

***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 use in humans.

Contributor:

Helene Andrews-Polymeris, Associate Professor, Department of Microbial Pathogenesis and Immunology, College of Medicine, Texas A&M Health Science Center, Bryan, Texas, USA and 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 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.^{2,3,4} 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 1 day.

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: 1

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. 6th ed. Washington, DC: U.S. Government Printing Office, 2020; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

Disclaimers:

You are authorized to use this product for research use only. It is not intended for human use.

Use of this product is subject to the terms and conditions of the BEI Resources Material Transfer Agreement (MTA). The MTA is available on our Web site at www.beiresources.org.

While BEI Resources uses reasonable efforts to include accurate and up-to-date information on this product sheet, neither ATCC® nor the U.S. Government makes any warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. Neither ATCC® nor the U.S. Government warrants that such information has been confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling, use and disposal. ATCC® and the U.S. Government are not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to ensure authenticity and reliability of materials on deposit, the U.S. Government, ATCC®, their suppliers and contributors to BEI Resources are not liable for

damages arising from the misidentification or misrepresentation of products.

Use Restrictions:

This material is distributed for internal research, non-commercial purposes only. This material, its product or its derivatives may not be distributed to third parties. Except as performed under a U.S. Government contract, individuals contemplating commercial use of the material, its products or its derivatives must contact the contributor to determine if a license is required. U.S. Government contractors may need a license before first commercial sale.

References:

1. Andrews-Polymenis, 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.

ATCC® is a trademark of the American Type Culture Collection.



BEI Resources is committed to ensuring digital accessibility for people with disabilities. This Product Sheet contains complex tables and may not be fully accessible. Please let us know if you encounter accessibility barriers and a fully accessible document will be provided:

E-mail: Contact@BEIResources.org. We try to respond to feedback within 24 hours.

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

Well Position	Deleted Region of Chromosome	Deletion Start	Deletion End	Locus Tag	14028S Gene Start	14028S Gene End	14028S Gene Strand	Description
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
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%)