

Product Information Sheet for NR-42848

***Salmonella enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_154/155_Kan**

Catalog No. NR-42848

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 3,517 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-42848 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_154/155_Kan, NR-42848."

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:

- McClelland, M., Personal Communication.

- 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.
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Table 1: *S. enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_154/155_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	4148486	4149736	STM14_4724	4148516	4149705	-	O-antigen translocase
A03	CDS	4175314	4176183	STM14_4760	4175344	4176152	-	Phospholipase A
A04	CDS	4194453	4195001	STM14_4780	4194483	4194970	-	Sec-independent translocase
A05	CDS	4217783	4218406	STM14_4806	4217813	4218375	-	Periplasmic protein disulfide isomerase I
A06	CDS	4258342	4261392	STM14_4852	4258372	4261361	+	Formate dehydrogenase alpha subunit
A07	CDS	4281960	4283333	STM14_4881	4281990	4283302	+	Two-component sensor protein
A08	CDS	4283330	4284028	STM14_4882	4283360	4283997	+	DNA-binding transcriptional regulator CpxR
A10	CDS	4288035	4288925	STM14_4888	4288065	4288894	-	CDP-diacylglycerol pyrophosphatase
A11	CDS	4309456	4310301	STM14_4913	4309486	4310270	+	Glycerol diffusion
B01	CDS	4462870	4464297	STM14_5085	4462900	4464266	+	Maltose ABC transporter periplasmic protein
B03	CDS	4472313	4472681	STM14_5093	4472343	4472650	-	Diacylglycerol kinase
B06	CDS	4669501	4670880	STM14_5304	4669531	4670849	-	UDP-N-acetylmuramate/L-alanyl-gamma-D-glutamyl-meso-diaminopimelate ligase
B07	CDS	4851868	4853805	STM14_5503	4851898	4853774	-	Lytic murein transglycosylase
B09	CDS	4857066	4857755	STM14_5509	4857096	4857724	-	DNA-binding response regulator CreB
B10	CDS	46190	47356	STM14_0048	46220	47325	-	pH-dependent sodium/proton antiporter
B11	CDS	285700	286104	STM14_0286	285730	286073	+	Outer membrane lipoprotein
C03	CDS	2898012	2898236	STM14_3301	2897886	2898195	+	Hypothetical protein
C04	ncRNA	2418383	2418563	STM14_2799	2418413	2418532	+	Regulatory RNA
C05	ncRNA	1863372	1863514	STM14_2125	1863402	1863483	-	Regulatory RNA
C06	ncRNA	1454807	1454913	STM14_1656	1454837	1454882	+	Regulatory RNA
C08	ncRNA	61461	61628	STM14_5594	61491	61597	+	
C09	CDS	2338171	2339169	STM14_2698	2338201	2339138	+	Galactose transport protein
C11	CDS	4284179	4284679	STM14_4883	4284209	4284648	-	Periplasmic repressor CpxP
C12	CDS	4869555	4870241	STM14_5521	4869585	4870210	-	Putative RNA methyltransferase
D01	CDS	4854855	4855502	STM14_5506	4854885	4855471	-	Phosphoglycerate mutase
D02	CDS	4853863	4854189	STM14_5504	4853893	4854158	-	Trp operon repressor
D03	CDS	4847109	4848491	STM14_5500	4847139	4848460	-	DNA repair protein RadA
D04	CDS	4846057	4847025	STM14_5499	4846087	4846994	-	Phosphoserine phosphatase
D05	CDS	4845296	4845940	STM14_5498	4845326	4845909	+	Hypothetical protein
D06	CDS	4837974	4838693	STM14_5490	4838004	4838662	-	Purine nucleoside phosphorylase
D07	CDS	4836541	4837764	STM14_5489	4836571	4837733	-	Phosphopentomutase
D09	CDS	4834246	4835043	STM14_5487	4834276	4835012	-	Deoxyribose-phosphate aldolase
D10	CDS	4830813	4831586	STM14_5483	4830843	4831555	-	Putative deoxyribonuclease YjjV

