

H11-CAG-LSL-Ilf3-EGFP-polyA

Cas9-KI Strategy

Designer: Daohua Xu

Project Overview

Project Name

Ilf3

Project type

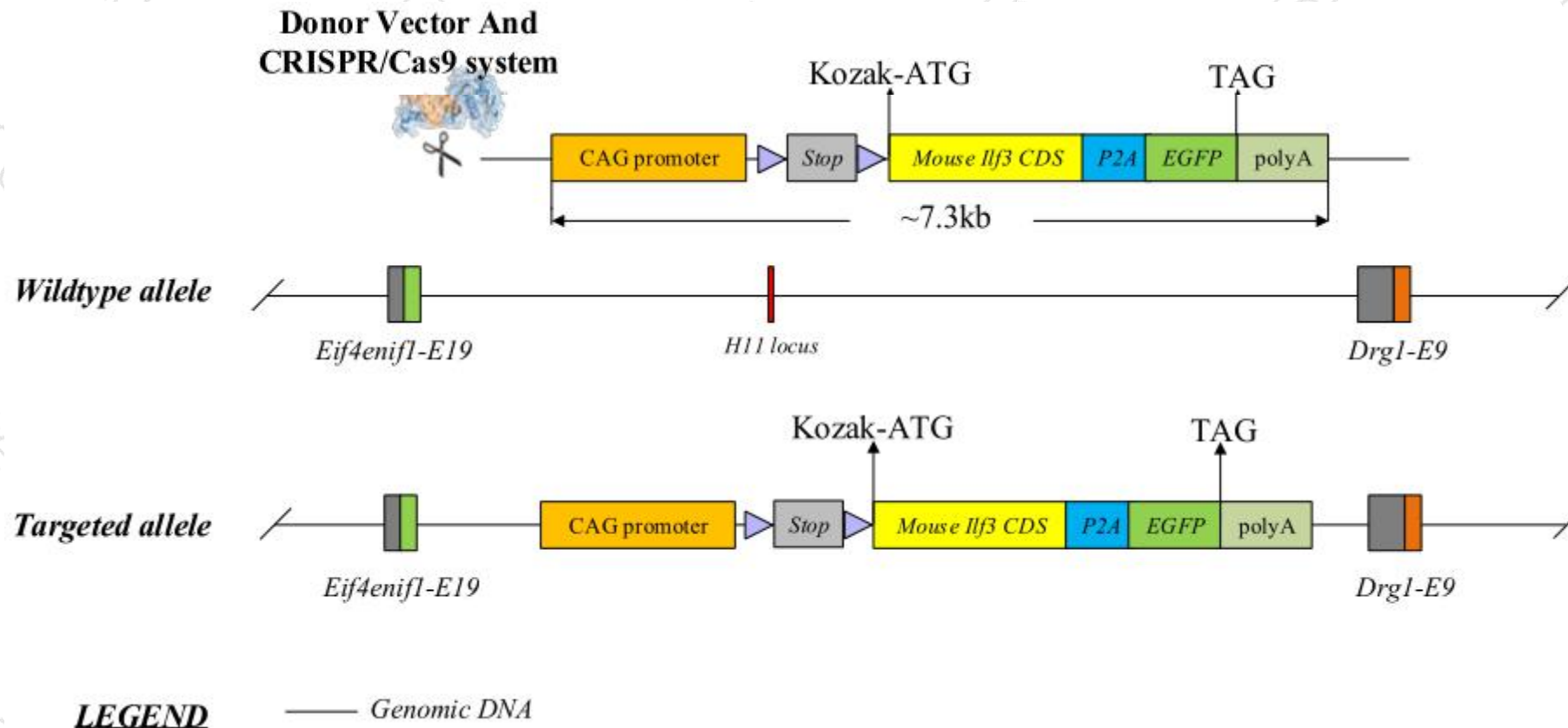
Cas9-KI (H11)

