Salmonella enterica subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_041/042_Kan

Catalog No. NR-42831

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-42831 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_041/042_Kan, NR-42831."

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_041/042_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	3317158	3318570	STM14_3797	3317188	3318538	-	Glucuronate isomerase
A02	CDS	3870340	3870804	STM14_4426	3870370	3870772	-	Putative cytoplasmic protein
A03	CDS	1728800	1729075	STM14_1969	1728830	1729043	+	Putative cytoplasmic protein
A04	CDS	3144314	3145462	STM14_3586	3144344	3145430	+	L-1,2-propanediol oxidoreductase
A06	CDS (LT2)				4688762	4689509		
A07	CDS	3325729	3326070	STM14_3808	3325759	3326038	+	Hydrogenase nickel incorporation protein HybF
A08	CDS	3303665	3305164	STM14_3777	3303695	3305132	+	Putative amino acid transporter
A09	CDS	683770	684321	STM14_0721	683800	684289	+	2'-(5''-triphosphoribosyl)-3'-dephospho-CoA:apo-citrate lyase
A10	CDS (LT2)				619767	619869		
A11	CDS	2154521	2155168	STM14_2511	2154551	2155226	+	Cobalt transport protein CbiM
A12	CDS	4046073	4046951	STM14_4622	4046103	4046919	+	2-oxo-3-deoxygalactonate kinase
B01	CDS	3694238	3695257	STM14_4237	3694268	3695225	+	RNA 3'-terminal-phosphate cyclase
B02	CDS	297612	298787	STM14_0302	297642	298755	-	Putative drug efflux protein
B03	CDS	4247348	4248244	STM14_4836	4247378	4248212	+	Putative oxidoreductase
B04	CDS	2235513	2236991	STM14_2596	2235543	2236959	+	Colanic acid exporter
B05	CDS	751487	751819	STM14_0803	751517	751787	-	Putative lipoprotein
B06	CDS	2448422	2449711	STM14_2827	2448452	2449679	+	Putative transport protein
B07	CDS	1612621	1613547	STM14_1843	1612663	1613515	-	Glutaminase
B08	CDS	2863295	2864203	STM14_3269	2863325	2864171	+	Putative cytoplasmic protein
B09	CDS	3747983	3748117	STM14_4293	3748013	3748085	-	Putative cytoplasmic protein
B11	CDS	4769156	4769764	STM14_5403	4769186	4769732	-	Putative cytoplasmic protein
B12	CDS	590674	591723	STM14_0618	590704	591691	+	Ureidoglycolate dehydrogenase
C01	CDS	4697810	4698586	STM14_5334	4697840	4698554	-	Putative inner membrane protein
C02	CDS	3695261	3696478	STM14_4238	3695291	3696446	+	Putative cytoplasmic protein
C03	CDS	1721679	1723187	STM14_1963	1721709	1723155	+	Putative carboxylesterase
C04	CDS	2650628	2652646	STM14_3048	2650658	2652614	+	Putative acetyltransferase
C05	CDS	409459	410862	STM14_0422	409489	410830	-	Cytochrome BD2 subunit I
C06	CDS	614444	615111	STM14_0648	614923	615079	+	
C07	CDS	1248928	1249302	STM14_1380	1248958	1249270	-	Putative outer membrane lipoprotein
C08	CDS	3200768	3201631	STM14_3647	3200798	3201599	+	Putative transcriptional regulator
C09	CDS	183755	184546	STM14_0188	183785	184514	+	Putative periplasmic protein
C10	CDS	1141981	1142661	STM14_1242	1142011	1142695	+	Transcriptional regulatory protein YedW
C11	CDS	2558813	2560060	STM14_2947	2558843	2560028	+	Activator
C12	CDS	2245871	2246617	STM14_2606	2245901	2246585	+	Glycosyl transferase

Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region Start	Deleted Region End	Gene Strand	Description
D01	CDS	4810309	4811370	STM14_5453	4810339	4811338	-	Putative glucosamine-fructose-6-phosphate aminotransferase
D02	CDS	2930916	2931215	STM14_3335	2930946	2931183	-	Putative transposase
D03	CDS	622753	623313	STM14_0661	622783	623281	-	Putative inner membrane protein
D04	CDS	3078269	3079045	STM14_3517	3078299	3079013	+	Hypothetical protein
D05	CDS	3876147	3876809	STM14_4432	3876177	3876777	-	3-keto-L-gulonate-6-phosphate decarboxylase
D06	CDS	4778926	4780098	STM14_5419	4778956	4780066	+	Isoaspartyl dipeptidase
D07	CDS	987674	988585	STM14_1074	987704	988553	-	Putative transcriptional regulator
D08	CDS	1589525	1589833	STM14_1812	1589555	1589801	+	Putative outer membrane protein
D09	CDS	1605806	1606708	STM14_1835	1605836	1606676	-	O-acetylserine/cysteine export protein
D10	CDS	4047898	4048743	STM14_4624	4047928	4048711	+	Sugar phosphatase
D11	CDS	1678352	1679197	STM14_1910	1678382	1679165	+	Putative arylamine N-acetyltransferase
D12	CDS	2154238	2154519	STM14_2510	2154268	2154487	+	Cobalt transport protein CbiN
E01	CDS	584652	586013	STM14_0613	584682	585981	-	Allantoinase
E02	CDS	576697	577179	STM14_0605	576727	577147	-	Ureidoglycolate hydrolase
E03	CDS	4828712	4829329	STM14_5480	4828742	4829297	-	Periplasmic protein
E04	CDS	1467965	1468966	STM14_1669	1467995	1468934	-	Hypothetical protein
E05	CDS	4245190	4246431	STM14_4834	4245220	4246399	+	Putative isomerase
E06	CDS	3769394	3770461	STM14_4315	3769424	3770429	+	Hypothetical protein
E07	CDS	2231580	2232983	STM14_2593	2231610	2232951	+	Putative colanic acid biosynthesis protein
E08	CDS	2234211	2235491	STM14_2595	2234241	2235459	+	Putative pyruvyl transferase
E09	CDS	2624099	2624938	STM14_3020	2624129	2624906	+	Ethanolamine utilization protein
E10	CDS	2320382	2321818	STM14_2679	2320412	2321786	+	Multidrug resistance outer membrane protein MdtQ
E11	CDS	4019549	4020079	STM14_4592	4019579	4020047	+	Putative cytoplasmic protein
E12	CDS	482831	483844	STM14_0507	482861	483812	+	2-aminoethylphosphonate transporter
F01	CDS	183433	183717	STM14_0187	183463	183685	-	Putative outer membrane protein
F02	CDS	4788866	4789030	STM14_5432	4788896	4788998	-	Putative cytoplasmic protein
F03	CDS	4338076	4340577	STM14_4941	4338106	4340545	+	PEP-protein phosphotransferase
F04	CDS	1603308	1603700	STM14_1832	1603338	1603668	-	Hypothetical protein
F05	CDS	592054	593718	STM14_0619	592084	593686	-	Membrane protein FdrA
F06	CDS	61794	63413	STM14_0063	61824	63381	+	Putative transcriptional regulator
F07	CDS	3083419	3084012	STM14_3523	3083449	3083980	-	Putative flavoprotein
F08	CDS	4254073	4255002	STM14_4846	4254103	4254970	+	Putative acetyl esterase
F10	CDS	4780569	4781453	STM14_5421	4780599	4781421	+	Putative inner membrane protein
F11	CDS	3518337	3518804	STM14_4026	3518367	3518772	+	Putative cytoplasmic protein
F12	CDS	4704513	4704797	STM14_5341	4704543	4704765	-	Putative inner membrane protein
G02	CDS	1364412	1364759	STM14_1544	1364442	1364727	+	Putative cytoplasmic protein
G03	CDS	4335406	4336029	STM14_4937	4335436	4335997	+	Putative periplasmic protein
G04	CDS	1655158	1655373	STM14_1887	1655188	1655341	-	Biofilm-dependent modulation protein
G05	CDS	3253466	3254173	STM14_3714	3253496	3254141	-	Putative ABC-type cobalt transport system permease component
G06	CDS	4523814	4524329	STM14_5137	4523949	4524297	-	LrgA family protein
G07	CDS	4701533	4702273	STM14_5338	4701563	4702131	-	4-hydroxy-2-oxoglutarate aldolase
G08	CDS	4549703	4551040	STM14_5166	4549733	4551008	+	Arginine:agmatin antiporter
G09	CDS	1919842	1920183	STM14_2185	1919872	1920151	-	Putative cytoplasmic protein
G10	CDS	3198201	3199037	STM14_3644	3198231	3199005	+	5-keto-4-deoxyuronate isomerase
G11	CDS	1751742	1752041	STM14_1994	1751772	1752009	-	Hypothetical protein
G12	CDS	681401	682864	STM14_0719	681431	682832	+	Citrate/succinate transport antiport protein
H02	CDS	4522928	4523815	STM14_5136	4522958	4523783	+	Putative transcriptional regulator
H03	CDS	1944987	1946732	STM14_2218	1945017	1946700	+	Putative penicillin-binding protein 3
H05	CDS	3098420	3098716	STM14_3541	3098450	3098681	+	Hypothetical protein
H06	CDS	4804900	4807665	STM14_5448	4804930	4807633	-	Putative transcriptional regulator
H07	CDS	4697123	4697485	STM14_5332	4697153	4697453	-	Putative cytoplasmic protein
H08	CDS	4704281	4704523	STM14_5340			-	Bifunctional antitoxin/transcriptional repressor RelB
H09	CDS	4820361	4820819	STM14_5465	4820391	4820787	+	Putative cytoplasmic protein
H10	CDS	1635982	1636371	STM14_1869	1636012	1636339	+	Putative translation initiation inhibitor
H11	CDS	2063072	2064556	STM14_2383	2063102	2064524	-	Cytoplasmic alpha-amylase
H12	CDS	4390603	4391736	STM14_5000	4390633	4391704	+	Thiamine biosynthesis protein ThiH

[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.

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