

Epb41 Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Epb41

Project type

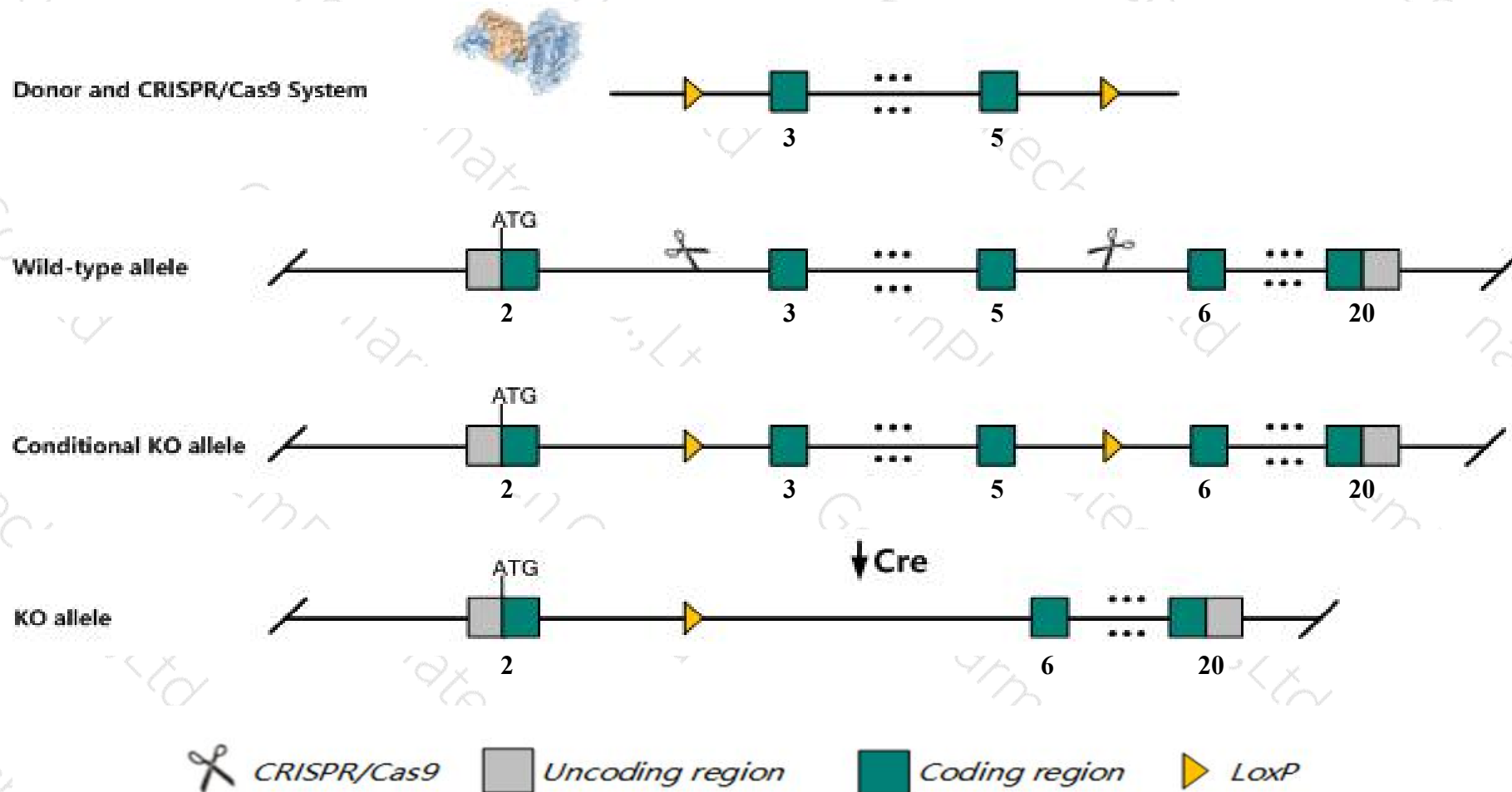
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Epb41* gene. The schematic diagram is as follows:



- The *Epb41* gene has 19 transcripts. According to the structure of *Epb41* gene, exon3-exon5 of *Epb41-201* (ENSMUST00000030739.10) transcript is recommended as the knockout region. The region contains 358bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Epb41* gene. The brief process is as follows: CRISPR/Cas9 system and Donor were microinjected into the fertilized eggs of C57BL/6JGpt mice. Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.
- The flox mice will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

- According to the existing MGI data, Homozygotes for a targeted null mutation exhibit moderate hemolytic anemia, erythrocytic abnormalities including aberrant morphology, reduced membrane stability, and lowered expression of spectrin and ankyrin.
- The *Epb41* gene is located on the Chr4. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- This Strategy is designed based on genetic information in existing databases. Due to the complexity of biological processes, all risk of loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)