Strain background

C57BL/6J

Overview of the Targeting Strategy

This model will use CRISPR/Cas9 technology to edit the *Ilf3* gene. The schematic diagram is as follows:



- Transcript Ilf3(NM_010561.3) is selected for presentation of the recommended strategy.
- Ilf3(NM_010561.3) gene has 20 exons, with the ATG start codon in exon 2 and TAA stop codon in exon20.
- We make H11-CAG-LSL-Ilf3-P2A-EGFP-polyA knockin mice via CRISPR/Cas9 system. Cas9 mRNA, sgRNA will be co-injected into zygotes. sgRNA direct Cas9 endonuclease cleavage at H11 locus and create a DSB(double-strand break). Such breaks will be repaired, and result in CAG-LSL-Ilf3-P2A-EGFP-polyA inserted in H11 locus.
- The pups will be genotyped by PCR, followed by southern blotting and sequence analysis.

- This strategy selects the mouse Ilf3 (NM_010561.3) transcript for production, please confirm with the customer.
- After mating with Cre tool mice, the Ilf3 and EGFP genes can be expressed in specific tissues or cells.
- Since the CAG-LSL-Ilf3-P2A-EGFP-polyA fragment is large (about 7.3 kb), the offspring mice need to be subjected to southern blotting.
- The two genes linked by P2A are identical in transcription and translation, but are split into two separate proteins that function as P2A-translated polypeptides.
- Homozygous mice that knocked out the Ilf3 gene all died within 12 hours of birth, and the phenotype was unknown after overexpression.
- The H11 locus is located on the mouse chromosome 11#. If this gene is knocked into the mouse and matched with other mouse strains, try to avoid the two target genes on the same chromosome. Otherwise, the double gene will not be positive and knocked into homozygous. Mouse offspring.

Gene information (NCBI)

Ilf3 interleukin enhancer binding factor 3 [*Mus musculus* (house mouse)]

Gene ID: 16201, updated on 18-Apr-2019

Summary

Official Symbol Ilf3 provided by [MGI](#)

Official Full Name interleukin enhancer binding factor 3 provided by [MGI](#)

Primary source [MGI:MGI:1339973](#)

See related [Ensembl:ENSMUSG000000032178](#)

Gene type protein coding

RefSeq status REVIEWED

Organism [Mus musculus](#)

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as NF90; NFAR; MBII-26; MPHOSPH4

Summary The protein encoded by this gene contains two double-stranded RNA binding domains and functions in the post-transcriptional regulation of gene expression. It is a component of an RNA-protein complex that may be involved in mediating the export of messenger RNAs. Alternative splicing results in multiple transcript variants encoding distinct isoforms. These isoforms are grouped into two categories, NFAR-1 or NFAR-2, based on variation at the C-terminus. [provided by RefSeq, Mar 2013]

Expression Ubiquitous expression in CNS E11.5 (RPKM 26.3), CNS E14 (RPKM 23.6) and 28 other tissues [See more](#)

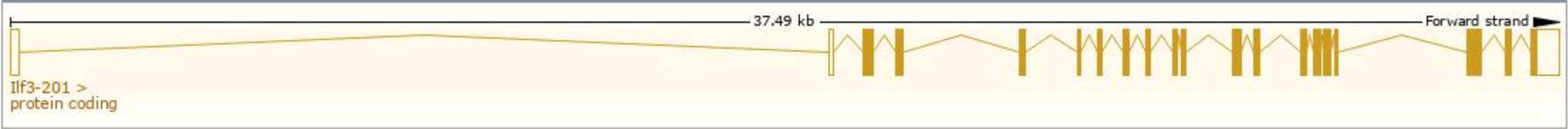
Orthologs [human](#) [all](#)

Transcript information (Ensembl)

