Relt Cas9-CKO Strategy

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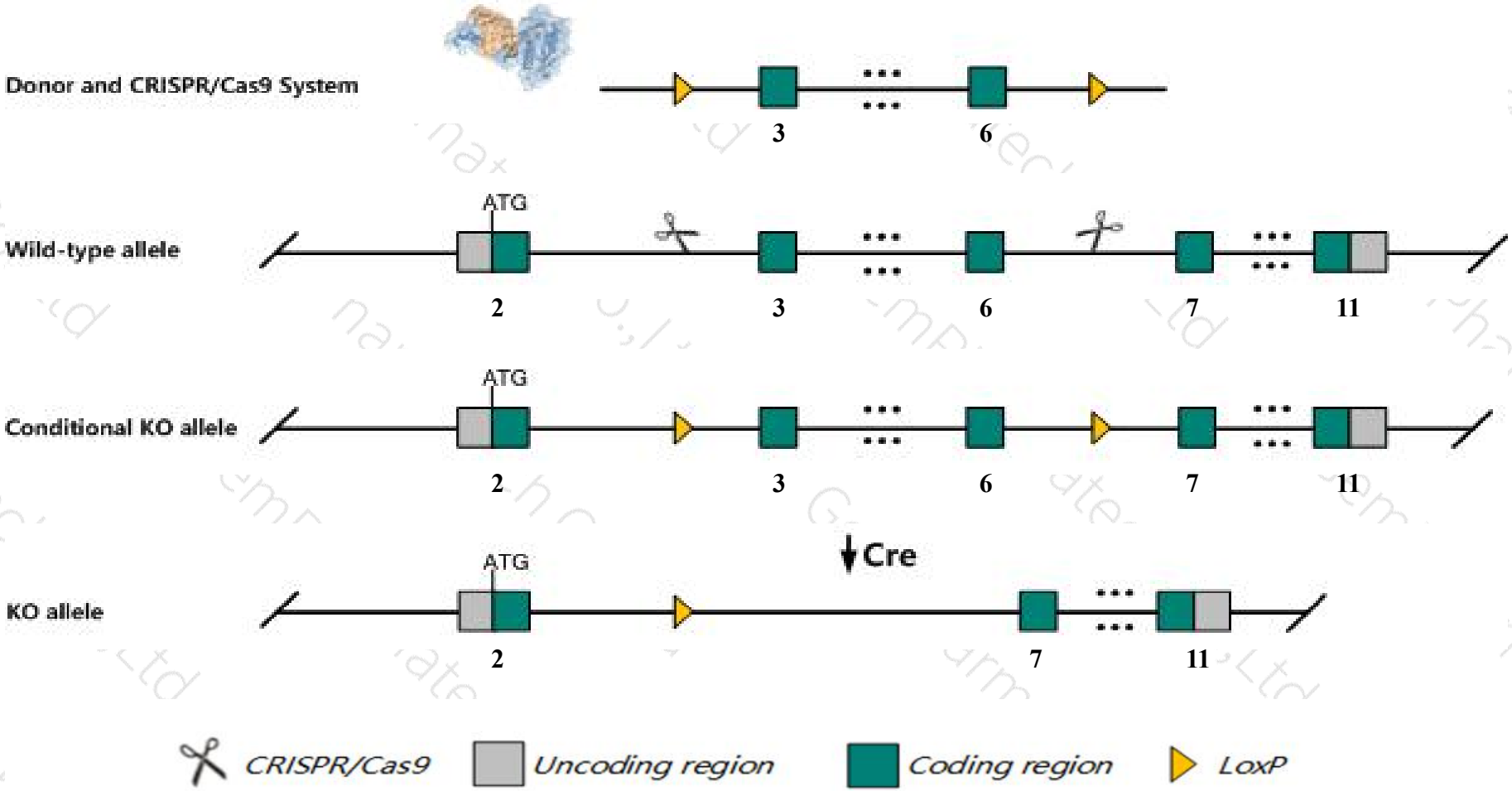
Project Overview

Project Name	***Relt***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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



Technical routes

- The *Relt* gene has 6 transcripts. According to the structure of *Relt* gene, exon3-exon6 of *Relt-201* (ENSMUST00000008462.10) transcript is recommended as the knockout region.The region contains 580bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Relt* 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.

