Product Information Sheet for NR-29405

Salmonella enterica subsp. enterica, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate 013/014_Kan

Catalog No. NR-29405

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.

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-29405 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 013/014_Kan, NR-29405."

Biosafety Level: 1
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. 6th ed. Washington, DC: U.S. Government Printing Office, 2020; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

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References:

1. Andrews-Polymenis, H. and M. McClelland, 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.

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Table 1: *S. enterica* subsp. *enterica*, Strain 14028s (Serovar Typhimurium) Single-Gene Deletion Mutant Library, Plate 013/014_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	chr_14028S	2219201	2220142	STM14_2581	2219171	2220172	+	Abequosyltransferase
A02	chr_14028S	2227252	2228070	STM14_2589	2227222	2228100	+	dTDP-glucose pyrophosphorylase
A03	chr_14028S	2332526	2332711	STM14_2692	2332496	2332741	-	Putative inner membrane protein
A04	chr_14028S	2462901	2463098	STM14_2842	2462871	2463128	+	Polymyxin resistance protein B
A05	chr_14028S	2522847	2524184	STM14_2907	2522817	2524214	+	Putative diaminopimelate decarboxylase
A06	chr_14028S[3]	2657738	2658031	STM14_3055	2657708	2658061	+	Putative glycerate kinase
A07	chr_14028S	2741893	2742471	STM14_3130	2741863	2742501	+	Hypothetical protein
A08	chr_14028S	2813630	2814133	STM14_3201	2813600	2814163	+	Antirepressor-like protein
A09	chr_14028S	2925730	2926266					
A10	chr_14028S	2984819	2986129	STM14_3401	2984789	2986159	+	Putative glycoporin
A11	chr_14028S	3294663	3295037	STM14_3766	3294633	3295067	+	Putative lactoylglutathione lyase
A12	chr_14028S	3457410	3457880	STM14_3960	3457380	3457910	-	Putative cytoplasmic protein
B02	chr_14028S	2228178	2229017	STM14_2590	2228148	2229047	+	dTDP-4-dehydrorhamnose reductase
B04	chr_14028S	2491475	2491567	STM14_2872	2491445	2491597	+	Putative cytoplasmic protein
B05	chr_14028S	2537533	2537847	STM14_2922	2537503	2537877	+	Putative cytoplasmic protein
B06	chr_14028S	2670167	2672320	STM14_3068	2670137	2672350	+	Putative diguanylate cyclase
B07	chr_14028S	2756991	2757695	STM14_3142	2756961	2757725	+	Hypothetical protein
B08	chr_14028S	2814466	2815083	STM14_3203	2814436	2815113	+	Antiterminator-like protein
B09	chr_14028S	2926707	2927124	STM14_3331	2926750	2927154	+	Putative transposase
B10	chr_14028S	3113590	3114327	STM14_3555	3113560	3114357	+	Putative metal-dependent hydrolase
B11	chr_14028S	3303160	3303657	STM14_3776	3303130	3303687	+	Putative cytoplasmic protein
B12	chr_14028S	3531907	3532545	STM14_4042	3531877	3532608	+	Putative inner membrane protein
C03	chr_14028S	2361219	2361281	STM14_2729	2361189	2361311	+	Putative inner membrane protein
C04	chr_14028S	2491707	2492585	STM14_2873	2491677	2492615	+	NADH dehydrogenase transcriptional repressor
C05	chr_14028S	2538080	2538268	STM14_2923	2538050	2538298	-	Putative regulatory protein
C06	chr_14028S[4]	2672685	2672864	STM14_3070	2672655	2672915	+	Putative inner membrane protein
C07	chr_14028S	2766790	2768091	STM14_3150	2766760	2768121	+	Putative phosphotransferase system IIB component
C08	chr_14028S	2839130	2839648	STM14_3242	2839091	2839678	-	Neutral amino-acid efflux protein
C09	chr_14028S	2928050	2928628	STM14_3333	2928020	2928658	+	Putative cytoplasmic protein
C10	chr_14028S	3157743	3158570	STM14_3601	3157686	3158600	-	Putative integral membrane protein
C11	chr_14028S	3308875	3309195	STM14_3784	3308845	3309225	+	Putative cytoplasmic protein
C12	chr_14028S	3536793	3537632	STM14_4047	3536763	3537662	+	Tartrate dehydratase subunit alpha
D02	chr_14028S	2245331	2245825	STM14_2605	2245301	2245855	+	Putative colanic acid biosynthesis acetyltransferase WcaF

