

***Salmonella enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_063/064_Kan**

Catalog No. NR-42842

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 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-42842 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_063/064_Kan, NR-42842."

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/bmb15/index.htm.

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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_063/064_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	3623691	3625598	STM14_4164	3623721	3625567	-	Putative ABC transporter ATP-binding protein
A02	CDS	3626493	3627560	STM14_4168	3626523	3627529	-	Putative hydrolase
A03	CDS	3620858	3621058	STM14_4160	3620888	3621027	+	Putative cytoplasmic protein
A04	CDS	3628792	3629196	STM14_4171	3628822	3629165	+	Hypothetical protein
A05	CDS	3635930	3637111	STM14_4181	3635960	3637080	-	Hypothetical protein
A06	CDS	3630185	3632272	STM14_4174	3630215	3632241	-	Putative inner membrane protein
A07	CDS	3682594	3683277	STM14_4227	3682624	3683246	-	Gluconate periplasmic binding protein
A08	CDS	3683336	3683911	STM14_4228	3683366	3683880	-	Putative DNA uptake protein
A09	CDS	3754045	3754641	STM14_4300	3754075	3754610	-	16S rRNA m(2)G966-methyltransferase
A10	CDS	3754628	3754900	STM14_4301	3754658	3754869	-	Hypothetical protein
B01	CDS	3760622	3761287	STM14_4307	3760652	3761256	-	Hypothetical protein
B02	CDS	3763270	3764319	STM14_4310	3763300	3764288	-	Putative permease
B03	CDS	3726991	3727686	STM14_4267	3727021	3727655	+	Putative cytoplasmic protein
B04	CDS	3727810	3728847	STM14_4268	3727840	3728816	+	Putative dehydrogenase
B05	CDS	3770767	3771963	STM14_4317	3770797	3771887	+	Putative periplasmic protein
B06	CDS	3775312	3776784	STM14_4321	3775342	3776753	-	Inner membrane transporter YhiP
B07	CDS	3776876	3777634	STM14_4322	3776906	3777588	+	Putative methyltransferase
B08	CDS	3781347	3782189	STM14_4326	3781377	3782158	-	Putative cytoplasmic protein
B09	CDS	3795374	3796402	STM14_4342	3795404	3796371	-	Putative tRNA-processing ribonuclease
B11	CDS	3798115	3800175	STM14_4345	3798145	3800144	+	Putative inner membrane protein
B12	CDS	3802264	3803751	STM14_4349	3802294	3803720	+	Putative Zn-dependent peptidase
C01	CDS	3805415	3807388	STM14_4353	3805445	3807357	+	Putative phosphodiesterase
C02	CDS	3812232	3814532	STM14_4357	3812262	3814501	+	Cellulose synthase regulator protein
C03	CDS	3814543	3817167	STM14_4358	3814573	3817136	+	Cellulose synthase catalytic subunit
C04	CDS	3820121	3821800	STM14_4364	3820151	3821769	-	Putative inner membrane protein
C05	CDS	3834277	3835968	STM14_4382	3834307	3835937	+	Phosphoethanolamine transferase
C06	CDS	3843602	3844042	STM14_4391	3843632	3844011	-	Hypothetical protein
C07	CDS	3846497	3847159	STM14_4394	3846527	3847128	-	Putative outer membrane lipoprotein
C09	CDS	3848402	3849112	STM14_4397	3848432	3849081	+	Putative outer membrane lipoprotein
C10	CDS	3887874	3888482	STM14_4442	3887904	3888451	+	Glutathione S-transferase
C11	CDS	3903283	3903756	STM14_4454	3903313	3903725	-	Putative tRNA/rRNA methyltransferase YibK
C12	CDS	3910577	3911008	STM14_4463	3910607	3910977	+	Putative rhodanese-like sulfurtransferase

Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region Start	Deleted Region End	Gene Strand	Description
D02	CDS	3941213	3942076	STM14_4498	3941243	3942045	-	Hypothetical protein
D04	CDS	3958268	3960586	STM14_4515	3958298	3960555	+	Alpha-xylosidase YicI
D05	CDS	3960598	3961980	STM14_4516	3960628	3961949	+	Putative transporter
D06	CDS	3979384	3980286	STM14_4541	3979414	3980255	-	Putative permease
D08	CDS	4021219	4022880	STM14_4597	4021249	4022849	+	Hypothetical protein
D11	CDS	4024665	4025891	STM14_4603	4024695	4025860	+	Putative cytoplasmic protein
D12	CDS	4068926	4069885	STM14_4643	4068956	4069854	-	DNA-binding transcriptional regulator YidZ
E01	CDS	4070816	4071382	STM14_4645	4070828	4071351	-	Putative oxidoreductase
E02	CDS	4071427	4072890	STM14_4646	4071457	4072859	+	Putative xanthine/uracil permease family protein
E05	CDS	4101443	4103047	STM14_4676	4101473	4102908	+	Regulatory ATPase RavA
E06	CDS	4111201	4112628	STM14_4684	4111231	4112597	+	Putative transport protein
E07	CDS	4119236	4120084	STM14_4693	4119266	4120053	+	Transcriptional regulator HdfR
E08	CDS	4153111	4154496	STM14_4728	4153141	4154465	-	Putative transport protein YifK
E09	CDS	4166774	4167481	STM14_4749	4166804	4167450	-	Hypothetical protein
E10	CDS	4168380	4169096	STM14_4751	4168410	4169065	-	Flavin mononucleotide phosphatase
E11	CDS	3944234	3944851	STM14_4502	3944264	3944820	-	Putative inner membrane protein
F01	CDS	4174682	4175149	STM14_4758	4174712	4175136	+	Hypothetical protein
F02	CDS	4180583	4181383	STM14_4765	4180613	4181352	-	Putative sugar phosphatase
F03	CDS	4181464	4182363	STM14_4766	4181494	4182332	-	Putative transport protein
F04	CDS	4189456	4190886	STM14_4775	4189501	4190855	-	DNA recombination protein RmuC
F05	CDS	3914097	3915059	STM14_4466	3914127	3915028	-	Putative periplasmic protein
F06	CDS	4206711	4207325	STM14_4791	4206741	4207294	-	Hypothetical protein
F07 ³	CDS	4222836	4223468	STM14_4812	4222866	4223401	+	GTPase EngB
F08	CDS	4216780	4217766	STM14_4805	4216810	4217735	-	Serine/threonine protein kinase
F10	CDS	4224051	4224566	STM14_4814	4224081	4224535	-	Hypothetical protein
F11	CDS	4250333	4250932	STM14_4840	4250363	4250901	-	Phosphatase
G01	CDS	4252229	4253218	STM14_4843	4252259	4253187	-	Putative acetyltransferase
G03	CDS	4284827	4285729	STM14_4884	4284857	4285698	-	Ferrous iron efflux protein F
G04	CDS	4304730	4305326	STM14_4907	4304760	4305295	+	Putative periplasmic protein
G05	CDS	4305427	4305855	STM14_4908	4305457	4305824	-	Putative inner membrane protein
G06	CDS	4359164	4359799	STM14_4962	4359197	4359768	-	DNA-binding transcriptional repressor FabR
G07	CDS	4359815	4360174	STM14_4963	4359845	4360143	-	Hypothetical protein
G08	CDS	4413786	4414223	STM14_5027	4413816	4414192	+	Hypothetical protein
G09	CDS	4396912	4397685	STM14_5007	4396942	4397654	-	NADH pyrophosphatase
G10	CDS	4399513	4400103	STM14_5010	4399543	4400072	-	Putative cytoplasmic protein
G12	CDS	4459713	4460123	STM14_5080	4459743	4460092	-	Phosphate-starvation-inducible protein PsiE
H01	CDS	4425828	4427459	STM14_5036	4425858	4427428	-	Putative transport protein
H02	CDS	4429884	4430156	STM14_5041	4429914	4430125	+	Hypothetical protein
H03	CDS	4477502	4478500	STM14_5100	4477532	4478469	-	tRNA-dihydrouridine synthase A
H04	CDS	4478668	4478910	STM14_5102	4478698	4478879	-	Phage shock protein G
H06	CDS	4519688	4521037	STM14_5133	4519718	4521006	-	Hypothetical protein
H07	CDS	4521187	4522833	STM14_5135	4521217	4522802	-	Na/H transport protein
H11	CDS	4598926	4599903	STM14_5224	4598956	4599872	-	Lysyl-tRNA synthetase

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.

³Mutant confirmed; duplication detected