Notice

- According to the existing MGI data,mice with a premature termination codon resulting in a null allele exhibit incisor and molar enamel malformations. homozygous knockout affects t cell proliferation and reduces tumor growth.
- Transcript *Relt-204* may not be affected.
- The *Relt* gene is located on the Chr7. 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）

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Relt RELT tumor necrosis factor receptor [Mus musculus (house mouse)]

Gene ID: 320100, updated on 13-Mar-2020

Summary

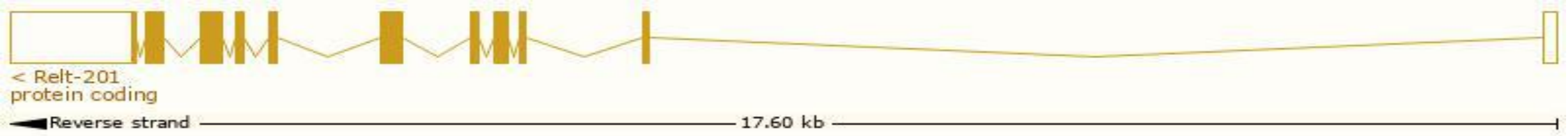
Official Symbol	Relt provided by MGI
Official Full Name	RELT tumor necrosis factor receptor provided by MGI
Primary source	MGI:MGI:2443373
See related	Ensembl:ENSMUSG00000008318
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	E430021K24Rik, Tnfrsf19l
Expression	Broad expression in spleen adult (RPKM 9.5), thymus adult (RPKM 6.5) and 24 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

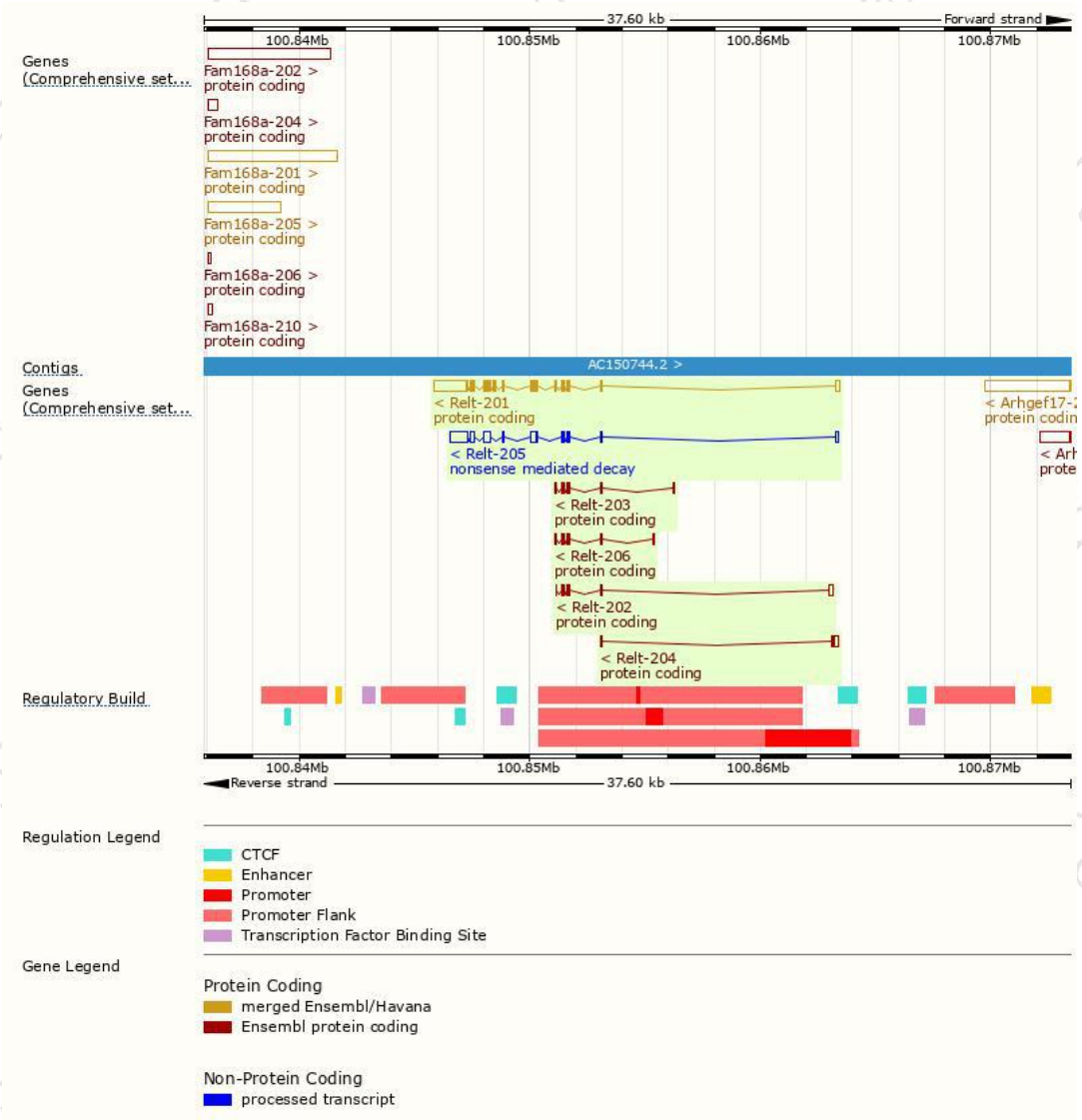
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Relt-201	ENSMUST00000008462.10	2851	436aa	Protein coding	CCDS21506	Q8BX43	TSL:1 GENCODE basic APPRIS P1
Relt-202	ENSMUST00000136231.1	516	119aa	Protein coding	-	D3Z306	CDS 3' incomplete TSL:2
Relt-203	ENSMUST00000139604.7	459	123aa	Protein coding	-	D3YWL9	CDS 3' incomplete TSL:5
Relt-206	ENSMUST00000156855.7	428	123aa	Protein coding	-	D3YWL9	CDS 3' incomplete TSL:5
Relt-204	ENSMUST00000142885.1	346	58aa	Protein coding	-	D3YTN8	CDS 3' incomplete TSL:3
Relt-205	ENSMUST00000155413.8	1966	119aa	Nonsense mediated decay	-	D6RIM1	TSL:1