Well Position	Deleted Region of Chromosome	Deletion Start	Deletion End	Locus Tag	14028S Gene Start	14028S Gene End	14028S Gene Strand	Description
D03	chr_14028S	2383075	2383305	STM14_2757	2383045	2383335	-	Virulence protein
D04	chr_14028S	2505677	2506447	STM14_2886	2505647	2506477	+	Putative transketolase
D06	chr_14028S	2673540	2673794	STM14_3073	2673495	2673824	-	Putative cytoplasmic protein
D07	chr_14028S	2768162	2768995	STM14_3151	2768132	2769025	+	N-acetylmuramic acid-6-phosphate etherase
D08	chr_14028S	2849644	2850039	STM14_3253	2849614	2850069	+	Putative cytoplasmic protein
D09	chr_14028S	2928685	2930607	STM14_3334	2928655	2930637	+	Putative DNA/RNA helicase
D10	chr_14028S	3215127	3215318					
D11	chr_14028S	3324355	3325347	STM14_3806	3324325	3325377	+	Putative periplasmic ferrichrome-binding protein
D12	chr_14028S	3571540	3571644	STM14_4085	3571510	3571674	-	Putative periplasmic protein
E02	chr_14028S	2246660	2247814	STM14_2607	2246630	2247844	+	Putative colanic acid biosynthesis protein
E03	chr_14028S	2386947	2388128	STM14_2761	2386917	2388158	+	Hypothetical protein
E04	chr_14028S	2506504	2507835	STM14_2887	2506474	2507871	+	Ascorbate-specific PTS system enzyme IIC
E05	chr_14028S	2578140	2580269	STM14_2964	2578110	2580299	+	Hypothetical protein
E06	chr_14028S	2685985	2693231	STM14_3082	2685955	2693261	+	
E07	chr_14028S	2770997	2772133	STM14_3154	2770967	2772163	+	Putative permease
E08	chr_14028S	2867443	2868603	STM14_3275	2867413	2868633	-	Hypothetical protein
E09	chr_14028S	2946560	2946811	STM14_3349	2946329	2946862	-	
E10	chr_14028S	3229957	3230790	STM14_3685	3229927	3230820	+	Putative outer membrane protein
E11	chr_14028S	3335329	3335784	STM14_3820	3335299	3335814	+	Putative ATP-dependent RNA helicase-like protein
E12	chr_14028S	3573957	3574559	STM14_4087	3573927	3574589	+	DNA-binding transcriptional regulator EnvR
F02	chr_14028S	2284161	2284517	STM14_2637	2284131	2284547	+	Putative cytoplasmic protein
F03	chr_14028S	2396597	2396740	STM14_2772	2396453	2396770	-	Virulence protein
F04	chr_14028S	2508269	2508652	STM14_2889	2508239	2508682	+	Putative phosphotransferase system enzyme II A component
F06	chr_14028S	2713067	2713816	STM14_3101	2713037	2713846	+	Putative dimethylsulfoxide reductase
F07	chr_14028S	2782918	2783400	STM14_3166	2782885	2783430	-	Transposase-like protein
F09	chr_14028S	2948638	2949507	STM14_3352	2948608	2949537	-	Virulence protein
F10	chr_14028S	3260983	3261396	STM14_3723	3260953	3261426	+	Putative mannitol dehydrogenase
F12	chr_14028S	3649857	3649967	STM14_4191	3649827	3649997	-	Putative outer membrane lipoprotein
G02	chr_14028S	2319585	2320286	STM14_2678	2319555	2320316	+	Acetoin dehydrogenase
G03	chr_14028S	2447623	2448366	STM14_2826	2447593	2448396	+	Putative aldolase
G04	chr_14028S	2520230	2521273	STM14_2905	2520200	2521303	+	Putative cytoplasmic protein
G05	chr_14028S	2601392	2602051	STM14_2995	2601362	2602081	-	Glutamine amidotransferase
G06	chr_14028S	2725577	2726185	STM14_3108	2725547	2726215	-	Putative cytoplasmic protein
G07	chr_14028S	2786285	2788597	STM14_3170	2786255	2788627	+	Tail fiber-like protein
G08	chr_14028S	2906328	2907551	STM14_3310	2906298	2907581	-	Putative ATPase
G09	chr_14028S	2949860	2950696	STM14_3354	2949830	2950726	-	Putative transcriptional activator
G10	chr_14028S	3263535	3264947	STM14_3726	3263505	3264977	+	Putative mannitol dehydrogenase
G11	chr_14028S	3374600	3375211	STM14_3867	3374570	3375241	-	Putative disulfide bond formation protein
G12	chr_14028S	3693621	3693836	STM14_4235	3693591	3693866	+	Putative cytoplasmic protein
H03	chr_14028S	2451016	2451738	STM14_2829	2450986	2451768	+	Putative transcriptional regulator
H04	chr_14028S	2521335	2522696	STM14_2906	2521305	2522726	+	Putative amino acid transporter
H05	chr_14028S	2613707	2613976	STM14_3010	2613677	2614006	-	Putative inner membrane protein
H06	chr_14028S	2736966	2737784	STM14_3125	2736936	2737814	-	Putative hydrolase
H07	chr_14028S[5]	1317350	1320385	STM14_1470	1317320	1320415	-	Phage tail component H-like protein
H08	chr_14028S	2912664	2913806	STM14_3319	2912634	2913836	-	Putative hexulose 6 phosphate synthase
H09	chr_14028S	2955844	2956218	STM14_3362	2955814	2956248	-	Tricarboxylic transport
H10	chr_14028S	3266249	3266938	STM14_3727	3266219	3266968	+	Putative regulatory protein
H11	chr_14028S	3456532	3457191	STM14_3959	3456502	3457221	+	Putative inner membrane protein
H12	chr_14028S	3704014	3704499	STM14_4247	3703984	3704529	+	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.

[3] Deleted region also overlaps STM14_3056 (48.2%)

[4] Deleted region also overlaps STM14_3071 (53.1%)

[5] Alternative deleted regions: 2794745 - 2797780