Product Information Sheet for NR-42824

Salmonella enterica subsp. enterica, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_027/028_Kan

Catalog No. NR-42824

For research use only. Not for use in humans.

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.[2,3,4] 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-42824 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 SGD_027/028_Kan, NR-42824."

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 (BMBL). 6th ed. Washington, DC: U.S. Government Printing Office, 2020.

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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_027/028_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	CDS	3372754	3374550	STM14_3866	3373786	3374519	-	Putative arylsulfate sulfotransferase
A02	CDS	3077279	3078229	STM14_3516	3077309	3078198	+	Putative nucleoside-diphosphate-sugar epimerase
A03	CDS	2535147	2536148	STM14_2919	2535177	2536117	-	Flagella biosynthesis regulator
A04	CDS	538624	539559	STM14_0564	538654	539528	+	Putative transposase
A05	CDS	1158498	1158860	STM14_1266	1158528	1158829	-	Suppression of copper sensitivity protein A
A06	CDS	2088025	2088324	STM14_2416	2088055	2088260	+	Putative inner membrane protein
A07	CDS	4567865	4568491	STM14_5179	4567895	4568460	-	Putative anaerobic dimethylsulfoxide reductase subunit B
A08	CDS	3201708	3202190	STM14_3648	3201765	3202159	-	Putative inner membrane protein
A09	CDS	1355292	1355618	STM14_1521	1355322	1355587	-	Putative cytoplasmic protein
A10	CDS	4278811	4279794	STM14_4877	4278841	4279763	+	Putative periplasmic dicarboxylate-binding protein
A11	CDS	3309225	3309710	STM14_3785	3309255	3309679	+	Putative cytoplasmic protein
A12	CDS	753223	754362	STM14_0805	753253	754331	+	Citrate utilization protein b
B01	CDS	1282096	1282425	STM14_1414	1282126	1282394	+	Putative periplasmic protein
B02	CDS	3625984	3626295	STM14_4166	3626014	3626264	+	Hypothetical protein
B03	CDS	3214693	3215070	STM14_3664	3214723	3214889	+	Putative virulence-associated protein
B04	CDS	2518730	2520136	STM14_2904	2518760	2520105	+	Putative amino acid transporter
B05	CDS	4717965	4718453	STM14_5355	4717995	4718422	+	Putative arginine repressor
B06	CDS	3375256	3375933	STM14_3868	3375286	3375902	-	Putative disulfide oxidoreductase
B07	CDS	2344235	2345602	STM14_2709	2344265	2345571	+	Putative L-serine dehydratase
B09	CDS	1460518	1461864	STM14_1662	1460548	1461833	+	Putative Na+-dicarboxylate symporter
B12	CDS	4133205	4133354	STM14_4709	4133274	4133323	-	Putative inner membrane protein
C01	CDS	2284669	2285004	STM14_2638	2284699	2284973	+	Putative inner membrane protein
C02	CDS	2044533	2045972	STM14_2358	2044563	2045941	+	Putative cell wall-associated hydrolase
C04	CDS	2702699	2702920	STM14_3088	2702729	2702889	+	Putative cytoplasmic protein
C05	CDS	16493	17026	STM14_0018	16523	16995	+	Hypothetical protein
C06	CDS	622504	622740	STM14_0660	622534	622709	-	Putative periplasmic protein
C08	CDS	4486168	4486623	STM14_5111	4486198	4486592	+	Putative outer membrane lipoprotein
C09	CDS	4033878	4034510	STM14_4613	4033908	4034479	+	Chaperone protein TorD

