

Product Information Sheet for NR-42834

***Salmonella enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_047/048_Kan**

Catalog No. NR-42834

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-42834 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_047/048_Kan, NR-42834."

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_047/048_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	4535593	4536213	STM14_5148	4535623	4536182	-	Formate-dependent nitrite reductase complex subunit NrfG
A02	CDS	1511938	1513311	STM14_1722	1511968	1513280	+	Multidrug efflux protein
A03	CDS	1910498	1912231	STM14_2177	1910528	1912200	-	Potassium/proton antiporter
A04	CDS	529941	533090	STM14_0559	529971	533059	+	Acridine efflux pump
A05	CDS	2072706	2073149	STM14_2394	2072736	2073118	-	Flagellar biosynthesis chaperone
A06	CDS	3488446	3488733	STM14_3993	3488476	3488702	-	DNA-binding transcriptional regulator Nlp
A07	CDS	1120718	1121227	STM14_1215	1120748	1121196	+	SOS cell division inhibitor
B01	CDS	1813838	1814602	STM14_2068	1813868	1814571	+	Phosphatidylglycerophosphatase B
B02	CDS	4310700	4310939	STM14_4915	4310730	4310908	-	Putative cytoplasmic protein
B03	CDS	2087238	2087933	STM14_2415	2087268	2087902	+	Hypothetical protein
B04	CDS	763876	766074	STM14_0818	763906	766043	+	Ornithine decarboxylase
B05	CDS	3619767	3619985	STM14_4158	3619797	3619954	-	Hypothetical protein
B06	CDS	640372	641490	STM14_0680	640402	641459	+	Carboxylate-amine ligase
C01	CDS	4137118	4138383	STM14_4712	4137148	4138352	+	ATP-dependent RNA helicase RhlB
C02	CDS	2576855	2578057	STM14_2963	2576885	2578026	-	Nucleoside transport
C03	CDS	2034258	2034686	STM14_2344	2034288	2034655	-	Universal stress protein UspC
C04	CDS	3446511	3448553	STM14_3946	3446541	3448522	-	Putative transglycosylase
C05	CDS	3893340	3893702	STM14_4447	3893370	3893671	-	Hypothetical protein
C06	CDS	3444653	3445426	STM14_3944	3444683	3445395	-	Galactitol utilization operon transcriptional repressor
C07	CDS	2067413	2067727	STM14_2388	2067443	2067696	+	Flagellar hook-basal body protein FlIE
C08	CDS	838713	839861	STM14_0899	838743	839830	+	Galactokinase
C09	CDS	82437	83222	STM14_0083	82467	83191	+	Carnitiny-CoA dehydratase
C10	CDS	3423251	3424615	STM14_3922	3423281	3424584	+	L-serine deaminase
C11	CDS	1157038	1157343	STM14_1263	1157068	1157312	+	Chaperone-modulator protein CbpM
D05	CDS	2580905	2581267	STM14_2967	2580935	2581236	-	Putative negative regulator
D08	CDS	2711622	2712053	STM14_3098	2711652	2712022	+	Nucleoside diphosphate kinase
D09	CDS	1989026	1990837	STM14_2290	1989056	1990806	+	Phosphogluconate dehydratase
E01	CDS	4122426	4122524	STM14_4697	4122456	4122493	-	ilvG operon leader peptide
E02	CDS	543575	543904	STM14_0570	543605	543873	-	Hypothetical protein
E03	CDS	3481825	3482301	STM14_3984	3481855	3482270	+	Transcription elongation factor GreA
E04	CDS	511382	511756	STM14_0537	511412	511725	-	Putative DNA uptake protein
E05	CDS	528095	528646	STM14_0556	528125	528615	+	Maltose O-acetyltransferase
E06	CDS	2835869	2836606	STM14_3238	2835899	2836575	+	Putative transferase

