

***Salmonella enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate 015/016_Kan**

Catalog No. NR-29406

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.^{1,2} The kanamycin-resistant mutant collection contains 3,517 mutants distributed among eleven 96-well plates. In these mutants, 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} The parent strain *S. enterica* subsp. *enterica*, strain 14028s is available from BEI Resources as NR-12154.

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 [pCLF4](#) 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.

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-29406 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 015/016_Kan, NR-29406.”

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:

1. Andrews-Polymeris, H. and M. McClelland, 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.

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Table 1: *S. enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate 015/016_Kan^{1,2}

Well Position	Deleted Region of Chromosome	Deletion Start	Deletion End	Locus Tag	14028S Gene Start	14028S Gene End	14028S Gene Strand	Description
A01	chr_14028S	3710759	3711604	STM14_4252	3710729	3711634	-	Putative dihydrodipicolinate synthetase
A02	chr_14028S	3906447	3907697	STM14_4458	3906417	3907727	-	Putative permease
A04	chr_14028S	3997386	3997613	STM14_4562	3997356	3997643	+	Putative periplasmic protein
A05	chr_14028S	4065101	4066003	STM14_4640	4065071	4066033	-	Putative inner membrane protein
A06	chr_14028S	4173291	4173689	STM14_4756	4173261	4173719	+	Hypothetical protein
A07	chr_14028S	4281544	4281879	STM14_4880	4281514	4281909	-	Putative inner membrane protein
A08	chr_14028S	4327039	4327836	STM14_4930	4327009	4327866	+	Putative inner membrane protein
A09	chr_14028S	4447011	4447475	STM14_5065	4446981	4447505	+	Putative phage tail core protein
A10	chr_14028S	4572943	4573152	STM14_5185	4572913	4573182	+	Hypothetical protein
A11	chr_14028S	4672285	4673658	STM14_5308	4672255	4673688	+	Sugar transporter
A12	chr_14028S	4765528	4766193	STM14_5398	4765498	4766214	+	Hypothetical protein
B01	chr_14028S	3787107	3788024	STM14_4331	3787077	3788054	+	Putative phosphosugar isomerase
B02	chr_14028S	3922012	3923166	STM14_4474	3921982	3923196	-	O-antigen ligase
B03	chr_14028S	3930460	3931524	STM14_4483	3930430	3931554	+	Glucosyltransferase I
B04	chr_14028S	4004941	4005894	STM14_4572	4004911	4005924	+	Putative cytoplasmic protein
B05	chr_14028S	4066129	4066938	STM14_4641	4066033	4066968	-	Putative reverse transcriptase
B06	chr_14028S	4186130	4187374	STM14_4772	4186061	4187404	-	Putative arylsulfatase regulator
B07	chr_14028S	4289218	4290492	STM14_4889	4289188	4290522	-	Putative Na ⁺ /galactoside symporter
B08	chr_14028S	4329132	4330196	STM14_4932	4329102	4330226	+	Putative cytoplasmic protein
B09	chr_14028S	4449761	4450015	STM14_5069	4449731	4450045	+	Putative inner membrane protein
B10	chr_14028S	4573458	4573670	STM14_5187	4573428	4573715	+	Putative regulatory protein
B11	chr_14028S	4678321	4678563	STM14_5313	4678291	4678593	+	Putative cytoplasmic protein
B12	chr_14028S	4770304	4770981	STM14_5404	4770016	4771011	-	Putative inner membrane protein
C01	chr_14028S	3800315	3801022	STM14_4346	3800285	3801052	+	EAL domain-containing protein
C02	chr_14028S	3923283	3924368	STM14_4475	3923253	3924398	+	Putative hexose transferase
C03	chr_14028S	3931581	3932555	STM14_4484	3931551	3932585	+	Lipopolysaccharide core biosynthesis protein
C04	chr_14028S	4007313	4008173	STM14_4574	4007283	4008203	+	Putative sugar kinase
C05	chr_14028S	4078515	4079828	STM14_4653	4078485	4079858	+	Putative phosphotransferase system fructose-specific component IIB
C06	chr_14028S	4200603	4201601	STM14_4787	4199808	4201631	-	Arylsulfotransferase
C07	chr_14028S	4291532	4292512	STM14_4891	4291502	4292542	-	Putative ADP-ribosylglycohydrolase
C08	chr_14028S	4387889	4388152	STM14_4992	4387859	4388182	-	Putative inner membrane protein
C09	chr_14028S	4451703	4451846	STM14_5073	4451529	4451876	-	Putative cytoplasmic protein
C10	chr_14028S	4574882	4575043	STM14_5190	4574852	4575073	+	Putative cytoplasmic protein
C11	chr_14028S	4679845	4680690	STM14_5316	4679815	4680720	-	Putative endonuclease
C12	chr_14028S	4771317	4772168	STM14_5405	4771287	4772198	-	Putative cytoplasmic protein
D01	chr_14028S ³	3832973	3833920	STM14_4380	3832943	3833950	-	Putative regulatory protein
D02	chr_14028S	3924529	3925278	STM14_4476	3924499	3925308	+	Lipopolysaccharide core biosynthesis protein
D03	chr_14028S	3982266	3983315	STM14_4544	3982236	3983345	+	Putative selenocysteine synthase