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Well Position	Deleted Region of Chromosome	Deletion Start	Deletion End	Locus Tag	14028S Gene Start	14028S Gene End	14028S Gene Strand	Description
C10	CDS	3985597	3986022	STM14_4548	3985627	3985991	+	Putative phosphotransferase system enzyme IIA
C11	CDS	1699931	1700701	STM14_1936	1699961	1700670	+	Periplasmic dipeptidase precursor
C12	CDS	1140624	1141988	STM14_1241	1140654	1141957	+	Copper resistance protein
D01	CDS	404024	405982	STM14_0417	404054	405951	-	DNA methylase
D02	CDS	2894442	2896622	STM14_3299	2894472	2896591	-	Putative ABC transporter transmembrane region
D03	CDS	4573190	4573405	STM14_5186	4573220	4573320	+	Putative cytoplasmic protein
D04	CDS	2538835	2539353	STM14_2925	2538865	2539322	-	Putative periplasmic protein
D05	CDS	1652607	1653392	STM14_1884	1652637	1653361	+	Putative lipoprotein
D06	CDS	4485843	4486181	STM14_5110	4485873	4486150	+	Putative inner membrane protein
D07	CDS	4276977	4278284	STM14_4875	4277007	4278253	+	Putative C4-dicarboxylate transport system
D08	CDS	1350732	1351706	STM14_1516	1350762	1351675	-	Putative ABC transporter protein
D09	CDS	3073139	3073519	STM14_3512	3073169	3073488	+	Putative cytoplasmic protein
D10	CDS	1160792	1161415	STM14_1268	1160822	1161384	-	Suppression of copper sensitivity protein
D11	CDS	2299459	2299740	STM14_2656	2299489	2299709	+	Putative outer membrane protein
D12	CDS	1130516	1131064	STM14_1226	1130546	1131033	-	Putative outer membrane protein
E02	CDS	1578501	1579649	STM14_1802	1578531	1579618	+	Proline/glycine betaine transport system
E03	CDS	3073516	3074724	STM14_3513	3073546	3074693	+	Putative permease
E04	CDS	1620366	1620665	STM14_1853	1620396	1620634	+	Putative hydrogenase protein
E05	CDS	4782113	4783297	STM14_5422	4782143	4783266	+	Putative sugar transporter
E06	CDS	1356571	1357359	STM14_1525	1356601	1357328	-	Aminoglycoside resistance protein
E07	CDS	1364115	1364426	STM14_1543	1364145	1364395	-	Putative periplasmic protein
E08	CDS	3129129	3130271	STM14_3567	3129159	3130240	+	Putative glycerate kinase 2
E09	CDS	1802408	1802707	STM14_2052	1802438	1802676	-	Putative cytoplasmic protein
E10	CDS	1619959	1620369	STM14_1852	1619989	1620338	+	Putative hydrogenase
E12	CDS	3659109	3659648	STM14_4203	3659139	3659617	+	Putative inner membrane protein
F01	CDS	1349098	1350732	STM14_1515	1349128	1350701	-	Putative ABC transporter periplasmic binding protein
F02	CDS	4722158	4723378	STM14_5361	4722188	4723347	+	Arginine deiminase
F03	CDS	2343841	2344230	STM14_2708	2343871	2344184	+	Putative transcriptional regulator
F04	CDS	4867622	4867936	STM14_5518	4867652	4867905	+	Putative periplasmic protein
F05	CDS	2893036	2894445	STM14_3298	2893066	2894414	-	Putative outer membrane efflux protein
F06	CDS	2507886	2508158	STM14_2888	2507916	2508127	+	Putative cytoplasmic protein
F07	CDS	1650694	1652478	STM14_1883	1650724	1652447	+	Putative alpha amylase
F08	CDS	1347423	1347953	STM14_1512	1347453	1347922	+	Putative inner membrane protein
F09	CDS	3312670	3313974	STM14_3791	3312700	3313943	+	Putative permease
F10	CDS	3346180	3346635	STM14_3836	3346210	3346604	-	Putative diadenosine tetraphosphatehydrolase
F11	CDS	3372754	3374550	STM14_3866	3372784	3373689	-	Putative arylsulfate sulfotransferase
F12	CDS	3310436	3311359	STM14_3787	3310466	3311328	+	Putative xylanase/chitin deacetylase
G01	CDS	3792807	3793163	STM14_4337	3792837	3793132	-	Putative phage endolysin
G02	CDS	3985101	3985586	STM14_4547	3985131	3985555	+	Putative phosphotransferase system enzyme IIB
G03	CDS	1618880	1619941	STM14_1851	1618910	1619910	+	Putative hydrogenase
G04	CDS	1048140	1049231	STM14_1130	1048170	1049200	+	Outer membrane protein F precursor
G05	CDS	2952594	2954009	STM14_3359	2952624	2953978	+	Regulatory protein
G06	CDS	2040221	2040556	STM14_2352	2040251	2040525	-	Putative outer membrane lipoprotein
G07	CDS	3344681	3346135	STM14_3835	3344680	3346104	-	Putative cation transporter
G09	CDS	4644931	4645857	STM14_5279	4644961	4645826	-	Putative permease
G10	CDS	4569273	4569926	STM14_5181	4569303	4569895	-	Putative anaerobic dehydrogenase component
G12	CDS	402095	402289	STM14_0414	402125	402258	-	Putative copper chaperone
H01	CDS	3850516	3851055	STM14_4400	3850546	3851024	-	Hypothetical protein
H02	CDS	4772403	4772864	STM14_5408	4772433	4772833	-	Hypothetical protein
H03	CDS	2712174	2713037	STM14_3100	2712204	2713006	+	Putative polyferredoxin
H04	CDS	4772861	4773535	STM14_5409	4772891	4773504	+	Putative dienelactone hydrolase
H05	CDS	827686	828669	STM14_0888	827716	828602	+	Transcriptional regulator
H06	CDS	3302343	3303026	STM14_3775	3302373	3302995	-	Putative response regulator
H07	CDS	4818950	4819675	STM14_5463	4818980	4819644	-	Putative transcriptional regulator
H08	CDS	2812004	2812690	STM14_3198	2812034	2812659	-	Hypothetical protein
H09	CDS	2824312	2824662	STM14_3223	2824342	2824631	-	Hypothetical protein

[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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