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Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region Start	Deleted Region End	Gene Strand	Description
D12	CDS	4812532	4814823	STM14_5456	4812562	4814792	+	Phosphoglycerol transferase I
E01	CDS	4775969	4776121	STM14_5413	4775999	4776090	-	Hypothetical protein
E02					4765118	4765157		
E03	CDS	4733281	4734792	STM14_5373	4733311	4734761	+	Leucyl aminopeptidase
E04	CDS	4732701	4733144	STM14_5372	4732731	4733152	+	DNA polymerase III subunit chi
E05	CDS	4716329	4716790	STM14_5352	4716359	4716759	+	Aspartate carbamoyltransferase regulatory subunit
E06	CDS	4712521	4715229	STM14_5349	4712503	4715198	-	Magnesium-transporting ATPase MgtA
E07	CDS	4711190	4712266	STM14_5348	4711220	4712235	+	Trehalose repressor
E08	CDS	4709635	4711052	STM14_5346	4709665	4711021	+	
E09	CDS	4707729	4707899	STM14_5344	4707759	4707868	-	Hypothetical protein
E10	CDS	4705386	4707524	STM14_5343	4705416	4707493	+	Anaerobic ribonucleoside triphosphate reductase
E11	CDS	4696110	4696496	STM14_5330	4696140	4696465	-	Cytochrome b562
E12	CDS	4694659	4696011	STM14_5329	4694620	4695980	-	Peptidase PmbA
F01	CDS	4668328	4669326	STM14_5302	4668358	4669295	+	Fructose-1,6-bisphosphatase
F02	CDS	4658032	4658670	STM14_5294	4658062	4658639	+	Methionine sulfoxide reductase A
F03	CDS	4654601	4655341	STM14_5289	4654631	4655310	-	PAPS (adenosine 3'-phosphate 5'-phosphosulfate) 3'(2'),5'-bisphosphate nucleotidase
F04	CDS	4647678	4649087	STM14_5282	4647708	4649056	-	D-alanine/D-serine/glycine permease
F05	CDS	4646651	4647382	STM14_5281	4646792	4647351	-	Peptidyl-prolyl cis-trans isomerase
F06	CDS	4640929	4641783	STM14_5269	4640959	4641752	-	L-xylulose 5-phosphate 3-epimerase
F07	CDS	4640269	4640919	STM14_5268	4640299	4640888	-	3-keto-L-gulonate-6-phosphate decarboxylase
F09	CDS	4639476	4639781	STM14_5266	4639506	4639750	-	L-ascorbate-specific enzyme IIB component of PTS
F11	CDS	4620543	4621547	STM14_5245	4620573	4621516	-	FtsH protease regulator HflC
G01	CDS	4617786	4619066	STM14_5243	4617816	4619035	-	Putative GTPase HflX
G03	CDS	4595680	4596075	STM14_5221	4595710	4596044	+	Fumarate reductase subunit C
G04	CDS	4595310	4595669	STM14_5220	4595340	4595638	+	Fumarate reductase subunit D
G05	CDS	4585553	4586029	STM14_5204	4585583	4585998	-	FxsA
G06	CDS	4583776	4585212	STM14_5202	4583806	4585181	+	Aspartate ammonia-lyase
G08	CDS	4544512	4546014	STM14_5161	4544542	4545983	-	Proline/glycine betaine transporter
G09	CDS	4543709	4544044	STM14_5159	4543739	4544013	+	Hypothetical protein
G10	CDS	4538892	4541039	STM14_5155	4538922	4541008	+	Formate dehydrogenase
G11	CDS	4536558	4537868	STM14_5151	4536588	4537837	-	Glutamate/aspartate:proton symporter
H01	CDS	4517512	4517835	STM14_5127	4517542	4517804	+	DNA-binding transcriptional regulator SoxS
H02	CDS	4482683	4484038	STM14_5106	4482713	4484007	-	Aromatic amino acid aminotransferase
H03	CDS	4481580	4482659	STM14_5105	4481610	4482628	-	Alanine racemase
H04	CDS	4479085	4480068	STM14_5103	4479115	4480037	+	Quinone oxidoreductase, NADPH-dependent
H05	CDS	4468240	4468737	STM14_5090	4468270	4468706	-	Chorismate pyruvate lyase
H06	CDS	4464422	4465531	STM14_5086	4464452	4465500	-	Maltose/maltodextrin transporter ATP-binding protein
H07	CDS	4461194	4462738	STM14_5082	4461224	4462707	+	Maltose transporter membrane protein
H08	CDS	4460289	4461179	STM14_5081	4460319	4461148	+	Maltose transporter permease
H12	CDS	4421773	4425561	STM14_5035	4421803	4425530	-	B12-dependent methionine 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.