Epb41 erythrocyte membrane protein band 4.1 [Mus musculus (house mouse)]

Gene ID: 269587, updated on 19-Mar-2019

Summary



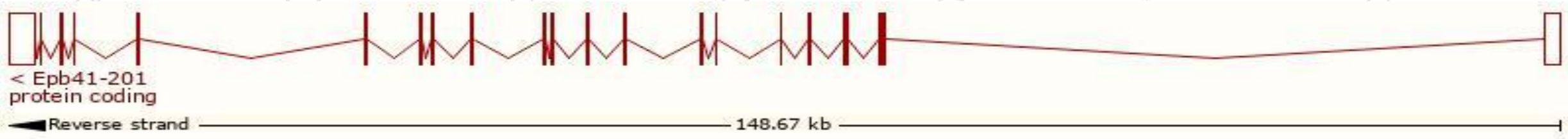
Official Symbol	Epb41 provided by MGI
Official Full Name	erythrocyte membrane protein band 4.1 provided by MGI
Primary source	MGI:MGI:95401
See related	Ensembl:ENSMUSG00000028906
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	4.1R, AI415518, D4Ertd442e, Elp-1, Elp1, Epb4.1, mKIAA4056
Expression	Ubiquitous expression in liver E14.5 (RPKM 23.5), liver E14 (RPKM 23.4) and 27 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

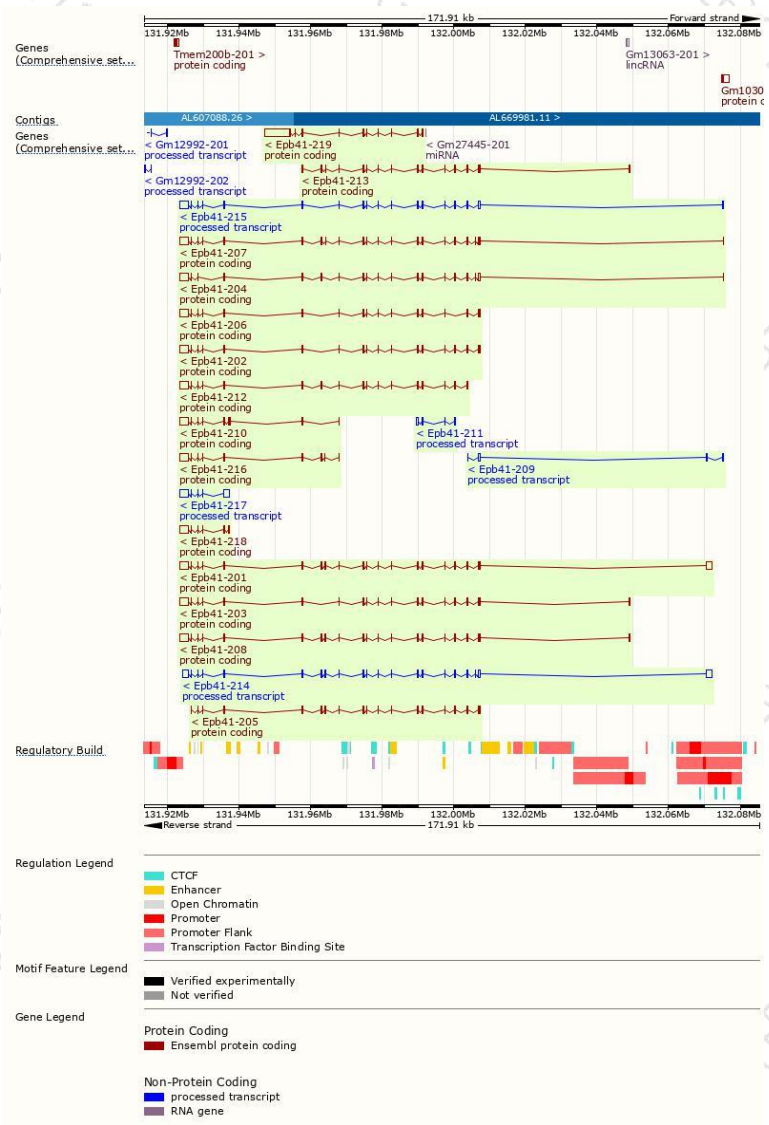
The gene has 19 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Epb41-201	ENSMUST00000030739.10	6565	858aa	Protein coding	CCDS18717	P48193	TSL:5 GENCODE basic APPRIS P3
Epb41-208	ENSMUST00000105981.8	5303	858aa	Protein coding	CCDS18717	P48193	TSL:5 GENCODE basic APPRIS P3
Epb41-203	ENSMUST00000084253.9	5141	804aa	Protein coding	CCDS51317	P48193	TSL:1 GENCODE basic APPRIS ALT2
Epb41-202	ENSMUST00000054917.11	4948	804aa	Protein coding	CCDS51317	P48193	TSL:5 GENCODE basic APPRIS ALT2
Epb41-205	ENSMUST00000105972.7	2577	858aa	Protein coding	CCDS18717	P48193	TSL:5 GENCODE basic APPRIS P3
Epb41-219	ENSMUST00000212761.1	8302	476aa	Protein coding	-	A0A1D5RLV1	CDS 5' incomplete TSL:1
Epb41-207	ENSMUST00000105975.7	5235	869aa	Protein coding	-	A2A841	TSL:5 GENCODE basic APPRIS ALT2
Epb41-204	ENSMUST00000105970.7	5059	639aa	Protein coding	-	A2A839	TSL:5 GENCODE basic APPRIS ALT2
Epb41-206	ENSMUST00000105974.7	4844	769aa	Protein coding	-	A2A842	TSL:5 GENCODE basic
Epb41-212	ENSMUST00000137846.7	4531	667aa	Protein coding	-	A2A838	CDS 5' incomplete TSL:5
Epb41-210	ENSMUST00000135579.7	3654	375aa	Protein coding	-	F6S4K9	CDS 5' incomplete TSL:1
Epb41-216	ENSMUST00000146443.7	3278	250aa	Protein coding	-	F7BUB8	CDS 5' incomplete TSL:5
Epb41-218	ENSMUST00000155990.7	3120	197aa	Protein coding	-	F7CR30	CDS 5' incomplete TSL:3
Epb41-213	ENSMUST00000141291.1	2375	715aa	Protein coding	-	A2AD32	CDS 3' incomplete TSL:5
Epb41-214	ENSMUST00000144754.7	5946	No protein	Processed transcript	-	-	TSL:1
Epb41-215	ENSMUST00000146021.7	5074	No protein	Processed transcript	-	-	TSL:1
Epb41-217	ENSMUST00000151746.7	4458	No protein	Processed transcript	-	-	TSL:1
Epb41-209	ENSMUST00000131953.1	644	No protein	Processed transcript	-	-	TSL:5
Epb41-211	ENSMUST00000136761.1	607	No protein	Processed transcript	-	-	TSL:2

