745	-	Putative cytoplasmic protein
F10	CDS	4242249	4244285	STM14_4832	4242279	4244254	+	Alpha-glucosidase
F11	CDS	1980071	1980946	STM14_2279	1980101	1980915	+	Putative inner membrane protein
F12	CDS	880293	880703	STM14_0944	880323	880672	-	Putative inner membrane protein
G01	CDS	2039327	2039965	STM14_2351	2039357	2039934	+	Hypothetical protein
G02	CDS	1345491	1345958	STM14_1509	1345521	1345927	-	Putative molecular chaperone
G03	CDS	795268	796314	STM14_0848	795298	796283	+	Putative transport protein
G04	CDS	3325453	3325701	STM14_3807	3325483	3325670	+	Hydrogenase 2 accessory protein HypG
G06	CDS	64862	66637	STM14_0065	64892	66607	+	Pyruvate carboxylase subunit B
G07	CDS	3443499	3444542	STM14_3943	3443529	3444511	-	Galactitol-1-phosphate dehydrogenase
G08	CDS	2252609	2253058	STM14_2612	2252639	2253027	+	Tyrosine phosphatase
G09	CDS	2182992	2184206	STM14_2546	2183022	2184175	-	Propionate kinase
G10	CDS	2405848	2406297	STM14_2785	2405878	2406266	+	Diheme cytochrome c550
G11	CDS	1582252	1582866	STM14_1806	1582282	1582835	+	Twin-arginine leader-binding protein DmsD
G12	CDS	4704801	4705265	STM14_5342	4704831	4705234	+	Anaerobic ribonucleotide reductase-activating protein
H01	CDS	165840	167042	STM14_0170	165870	167011	+	Type IV pilin biogenesis protein
H02	CDS	1220809	1221507	STM14_1351	1220839	1221476	-	Flagellar basal body L-ring protein
H03	CDS	3421604	3423043	STM14_3921	3421634	3422904	+	Putative transport protein
H04	CDS	1121444	1122049	STM14_1217	1121474	1122018	-	Putative DNA transformation protein
H05	CDS	3252887	3253465	STM14_3713	3252917	3253434	-	Putative inner membrane protein
H06	CDS	371424	371747	STM14_0383	371478	371716	-	Putative cytoplasmic protein
H07	CDS	4249339	4250142	STM14_4839	4249369	4250111	-	Putative glycerol-3-phosphate regulon repressor
H08	CDS	3149905	3151323	STM14_3591	3149935	3151292	-	L-fuculokinase
H09	CDS	1599665	1601707	STM14_1826	1599695	1601676	-	Dipeptidyl carboxypeptidase II

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.