Well Position	Deleted Region of Chromosome	Deletion Start	Deletion End	Locus Tag	14028S Gene Start	14028S Gene End	14028S Gene Strand	Description
D06	chr_14028S	4235713	4236516	STM14_4826	4235683	4236546	+	Putative membrane-associated metal-dependent hydrolase
D09	chr_14028S	4529431	4529517	STM14_5142	4529401	4529547	-	Putative cytoplasmic protein
D10	chr_14028S	4576449	4577141	STM14_5193	4576419	4577171	+	Non-specific acid phosphatase
D11	chr_14028S	4684050	4685156	STM14_5320	4684020	4685186	+	Hypothetical protein
D12	chr_14028S	4776504	4776890	STM14_5414	4776390	4776920	-	Putative hyperinvasive locus E
E01	chr_14028S	3853004	3853279	STM14_4405	3852974	3853309	+	
E02	chr_14028S	3925488	3926126	STM14_4477	3925458	3926156	+	Lipopolysaccharide core biosynthesis protein
E03	chr_14028S	3989266	3989499	STM14_4552	3989236	3989529	-	Putative inner membrane protein
E04	chr_14028S ⁴	4014522	4015496	STM14_4587	4014492	4015577	+	Putative periplasmic protein
E05	chr_14028S	4129389	4129646	STM14_4703	4129359	4129676	-	Putative cytoplasmic protein
E06	chr_14028S	4238624	4239256	STM14_4829	4238594	4239286	+	Outer membrane porin L
E07	chr_14028S	4299685	4300626	STM14_4901	4299655	4300656	-	Putative sugar transport protein
E08	chr_14028S ⁵	4430284	4431165	STM14_5043	4430491	4431195	+	
E09	chr_14028S	4551210	4551911	STM14_5167	4551180	4551941	+	Transcriptional activator
E10	chr_14028S	4590263	4591060	STM14_5210	4590233	4591090	+	Putative inner membrane protein
E11	chr_14028S	4718670	4720013	STM14_5357	4718640	4720043	+	Putative arginine repressor
E12	chr_14028S	4777189	4777863	STM14_5417	4777159	4777893	-	Putative aspartate racemase
F01	chr_14028S	3872405	3873622	STM14_4429	3872375	3873652	-	Hypothetical protein
F02	chr_14028S	3926209	3927159	STM14_4478	3926179	3927189	+	Lipopolysaccharide glucosyltransferase
F03	chr_14028S	3993273	3993482	STM14_4558	3993243	3993512	+	Putative phosphotransferase system HPr protein
F04	chr_14028S	4020934	4021125	STM14_4596	4020868	4021155	-	
F05	chr_14028S	4129703	4129984	STM14_4704	4129673	4130014	-	Putative cytoplasmic protein
F06	chr_14028S	4246477	4247295	STM14_4835	4246447	4247325	+	Putative aldolase
F07	chr_14028S	4300715	4301677	STM14_4902	4300685	4301707	-	Putative sugar transport protein
F08	chr_14028S	4431487	4432155	STM14_5045	4431457	4432185	+	Putative cytoplasmic protein
F09	chr_14028S	4562166	4562372	STM14_5175	4562136	4562402	+	Putative cytoplasmic protein
F10	chr_14028S	4601713	4601967	STM14_5227	4601683	4601997	-	Putative inner membrane protein