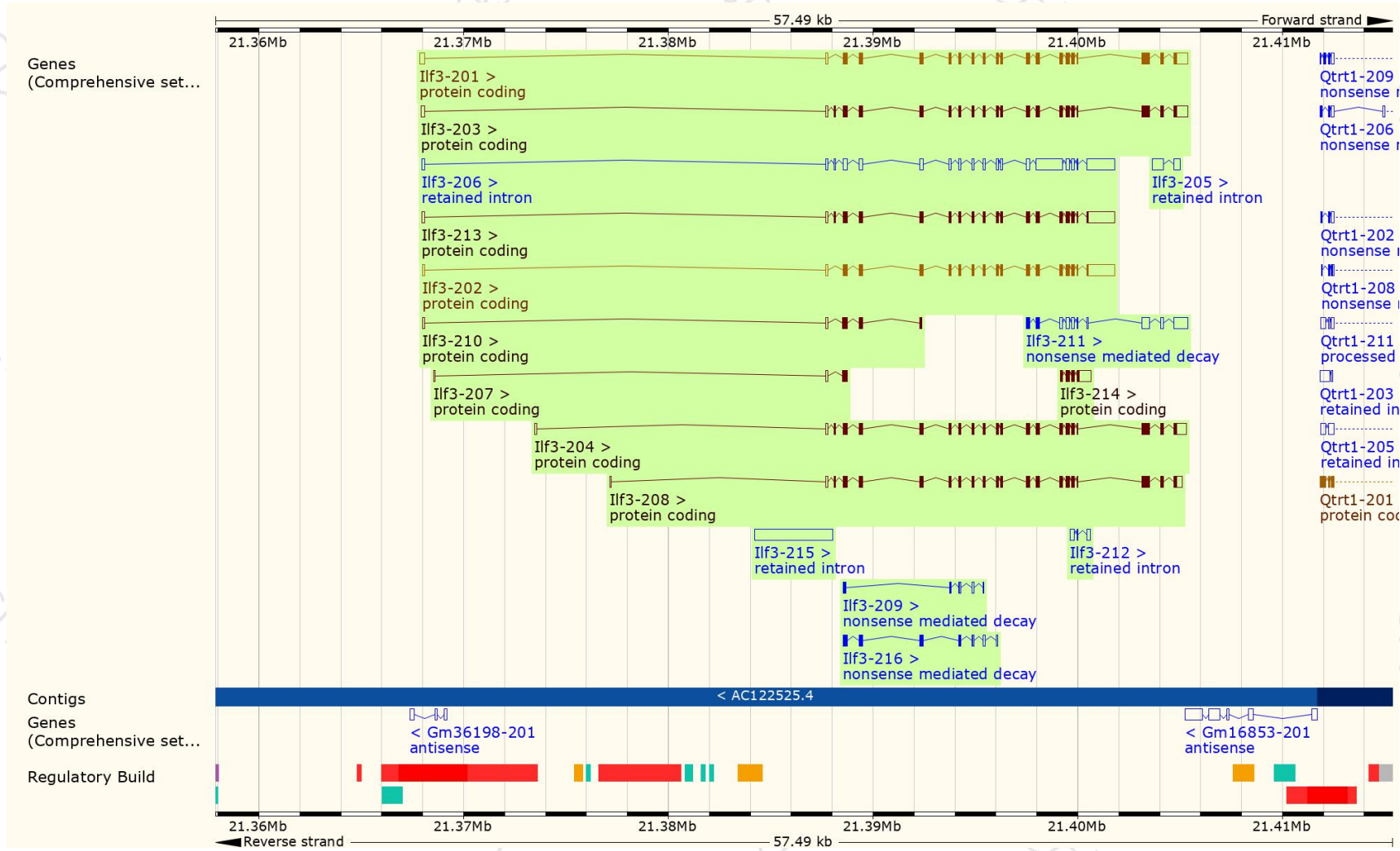
The gene has 16 transcripts,all transcripts are shown below:

Show/hide columns (1 hidden) Filter							
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ilf3-202	ENSMUST00000115414.2	3606	703aa	Protein coding	CCDS40553	Q45VK5	TSL:1 GENCODE basic APPRIS ALT2
Ilf3-201	ENSMUST00000067646.11	3549	898aa	Protein coding	CCDS40552	Q9Z1X4	TSL:1 GENCODE basic APPRIS P4
Ilf3-213	ENSMUST00000216892.1	3684	716aa	Protein coding	-	Q45VK6 Q9Z1X4	TSL:1 GENCODE basic
Ilf3-203	ENSMUST00000213518.1	3513	908aa	Protein coding	-	A0A1L1STE4	TSL:1 GENCODE basic
Ilf3-204	ENSMUST00000213603.1	3430	911aa	Protein coding	-	Q9Z1X4	TSL:5 GENCODE basic
Ilf3-208	ENSMUST00000214758.1	3177	911aa	Protein coding	-	Q9Z1X4	TSL:5 GENCODE basic
Ilf3-214	ENSMUST00000217348.1	1125	177aa	Protein coding	-	A0A1L1SQR7	CDS 5' incomplete TSL:2
Ilf3-210	ENSMUST00000214852.1	698	168aa	Protein coding	-	A0A1L1SSU8	CDS 3' incomplete TSL:3
Ilf3-207	ENSMUST00000214474.1	397	77aa	Protein coding	-	A0A1L1SQ69	CDS 3' incomplete TSL:3
Ilf3-211	ENSMUST00000215169.1	2118	126aa	Nonsense mediated decay	-	A0A1L1SU19	CDS 5' incomplete TSL:5
Ilf3-216	ENSMUST00000217498.1	900	214aa	Nonsense mediated decay	-	A0A1L1ST48	CDS 5' incomplete TSL:5
Ilf3-209	ENSMUST00000214821.1	443	74aa	Nonsense mediated decay	-	A0A1L1SR62	CDS 5' incomplete TSL:5
Ilf3-206	ENSMUST00000214217.1	4679	No protein	Retained intron	-	-	TSL:2
Ilf3-215	ENSMUST00000217476.1	3813	No protein	Retained intron	-	-	TSL:NA
Ilf3-205	ENSMUST00000214200.1	871	No protein	Retained intron	-	-	TSL:2
Ilf3-212	ENSMUST00000215438.1	415	No protein	Retained intron	-	-	TSL:2

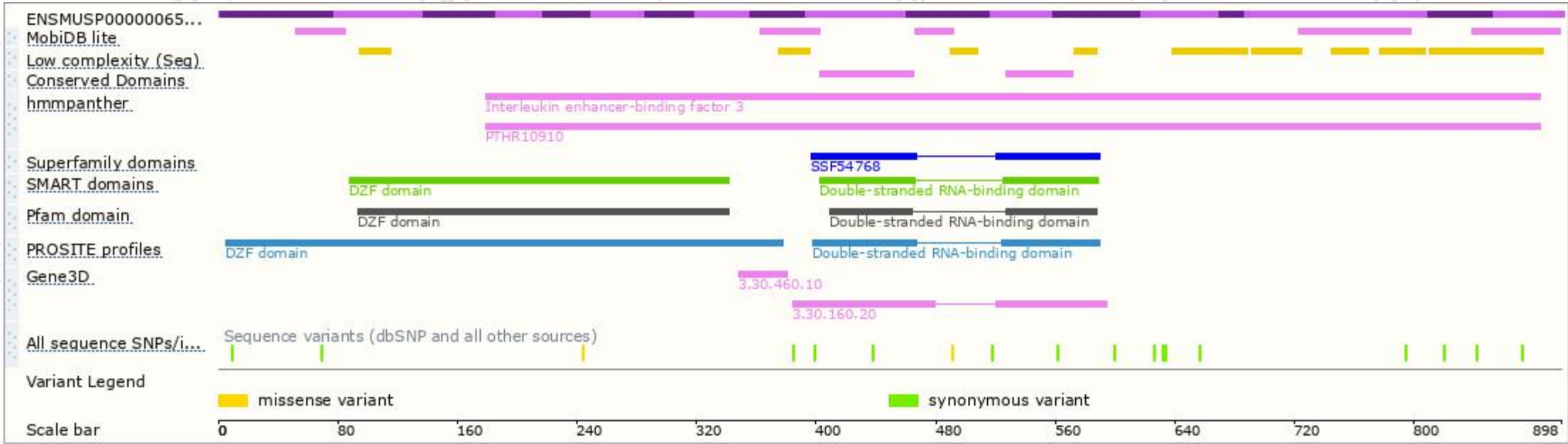
The strategy is based on the design of *Ilf3-201* transcript,The transcription is shown below



