Product Information Sheet for NR-42841

Salmonella enterica subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate SGD_061/062_Kan

Catalog No. NR-42841

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-42841 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_061/062_Kan, NR-42841."

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_061/062_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	2769141	2769989	STM14_3152	2769171	2769958	-	Putative DNA-binding transcriptional regulator
A02	CDS	2729200	2729400	STM14_3112	2729230	2729369	+	Hypothetical protein
A03	CDS	2757727	2759169	STM14_3143	2757757	2759081	+	Putative sensor kinase
A04	CDS	2773243	2773503	STM14_3156	2773273	2773472	-	Putative ferredoxin
A05	CDS	2734484	2734978	STM14_3120	2734514	2734947	+	DNA-binding transcriptional regulator IscR
A06	CDS	2861585	2861923	STM14_3266	2861615	2861892	-	translation inhibitor protein RaiA
A07	CDS	2839740	2840123	STM14_3243	2839770	2840092	+	Autonomous glycyl radical cofactor GrcA
A08	CDS	2841247	2842284	STM14_3245	2841277	2842253	+	Putative methyltransferase
A09	CDS	2858736	2859467	STM14_3262	2858766	2859436	+	Hypothetical protein
A10	CDS	2847889	2848212	STM14_3251	2847919	2848181	-	Hypothetical protein
A12	CDS	2842980	2843660	STM14_3247	2843010	2843629	-	Putative cytoplasmic protein
B01	CDS	2843714	2846374	STM14_3248	2843744	2846343	-	Putative acetyl-CoA synthetase
B03	CDS	2873547	2874743	STM14_3284	2873532	2874712	-	Hypothetical protein
B04	CDS	2879495	2879785	STM14_3292	2879525	2879754	+	Hypothetical protein
B05	CDS	2879775	2880251	STM14_3293	2879805	2880199	+	Hypothetical protein
B08	CDS	2959079	2960347	STM14_3366	2959109	2960316	-	Hypothetical protein
B09	CDS	3026080	3026424	STM14_3456	3026110	3026393	+	Hypothetical protein
B10	CDS	3006742	3007875	STM14_3432	3006772	3007844	-	Nitric oxide reductase
B11	CDS	3089457	3090506	STM14_3531	3089487	3090475	+	tRNA pseudouridine synthase D
C01	CDS	3116367	3117038	STM14_3556	3116397	3117007	+	Hypothetical protein
C02	CDS	3153667	3154062	STM14_3595	3153697	3154031	+	Hypothetical protein
C03	CDS	3138652	3140016	STM14_3578	3138712	3139985	-	Putative nucleotide binding protein
C05	CDS	3158652	3159458	STM14_3602	3158682	3159427	+	Putative enzyme
C06	CDS	3181977	3182507	STM14_3621	3182007	3182476	+	Dinucleoside polyphosphate hydrolase
C07	CDS	3184067	3184780	STM14_3629	3184097	3184749	-	Putative transport protein
C10	CDS	3240866	3241195	STM14_3697	3240896	3241164	-	Z-ring-associated protein
C12	CDS	3246728	3247363	STM14_3706	3246758	3247290	+	Arginine exporter protein
D01	CDS	3247563	3248423	STM14_3707	3247593	3248392	+	Mechanosensitive channel MscS
D02	CDS	3245888	3246634	STM14_3705	3245918	3246588	+	Hypothetical protein
D03	CDS	3257799	3258557	STM14_3719	3257829	3258526	-	Putative Zn-dependent protease
D04	CDS	3286933	3287652	STM14_3754	3286963	3287621	+	tRNA (guanine-N(7))-methyltransferase