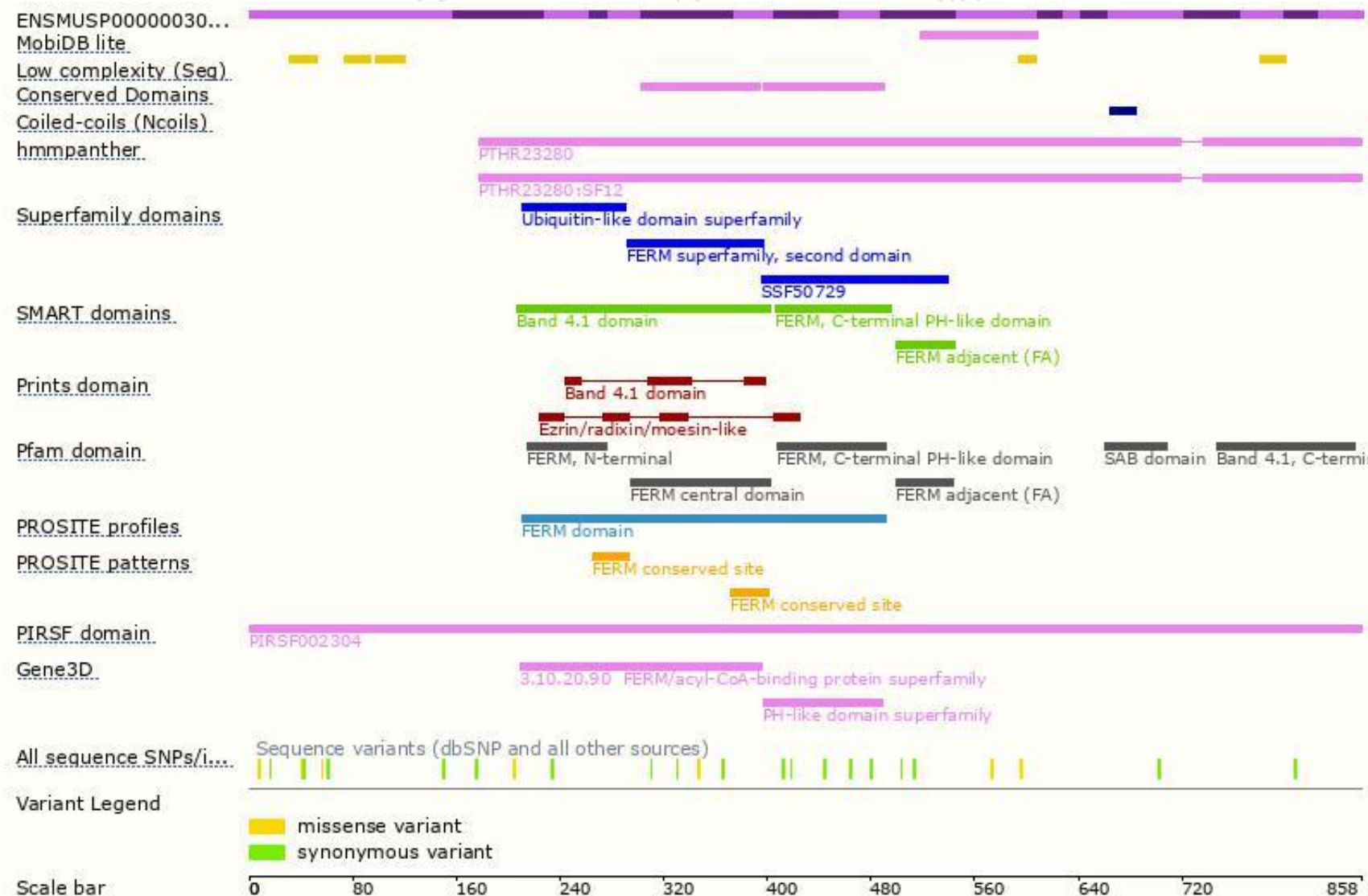
The strategy is based on the design of *Epb41-201* transcript,The transcription is shown below