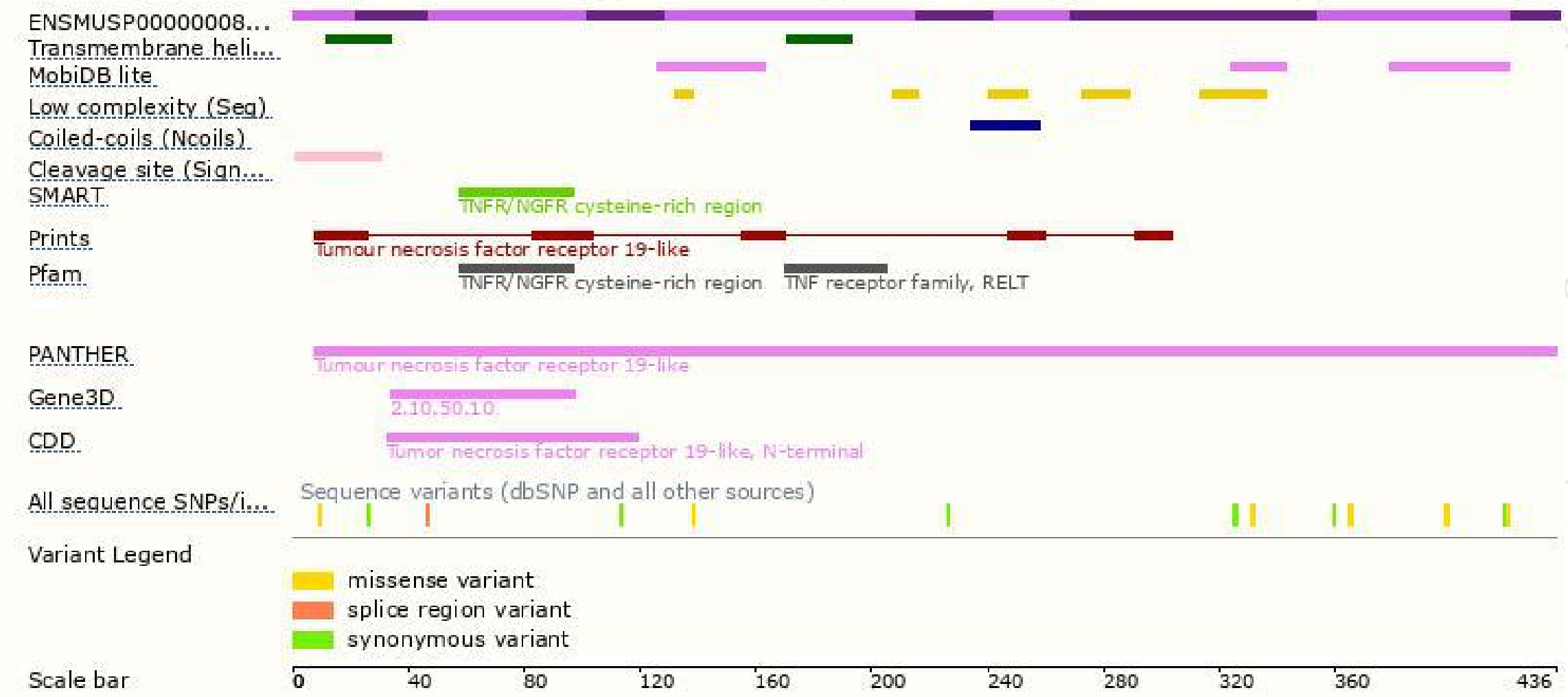
The strategy is based on the design of *Relt-201* transcript,the transcription is shown below:



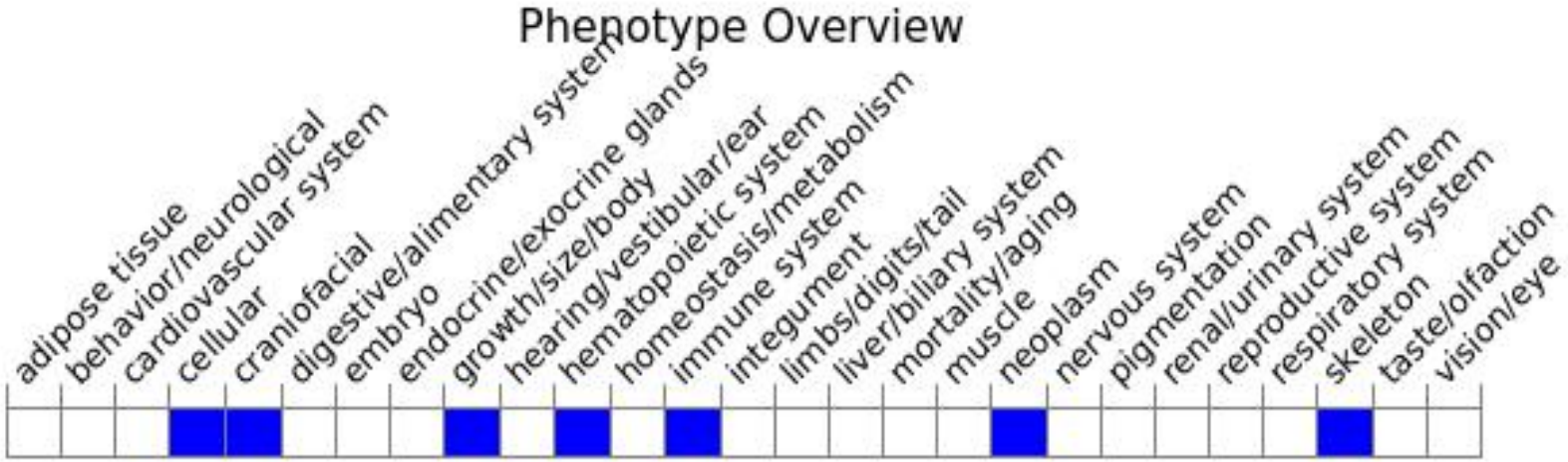
Genomic location distribution



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/).

According to the existing MGI data,mice with a premature termination codon resulting in a null allele exhibit incisor and molar enamel malformations. Homozygous knockout affects T cell proliferation and reduces tumor growth.

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

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