Genomic location distribution



Protein domain



CDS of mouse Ilf3 (2736bp , NM_010561.3)



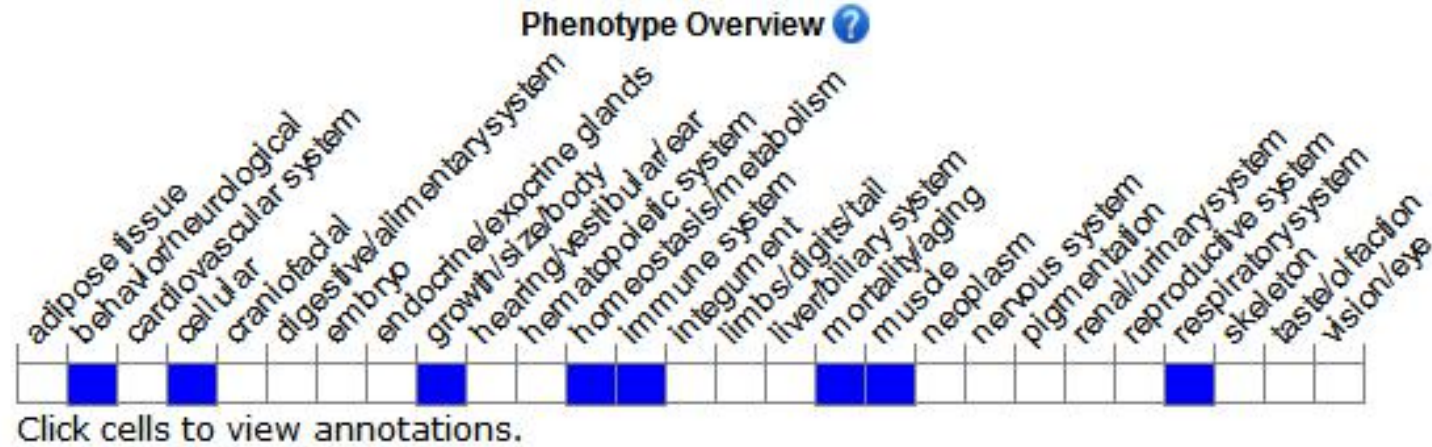
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GemPharmatech

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CCTGGGTCCAGCCAGAGCTACAGTGGTCCTGCCAGCTCCTACCAGTCCTCACAGGGTGGCTACAGTCGGAACACAGAGCACAGCATGAACTACCAGTACAGATAA

Protein of mouse Ilf3 (911aa)

MALYHHHFITRRRRRRPMRIFVNDDRHVMAKHSSVYPTQEELEAVQNMVSHTERALKAVSDWIDEQEKGNSE
AENMDTPPDDESKEGAGEQKAEHMTRTLRGVMRVGLVAKGLLLKGDLDLELVLLCKEKPTTALLDKVADNLA
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GYSRNTEHSMNYQYR

Mouse phenotype description(MGI)



Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(<http://www.informatics.jax.org/>).

According to the existing MGI data, Mice homozygous for a knock-out allele are born small and weak, show tachypnea and multi-organ apoptosis, and die neonatally due to neuromuscular respiratory failure. The diaphragm and other skeletal muscles show disorganization and paucity of myofibers, myocyte degeneration and elevated apoptosis.

If you have any questions, you are welcome to inquire.

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