

Francisella tularensis subsp. novicida "Two-Allele" Transposon Mutant Library, Plate 33

Catalog No. NR-51315

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.

A comprehensive 16,508-member transposon mutant library¹ of sequence-defined transposon insertion mutants of *Francisella tularensis* subsp. *novicida*, strain U112 was prepared to allow the systematic identification of virulence determinants and other factors associated with *Francisella* pathogenesis. Genes refractory to insertional inactivation helped define the genes essential for viability of the organism.

To facilitate genome-scale screening using the mutant collection, a "two-allele" single-colony purified sublibrary, made up of approximately two purified mutants per gene, was assembled.

NR-51315 contains 18 mutants that were not available at the time of the release of Plates 1 to 32 of the "two-allele" 3,050-member sublibrary. Detailed information for these 18 mutants is shown in Tables 1 to 3.

Francisella tularensis subsp. *novicida*, strain U112 is excluded from Select Agent status. Please see [CDC Select Agent Program, Notification of Exclusion](#).

Material Provided:

Each inoculated well of the 96-well plate contains approximately 50 µL of culture in Tryptic Soy broth containing 0.1% L-cysteine and 10 µg/mL kanamycin supplemented with 5% glycerol.

Packaging/Storage:

NR-51315 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:

Wells A04, A05 and A06 should be grown in Tryptic Soy broth or agar containing 0.1% L-cysteine and 30 µg/mL erythromycin

Tryptic Soy broth or agar containing 0.1% L-cysteine and 10 µg/mL kanamycin

Incubation:

Temperature: 37°C

Atmosphere: Aerobic with 5% CO₂

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: *Francisella tularensis* subsp. *novicida* "Two-Allele" Transposon Mutant Library, Plate 33, NR-51315."

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.

Disclaimers:

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Product Information Sheet for NR-51315

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References:

- Gallagher, L. A., et al. "A Comprehensive Transposon Mutant Library of *Francisella novicida*, A Bioweapon Surrogate." *Proc. Natl. Acad. Sci. USA* 104 (2007): 1009-1014. PubMed: 17215359.

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Table 1: Plate 33 – Transposon Type and Mutated Gene¹

Well Position	Strain Name	Transposon Type	Gene	Description	Function Class
A01	tnfn1_pw060328p02q123	T20	-	conserved protein of unknown function	unknown function - conserved
A02	tnfn1_pw060328p03q104	T20	cydC	ABC-type transport ATP-binding protein CydC	transport
A03	tnfn1_pw060328p08q123	T20	aceF	pyruvate dehydrogenase complex, E2 component, dihydrolipoamide acyltransferase	carbohydrate metabolism - degradation, utilization, assimilation
A04	tnfn1_pw060418p01q10	T15	atpH	ATP synthase, F1 sector, subunit delta	energy metabolism
A05	tnfn1_pw060418p01q102	T15	-	beta-lactamase class A	other metabolism - degradation, utilization, assimilation
A06	tnfn1_pw060418p01q103	T15	-	conserved protein of unknown function	unknown function - conserved
A07	tnfn1_pw060418p01q105	<KAN-2>	-	transcriptional regulator, ArsR family	signal transduction and regulation
A08	tnfn1_pw060418p01q106	<KAN-2>	-	protein of unknown function	unknown function - novel
A09	tnfn1_pw060418p01q124	T20	iscS	cysteine desulfurase	amino acid metabolism - biosynthesis
A10	tnfn1_pw060418p01q151	T20	panD	aspartate 1-decarboxylase	cofactors, prosthetic groups, electron carriers metabolism
A11	tnfn1_pw060418p01q152	T20	fadA	acetyl-CoA acetyltransferase	other metabolism - degradation, utilization, assimilation
A12	tnfn1_pw060418p01q158	T20	-	sugar transporter, MFS superfamily	transport - carbohydrates (sugars, polysaccharides)
B01	tnfn1_pw060418p01q179	T18	-	protein of unknown function	unknown function - novel
B02	tnfn1_pw060418p02q148	T20	-	glycosyl transferase, family 8	cell wall / LPS / capsule
B03	tnfn1_pw060420p01q127	T20	-	serine-type D-Ala-D-Ala carboxypeptidase	cell wall / LPS / capsule
B04	tnfn1_pw060420p04q175	T20	-	transcriptional regulator	signal transduction and regulation
B05	tnfn1_pw060510p02q103	T20	prlC	oligopeptidase A	amino acid metabolism
B06	tnfn1_pw060510p03q138	T18	-	transcriptional regulator, LysR family	signal transduction and regulation

¹All information in this table was provided by the depositor at the time of deposition.