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E07	CDS	3377567	3377776	STM14_3871	3377597	3377745	+	Glycogen synthesis protein GlgS
E08	CDS	4455796	4456038	STM14_5076	4455823	4456007	-	Putative outer membrane protein
E09	CDS	603182	603394	STM14_0631	603212	603363	+	Hypothetical protein
E10	CDS	1869102	1872845	STM14_2132	1869132	1872814	+	Nitrate reductase 1 alpha subunit
E11	CDS	3092213	3092569	STM14_3536	3092243	3092538	+	Hypothetical protein
F01	CDS	3359706	3361055	STM14_3850	3359736	3361024	-	Sensor protein QseC
F02	CDS	3415051	3415356	STM14_3911	3415081	3415325	-	Putative inner membrane protein
F03	CDS	774532	774729	STM14_0826	774562	774590	+	K+-transporting ATPase, F subunit
F04	CDS	4011404	4011502	STM14_4579	4011434	4011471	+	IlvB operon leader peptide
F05	CDS	4515079	4515360	STM14_5123	4515109	4515329	+	Putative inner membrane protein
F06	CDS	1909487	1910401	STM14_2176	1909517	1910370	-	L,D-carboxypeptidase A
F07	CDS	2753703	2755163	STM14_3139	2753733	2755132	-	Putative di-/tripeptide transport protein
F09	CDS	1360570	1361208	STM14_1532	1360600	1361177	-	Leucine export protein LeuE
F10	CDS	4547261	4547929	STM14_5164	4547291	4547898	+	DNA-binding transcriptional regulator BasR
F11	CDS	4023608	4024027	STM14_4600	4023638	4023990	+	Heat shock protein IbpA
G01	CDS	4146672	4147349	STM14_4722	4146831	4147318	-	TDP-fucosamine acetyltransferase
G02	CDS	1944112	1944921	STM14_2217	1944142	1944890	+	23S rRNA methyltransferase A
G04	CDS	4195813	4196607	STM14_4782	4195855	4196576	-	DNase TatD
G05	CDS	98967	99497	STM14_0103	98997	99466	-	Glutathione-regulated potassium-efflux system ancillary protein KefF
G06	CDS	1430619	1431170	STM14_1628	1430649	1431139	-	Putative glutathione peroxidase
G07	CDS	3627557	3627775	STM14_4169	3627599	3627744	-	Hypothetical protein
G08	CDS	158843	159340	STM14_0161	158942	159309	-	SecA regulator SecM
G09	CDS	1602052	1602234	STM14_1829	1602082	1602203	-	Putative cytoplasmic protein
G10	CDS	3892840	3893049	STM14_4446	3892870	3893018	+	Putative cytoplasmic protein
G11	CDS	1669184	1670572	STM14_1903	1669214	1670541	-	Nitrate extrusion protein
G12	CDS	3406607	3407743	STM14_3899	3406637	3407712	+	Putative methyltransferase
H03	CDS	4724580	4725584	STM14_5364	4724610	4725553	+	Ornithine carbamoyltransferase subunit I
H04	CDS	524651	525040	STM14_0550	524681	525009	+	Putative methyltransferase
H06	CDS	1399093	1400001	STM14_1589	1399123	1399970	+	Nucleotide excision repair endonuclease
H07	CDS	4586088	4587329	STM14_5205	4586118	4587298	+	Inner membrane protein YjeH
H08	CDS	3622874	3623425	STM14_4162	3622904	3623394	+	Glutathione-regulated potassium-efflux system ancillary protein KefG
H09	CDS	210499	211728	STM14_0212	210529	211697	-	Putative xylanase/chitin deacetylase
H10	CDS	1577076	1578329	STM14_1801	1577106	1578334	+	Putative voltage-gated ClC-type chloride channel ClcB
H11	CDS	3251016	3252062	STM14_3710	3251046	3252031	+	Erythrose 4-phosphate dehydrogenase
H12	CDS	3675024	3677351	STM14_4219	3675054	3677320	-	Putative RNase R

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.