F11	chr_14028S	4727254	4727682	STM14_5368	4727224	4727712	+	Putative inner membrane protein
F12	chr_14028S	4789135	4789776	STM14_5433	4789105	4789806	+	Putative inner membrane protein
G01	chr_14028S	3903854	3904798	STM14_4455	3903824	3904828	+	Putative transcriptional regulator
G03	chr_14028S	3994475	3995899	STM14_4560	3994445	3995929	+	Putative sugar kinase
G04	chr_14028S	4044357	4045445	STM14_4620	4044327	4045475	+	2-oxo-3-deoxygalactonate 6-phosphate aldolase
G05	chr_14028S	4163281	4163763	STM14_4742	4163251	4163793	-	Putative inner membrane protein
G06	chr_14028S	4262721	4263692	STM14_4855	4262691	4263722	+	Putative inner membrane lipoprotein
G07	chr_14028S	4302721	4302954	STM14_4904	4302691	4302984	-	Autoinducer-2 (AI-2) modifying protein LsrG
G08	chr_14028S	4432412	4432642	STM14_5046	4432382	4432672	+	Putative inner membrane protein
G09	chr_14028S	4565530	4567821	STM14_5178	4565422	4567851	-	Putative anaerobic dimethylsulfoxide reductase subunit A
G10	chr_14028S	4608265	4608945	STM14_5232	4608235	4608975	+	Putative arginine-binding periplasmic protein
G11	chr_14028S	4740826	4741797	STM14_5380	4740796	4741827	+	L-idonate 5-dehydrogenase
G12	chr_14028S	4821909	4822913	STM14_5467	4821879	4822943	+	Hypothetical protein
H01	chr_14028S	3905177	3906313	STM14_4457	3905147	3906343	-	Putative mandelate racemase
H02	chr_14028S	3928256	3929275	STM14_4480	3928226	3929305	+	UDP-D-galactose:(glucosyl)lipopolysaccharide-1, 6-D-galactosyltransferase
H03	chr_14028S	3995952	3997250	STM14_4561	3995922	3997280	+	Putative PTS system galactitol-specific enzyme IIC component
H04	chr_14028S	4049079	4050281	STM14_4625	4049049	4050311	-	Putative permease
H05	chr_14028S	4163835	4164137	STM14_4743	4163805	4164167	-	Putative cytoplasmic protein
H06	chr_14028S	4263977	4264975	STM14_4857	4263947	4265005	-	Putative cytoplasmic protein
H07	chr_14028S	4320499	4322229	STM14_4926	4320469	4322259	+	Putative arylsulfate sulfotransferase
H08	chr_14028S	4440488	4440790	STM14_5056	4440458	4440820	+	Putative phage glucose translocase
H10	chr_14028S	4630585	4631163	STM14_5257	4630555	4631193	-	Putative inner membrane protein
H11	chr_14028S	4756125	4759742	STM14_5392	4756095	4759772	+	Putative type II restriction enzyme methylase subunit
H12	chr_14028S	4866596	4867207	STM14_5517	4866566	4867237	+	Putative inner membrane protein

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.

³Deleted region also overlaps STM14_4379 (2.5%)

⁴Deleted region also overlaps STM14_4586 (3.7%)

⁵Deleted region also overlaps STM14_5042 (82.8%)