Salmonella enterica subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_055/056_Kan

Catalog No. NR-42838

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 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-42838 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_055/056_Kan, NR-42838."

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. 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.

Support Provided by NIAID

Table 1: *S. enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_055/056_Kan[1,2]

Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region in Start	Deleted Region in End	Gene Strand	Description
A01	CDS	10841	11245	STM14_0011	10871	11214	+	Hypothetical protein
A05	CDS	124122	124829	STM14_0125	124152	124798	+	Thiamine transporter ATP-binding subunit
A06	CDS	196688	197035	STM14_0199	196718	197004	+	Putative periplasmic protein
A07	CDS	163177	163920	STM14_0167	163207	163889	+	Hypothetical protein
A08	CDS	197237	198847	STM14_0200	197267	198816	-	Multicopper oxidase
A09	CDS	190308	190670	STM14_0192	190338	190639	-	Hypothetical protein
A12	CDS	202886	203812	STM14_0204	202916	203781	-	Putative ABC-type multidrug transport system ATPase component
B02	CDS	209997	210437	STM14_0211	210027	210406	-	Putative PTS enzyme
B05	CDS	241313	241936	STM14_0245	241343	241905	+	Hypothetical protein
B09	CDS	287077	287730	STM14_0288	287107	287699	+	DL-methionine transporter permease subunit
B11	CDS	248022	248408	STM14_0251	248052	248377	+	Hypothetical protein
C02	CDS	280998	281216	STM14_0280	281028	281185	+	Hypothetical protein
C05	CDS	363911	365155	STM14_0374	363941	365124	-	Fermentation/respiration switch protein
C07	CDS	353282	355726	STM14_0365	353312	355695	+	Acyl-CoA dehydrogenase
C12	CDS	437792	438058	STM14_0452	437822	438027	-	Hypothetical protein
D07	CDS	500812	501390	STM14_0525	500842	501413	+	Hypothetical protein
D08	CDS	488510	489001	STM14_0514	488522	488970	-	Nucleotide-binding protein
D10	CDS	556097	556576	STM14_0583	556127	556545	+	Hypothetical protein
D12	CDS	516834	517418	STM14_0543	516990	517387	-	Putative transcriptional regulator
E01	CDS	511860	512258	STM14_0538	511890	512227	-	Putative esterase
E02	CDS	512364	513059	STM14_0539	512394	513028	+	Queuosine biosynthesis protein QueC
E03	CDS	567133	567819	STM14_0595	567163	567788	-	Putative ABC transporter ATP-binding protein YbbA
E04	CDS	561250	561666	STM14_0587	561280	561635	-	DNA-binding transcriptional regulator CueR
E05	CDS	561667	562119	STM14_0588	561697	562088	+	Hypothetical protein
E06	CDS	562116	563033	STM14_0589	562146	563002	+	Putative inner membrane protein
E07	CDS	563844	564623	STM14_0591	563874	564592	-	Putative transport protein
E09	CDS	565623	566393	STM14_0593	565653	566362	+	Short chain dehydrogenase
E10	CDS	567816	570230	STM14_0596	567846	570199	-	Putative inner membrane protein
E12	CDS	661373	661786	STM14_0697	661403	661755	-	Hypothetical protein
F01	CDS	626329	627576	STM14_0664	626359	627545	+	Hypothetical protein
F02	CDS	664370	665458	STM14_0701	664400	665427	+	Hypothetical protein
F03	CDS	665585	666745	STM14_0702	665615	666714	-	Putative aminotransferase

Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region in Start	Deleted Region in End	Gene Strand	Description
F04	CDS	677868	678296	STM14_0713	677898	678265	+	Putative universal stress protein
F07	CDS	698525	698788	STM14_0743	698555	698757	+	Hypothetical protein
F08	CDS	726055	726990	STM14_0769	726085	726959	+	Ribonucleoside hydrolase 1
F09	CDS	717062	717535	STM14_0761	717092	717504	-	Hypothetical protein
F11	CDS	732227	733105	STM14_0776	732257	733074	+	Putative transport protein
G01	CDS	758318	758599	STM14_0811	758336	758568	+	LexA regulated protein
G02	CDS	758733	759503	STM14_0813	758763	759472	+	Hypothetical protein
G03	CDS	750031	751437	STM14_0802	750061	751406	-	Putative outer membrane protein
G04	CDS	818322	819110	STM14_0872	818352	819079	-	Hypothetical protein
G05	CDS	778545	779288	STM14_0830	778575	779257	-	Putative hydrolase-oxidase
G06	CDS	779304	779960	STM14_0831	779334	779929	-	Putative carboxylase
G07	CDS	779954	780886	STM14_0832	779984	780855	-	Putative carboxylase
G08	CDS	780876	781610	STM14_0833	780906	781579	-	Hypothetical protein
G09	CDS	822359	823297	STM14_0882	822389	823266	+	Zinc transporter ZitB
G10	CDS	848539	849357	STM14_0910	848569	849326	+	Phosphotransferase
G11	CDS	858601	859077	STM14_0918	858631	859046	+	Putative kinase inhibitor protein
G12	CDS	850660	851943	STM14_0912	850690	851912	+	Putative pectinesterase
H01	CDS	849521	850516	STM14_0911	849551	850485	-	6-phosphogluconolactonase
H02	CDS	883010	884746	STM14_0948	883040	884715	+	Putative ABC-type multidrug transport system ATPase component
H03	CDS	869674	870582	STM14_0929	869704	870551	+	Putative cytoplasmic protein
H04	CDS	873730	874434	STM14_0935	873760	874403	-	Putative permease
H05	CDS	877202	878164	STM14_0941	877232	878133	+	Hypothetical protein
H07	CDS	880665	881771	STM14_0945	880695	881740	+	Putative transport protein
H08	CDS	881887	883017	STM14_0947	881917	882986	+	Putative transport protein
H09	CDS	890384	891358	STM14_0954	890414	891327	-	Hypothetical protein
H10	CDS	885734	886408	STM14_0950	885764	886377	+	Putative DNA-binding transcriptional regulator
H11	CDS	891514	891774	STM14_0955	891544	891743	+	Hypothetical protein
H12	CDS	892530	893456	STM14_0959	892560	893425	-	Putative SAM-dependent methyltransferase

[1]All information in this table was provided by the depositor at the time of deposition.
[2]Construction 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.