Product Information Sheet for NR-42833

Salmonella enterica subsp. enterica, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_045/046_Kan

Catalog No. NR-42833

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-42833 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_045/046_Kan, NR-42833."

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_045/046_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
A02	CDS	3915046	3916080	STM14_4467	3915076	3916049	+	Glycosyl transferase
A03	CDS	1666787	1668274	STM14_1900	1666817	1668243	+	Methyl viologen resistance
A05	CDS	2244156	2245277	STM14_2604	2244186	2245246	+	GDP-D-mannose dehydratase
A06	CDS	1546357	1547397	STM14_1762	1546315	1547366	-	Putative oxidoreductase
A07	CDS	1623778	1624881	STM14_1857	1623808	1624850	+	Putative hydrogenase-1 small subunit
A08	CDS	3012403	3012813	STM14_3440	3012433	3012782	+	Hydrogenase 3 large subunit processing protein
A09	CDS	1204583	1205701	STM14_1329	1204613	1205670	+	N-methyltryptophan oxidase
A10	CDS	2402944	2403156	STM14_2780	2402974	2403086	+	Heme exporter protein C
A11	CDS	4546181	4547251	STM14_5163	4546211	4547220	+	Sensor protein BasS/PmrB
A12	CDS	4485067	4485483	STM14_5108	4485097	4485452	-	Putative cytoplasmic protein
B01	CDS	4239354	4240775	STM14_4830	4239384	4240744	+	GPH family transport protein
B02	CDS	1839558	1839962	STM14_2096	1839588	1839931	-	Putative ferredoxin
B03	CDS	718694	719401	STM14_0763	718724	719370	-	Putative cytoplasmic protein
B05	CDS	2364078	2364797	STM14_2735	2364108	2364766	-	Putative permease
B06	CDS	1981459	1981689	STM14_2281	1981489	1981658	-	DNA polymerase III subunit theta
B08	CDS	563180	563857	STM14_0590	563210	563826	-	Putative ABC transporter ATP-binding protein YbbL
B09	CDS	3956456	3958165	STM14_4514	3956486	3958134	-	Putative inner membrane protein
B10	CDS	2471105	2472129	STM14_2851	2471242	2472098	-	
B11	CDS	1613547	1613906	STM14_1844	1613577	1613875	-	Putative cytoplasmic protein
B12	CDS	640057	640305	STM14_0679	640087	640274	+	Putative inner membrane protein
C01	CDS	3280066	3280770	STM14_3744	3280096	3280739	-	Hypothetical protein
C02	CDS	1907534	1907788	STM14_2173	1907564	1907757	+	Putative transglycosylase-associated protein
C03	CDS	2147847	2148776	STM14_2503	2147877	2148745	+	Hypothetical protein
C04	CDS	3817917	3818120	STM14_4360	3817947	3818089	+	Putative cytoplasmic protein
C05	CDS	558639	561140	STM14_0586	558669	561109	+	Copper exporting ATPase
C06	CDS	774958	775164	STM14_0827	774988	775133	-	Putative periplasmic protein
C07	CDS	1390918	1391724	STM14_1580	1390948	1391693	+	Exonuclease III