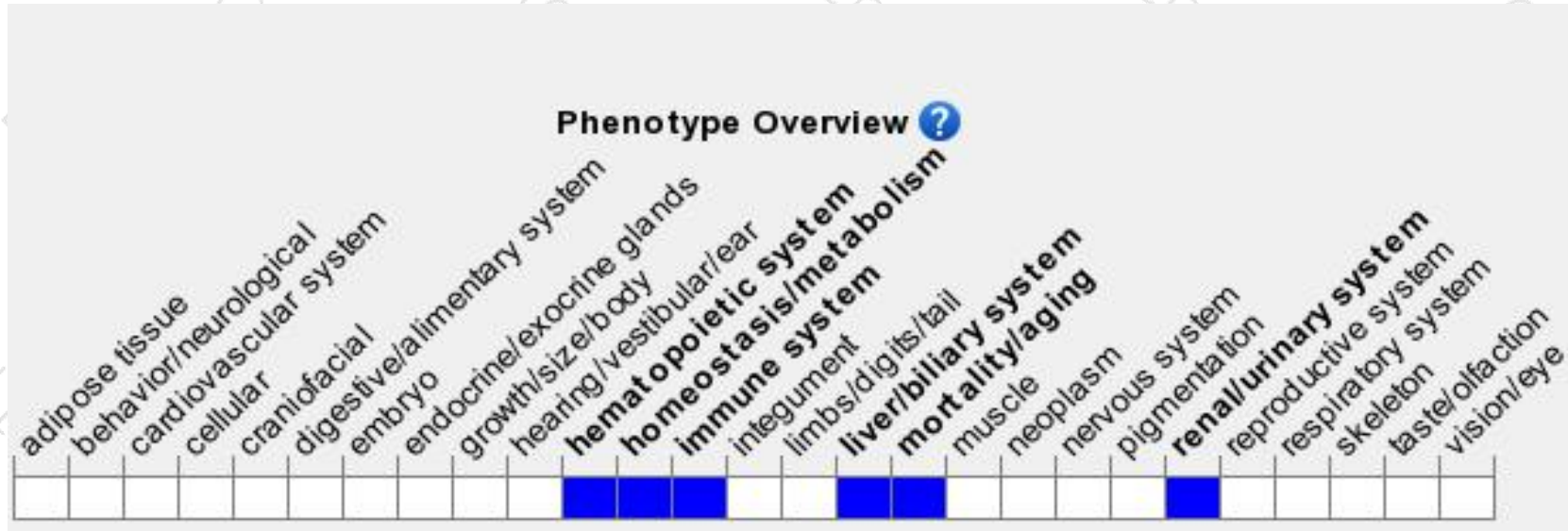
Genomic location distribution



Protein domain



Mouse phenotype description(MGI)



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

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

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