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Well Position	Gene Type	Gene Start	Gene End	Target Gene (Locus Tag)	Deleted Region Start	Deleted Region End	Gene Strand	Description
D05	CDS	3275310	3276041	STM14_3738	3275292	3276010	-	Hypothetical protein
D06	CDS	3286607	3286933	STM14_3753	3286637	3286902	+	Hypothetical protein
D07	CDS	3285838	3286557	STM14_3752	3285868	3286526	+	Hypothetical protein
D08	CDS	3280789	3281355	STM14_3745	3280819	3281324	-	Putative integral membrane protein
D09	CDS	3281352	3281642	STM14_3746	3281382	3281611	-	Hypothetical protein
D10	CDS	3281650	3282243	STM14_3747	3281680	3282212	-	Putative deoxyribonucleotide triphosphate pyrophosphatase
D11	CDS	3282236	3283372	STM14_3748	3282266	3283341	-	Coproporphyrinogen III oxidase
D12	CDS	3288887	3289162	STM14_3756	3288917	3289131	-	Hypothetical protein
E01	CDS	3340573	3341232	STM14_3828	3340603	3341201	-	Hypothetical protein
E02	CDS	3322681	3323547	STM14_3804	3322711	3323516	-	Putative glutathione S-transferase YghU
E03	CDS	3332203	3332490	STM14_3816	3332233	3332459	+	Putative cytoplasmic protein
E04	CDS	3368507	3369178	STM14_3861	3368537	3369147	-	Hypothetical protein
E05	CDS	3369184	3370347	STM14_3862	3369214	3370316	-	Putative glutathionylspermidine synthase
E06	CDS	3370409	3371239	STM14_3863	3370439	3371208	+	Hypothetical protein
E07	CDS	3371338	3372111	STM14_3864	3371368	3372080	-	Zinc transporter ZupT
E08	CDS	3390205	3390816	STM14_3883	3390235	3390785	-	Putative glycerol-3-phosphate acyltransferase PlsY
E09	CDS	3386797	3387411	STM14_3879	3386827	3387380	-	Putative signal transduction protein
E10	CDS	3361775	3362089	STM14_3853	3361805	3362058	-	Putative cytoplasmic protein
E11	CDS	3347190	3349361	STM14_3839	3347220	3349330	+	Hypothetical protein
E12	CDS	3358506	3358898	STM14_3848	3358536	3358867	+	Putative outer membrane protein
F01	CDS	3358897	3359709	STM14_3849	3359080	3359678	-	DNA-binding transcriptional regulator QseB
F02	CDS	3407829	3408326	STM14_3900	3407859	3408295	-	Putative metal-dependent hydrolase
F03	CDS	3409259	3410257	STM14_3903	3409289	3410226	-	Putative dehydrogenase
F04	CDS	3410530	3411498	STM14_3905	3410560	3411467	-	Putative tellurite resistance protein
F05	CDS	3411753	3412997	STM14_3906	3411783	3412966	-	Serine/threonine transporter SstT
F06	CDS	3417861	3418226	STM14_3916	3417891	3418195	-	Putative inner membrane protein
F07	CDS	3418265	3419161	STM14_3917	3418295	3419130	+	Putative transcriptional regulator
F08	CDS	3419266	3419967	STM14_3918	3419296	3419936	-	Putative cytoplasmic protein
F11	CDS	3499762	3500049	STM14_4009	3499792	3500018	-	Putative sigma(54) modulation protein
G01	CDS	3503508	3504161	STM14_4015	3503538	3504130	+	Isoprenoid biosynthesis protein
G03	CDS	3450936	3451454	STM14_3951	3450966	3451423	-	Putative intracellular proteinase
G04	CDS	3451434	3451877	STM14_3952	3451464	3451846	+	Hypothetical protein
G05	CDS	3452219	3452722	STM14_3954	3452249	3452691	+	Putative transport protein
G06	CDS	3452716	3453240	STM14_3955	3452746	3453161	+	Putative lipid carrier protein
G07	CDS	3453457	3454452	STM14_3956	3453487	3454421	-	Putative protease
G08	CDS	3454461	3455339	STM14_3957	3454473	3455308	-	Putative protease
G09	CDS	3455517	3456524	STM14_3958	3455547	3456493	-	Hypothetical protein
G12	CDS	3528752	3529156	STM14_4039	3528782	3529125	-	Cytochrome d ubiquinol oxidase subunit III
H01	CDS	3506818	3507747	STM14_4017	3506848	3507716	+	Putative FeS oxidoreductase
H02	CDS	3527441	3528565	STM14_4038	3527471	3528534	+	Putative ATPase
H03	CDS	3543899	3545866	STM14_4058	3543929	3545835	+	p-hydroxybenzoic acid efflux subunit AaeB
H04	CDS	3545872	3546804	STM14_4059	3545902	3546773	+	p-hydroxybenzoic acid efflux subunit AaeA
H06	CDS	3547197	3548126	STM14_4061	3547227	3548095	-	Putative DNA-binding transcriptional regulator
H07	CDS	3558766	3560706	STM14_4070	3558796	3560675	+	Regulatory protein CsrD
H08	CDS	3555205	3555798	STM14_4065	3555235	3555767	+	Maf-like protein
H09	CDS	3569170	3570135	STM14_4082	3569200	3570104	-	tRNA-dihydrouridine synthase B
H10	CDS	3565933	3566175	STM14_4078	3565963	3566144	-	Hypothetical protein
H11	CDS	3579521	3579742	STM14_4092	3579551	3579711	-	Putative outer membrane lipoprotein
H12	CDS	3617730	3618452	STM14_4156	3617760	3618421	+	Putative regulatory 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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