Table 2: Plate 33 – Sequencing and Insert Location¹

Well Position	Strain Name	Sequencing Confirmation ²	Effective Genome Position of Insertion ³	Locus Tag	ORF Left End	ORF Right End	Direction of ORF ⁴	Length of ORF (codons)	Effective Position of Insertion in ORF ⁵
A01	tnfn1_pw060328p02q123	C	741261	FTN_0697	741230	741517	F	96	32(288)
A02	tnfn1_pw060328p03q104	C	678748	FTN_0641	677863	679509	F	549	762(1647)
A03	tnfn1_pw060328p08q123	C	1586468	FTN_1493	1586220	1588112	F	631	1645(1893)
A04	tnfn1_pw060418p01q10	U	1764488	FTN_1649	1764017	1764538	F	174	51(522)
A05	tnfn1_pw060418p01q102	U	1134802	FTN_1072	1134341	1135201	R	287	462(861)
A06	tnfn1_pw060418p01q103	U	248954	FTN_0225	248455	249375	F	307	422(921)
A07	tnfn1_pw060418p01q105	C	858810	FTN_0801	858600	858947	R	116	138(348)
A08	tnfn1_pw060418p01q106	C	770015	FTN_0715	766940	770707	F	1256	3076(3768)
A09	tnfn1_pw060418p01q124	C	1311156	FTN_1245	1310691	1311863	R	391	708(1173)
A10	tnfn1_pw060418p01q151	C	1432485	FTN_1354	1432438	1432770	R	111	48(333)
A11	tnfn1_pw060418p01q152	C	1528045	FTN_1439	1527712	1528896	F	395	334(1185)
A12	tnfn1_pw060418p01q158	C	1070823	FTN_1011	1070332	1071615	F	428	793(1284)
B01	tnfn1_pw060418p01q179	C	1384035	FTN_1311	1384032	1384406	F	125	4(375)
B02	tnfn1_pw060418p02q148	C	1324223	FTN_1255	1323964	1324851	R	296	260(888)
B03	tnfn1_pw060420p01q127	U	670284	FTN_0635	669592	670983	F	464	700(1392)
B04	tnfn1_pw060420p04q175	C	905906	FTN_0850	905800	906207	R	136	107(408)
B05	tnfn1_pw060510p02q103	C	424121	FTN_0425	423396	425399	R	668	1279(2004)
B06	tnfn1_pw060510p03q138	C	391600	FTN_0392	390793	391710	R	306	111(918)

¹All information in this table was provided by the depositor at the time of deposition.

²C: Confirmed; U: Unconfirmed

³The Effective Genome Position of Insertion indicates the effective insertion position that is most likely to be functionally relevant in light of the 9-bp Tn5 target-site duplication and the direction of transcription of the predicted gene of insertion. E.g., for genes oriented to the right, the value reported indicates the genomic position of the nucleotide immediately adjacent to the left end of the insertion element when accounting for the 9-bp target site duplication.

⁴F, forward relative to genome; R, reverse

⁵Nucleotide of Insertion(Length of ORF in Nucleotides)

Table 3: Plate 33 – Sequence Mapping Quality Metrics¹

Well Position	Strain Name	Junction Info	Position in Sequence Read of Last Vector (Nucleotide)	Length of Match to Transposon	Transposon Match Score	Average Phred Score for Transposon Match	Genome Position Info	Length of Match to Genome	Genome Match Score	Average Phred Score for Genome Match
A01	tnfn1_pw060328p02q123	EXACT(0)	156	146	130	38	EXACT(0)	200	180	49
A02	tnfn1_pw060328p03q104	EXACT(0)	155	145	129	51	EXACT(0)	200	188	67
A03	tnfn1_pw060328p08q123	EXACT(0)	155	152	130	43	EXACT(0)	200	153	43
A04	tnfn1_pw060418p01q10	EXACT(0)	196	107	88	33	EXACT(0)	200	188	59
A05	tnfn1_pw060418p01q102	EXACT(0)	179	108	95	18	EXACT(0)	174	151	22
A06	tnfn1_pw060418p01q103	EXACT(0)	107	107	101	45	EXACT(0)	134	131	55
A07	tnfn1_pw060418p01q105	EXACT(0)	122	109	65	22	EXACT(0)	200	187	48
A08	tnfn1_pw060418p01q106	EXACT(0)	121	106	96	31	EXACT(0)	200	194	51
A09	tnfn1_pw060418p01q124	EXACT(0)	154	145	116	32	EXACT(0)	200	191	40
A10	tnfn1_pw060418p01q151	EXACT(0)	155	152	130	42	EXACT(0)	200	144	37
A11	tnfn1_pw060418p01q152	EXACT(0)	155	152	128	36	EXACT(0)	200	192	54
A12	tnfn1_pw060418p01q158	EXACT(0)	157	145	129	32	EXACT(0)	200	180	65
B01	tnfn1_pw060418p01q179	EXACT(0)	121	81	66	23	EXACT(0)	200	170	37
B02	tnfn1_pw060418p02q148	EXACT(0)	156	153	137	40	EXACT(0)	200	186	57
B03	tnfn1_pw060420p01q127	EXACT(0)	152	142	109	31	EXACT(0)	190	177	41
B04	tnfn1_pw060420p04q175	EXACT(0)	156	152	127	35	EXACT(0)	200	183	56
B05	tnfn1_pw060510p02q103	EXACT(0)	155	145	129	46	EXACT(0)	200	197	66
B06	tnfn1_pw060510p03q138	EXACT(0)	119	119	98	39	EXACT(0)	200	189	56

¹All information in this table was provided by the depositor at the time of deposition.