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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
C09	CDS	837679	838719	STM14_0898	837709	838688	+	Aldose 1-epimerase
C11	CDS	4319348	4319560	STM14_4924	4319378	4319529	-	50S ribosomal protein L31
C12	CDS	4024328	4024663	STM14_4602	4024361	4024632	-	Hypothetical protein
D01	CDS	1591778	1592104	STM14_1817	1591808	1592073	-	Hypothetical protein
D02	CDS	1544982	1545422	STM14_1759	1545012	1545391	+	Putative inner membrane protein
D03	CDS	3451915	3452232	STM14_3953	3451945	3452201	-	GIY-YIG nuclease superfamily protein
D04	CDS	1838582	1839220	STM14_2095	1838612	1839189	+	Outer membrane protein W
D05	CDS	2610811	2611260	STM14_3006	2610841	2611229	+	Putative inner membrane protein
D06	CDS	2610174	2610749	STM14_3005	2610204	2610718	+	Hypothetical protein
D07	CDS	1879445	1879675	STM14_2140	1879475	1879644	+	Cation transport regulator
D08	CDS	3881664	3883202	STM14_4438	3881694	3883171	+	Aldehyde dehydrogenase B
D09	CDS	3540575	3541513	STM14_4052	3540605	3541482	+	Malate dehydrogenase
D10	CDS	1193154	1194308	STM14_1315	1193184	1194277	+	Glucans biosynthesis protein
D11	CDS	1785562	1786068	STM14_2030	1785592	1786037	-	Thiol peroxidase
D12	CDS	4489545	4489661	STM14_5113	4489575	4489630	-	Putative cytoplasmic protein
E01	CDS	4632426	4634066	STM14_5259	4632474	4634035	-	Isovaleryl CoA dehydrogenase
E02	CDS	2967460	2967861	STM14_3377	2967490	2967830	+	DNA binding protein, nucleoid-associated
E03	CDS	1860732	1861637	STM14_2120	1860762	1861606	+	Hypothetical protein
E04	CDS	3560910	3561884	STM14_4071	3560940	3561853	-	Putative oxidoreductase
E05	CDS	1809601	1810353	STM14_2060	1809631	1810322	-	Putative regulatory protein
E06	CDS	442110	442301	STM14_0460	442140	442270	-	Hypothetical protein
E07	CDS	919153	921024	STM14_0991	919183	920993	-	Glutathione transporter ATP-binding protein
E08	CDS	3314379	3315563	STM14_3795	3314409	3315532	-	Mannonate dehydratase
E09	CDS	469393	469932	STM14_0490	469423	469901	+	Hypothetical protein
E11	CDS	2608151	2608942	STM14_3003	2608181	2608911	+	Short chain dehydrogenase
E12	CDS	2036100	2037002	STM14_2346	2036130	2036872	+	Trehalose-6-phosphate phosphatase
F01	CDS	892059	892325	STM14_0957	892089	892294	+	Hypothetical protein
F02	CDS	1931961	1932305	STM14_2204	1931991	1932274	-	Putative translation initiation inhibitor
F04	CDS	3238779	3240095	STM14_3695	3238809	3240064	+	Proline aminopeptidase P II
F05	CDS	4734836	4734955	STM14_5374	4734866	4734924	+	Putative cytoplasmic protein
F06	CDS	3864222	3866249	STM14_4420	3864252	3866218	-	Periplasmic alpha-amylase precursor
F07	CDS	5966	7396	STM14_0006	5996	7365	+	Putative alanine/glycine transport protein
F08	CDS	1366163	1366957	STM14_1548	1366193	1366926	-	Putative regulatory protein
F09	CDS	4720099	4721103	STM14_5358	4720129	4721072	+	Ornithine carbamoyltransferase
F10	CDS	474292	475266	STM14_0499	474322	475235	+	Putative oxidoreductase
F11	CDS	1961884	1962075	STM14_2240	1961914	1962044	-	Putative inner membrane lipoprotein
F12	CDS	1118785	1119237	STM14_1213	1118815	1119206	-	Hypothetical protein
G02	CDS	4630147	4630545	STM14_5256	4630177	4630514	-	Putative inner membrane protein
G04	CDS	2056152	2056871	STM14_2374	2056182	2056810	+	Flagellar biosynthesis sigma factor
G06	CDS	935679	936305	STM14_1012	935709	936274	+	Putative glutathione S-transferase
G07	CDS	4579601	4580176	STM14_5198	4579631	4580145	+	Putative transcriptional regulator
G09	CDS	3175908	3176228	STM14_3612	3175938	3176197	+	Hypothetical protein
G10	CDS	3850130	3850342	STM14_4399	3850160	3850311	-	Major cold shock protein
G12	CDS	644696	645910	STM14_0684	644726	645879	-	Enterobactin/ferric enterobactin esterase
H02	CDS	3020033	3020389	STM14_3450	3020063	3020358	-	Hydrogenase nickel incorporation protein
H03	CDS	2411612	2412106	STM14_2792	2411642	2412075	-	Ecotin precursor
H04	CDS	1191650	1193137	STM14_1314	1191683	1193106	-	Putative phospholipase
H05	CDS	3124976	3126271	STM14_3565	3125006	3126240	+	23S rRNA 5-methyluridine methyltransferase
H06	CDS	1948625	1948864	STM14_2227	1948655	1948833	-	Hypothetical protein
H07	CDS	3145479	3146126	STM14_3587	3145509	3146095	+	L-fuculose phosphate aldolase
H09	CDS	3575003	3576160	STM14_4090	3575033	3576129	-	Acriflavine resistance protein E precursor
H10	CDS	586072	587370	STM14_0614	586102	587339	-	Putative purine permease YbbY
H11	CDS	2184264	2185166	STM14_2547	2184294	2185135	-	Propanediol utilization protein
H12	CDS	445589	446761	STM14_0466	445619	446730	+	Protein AraJ

[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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