

***Rnf41* Cas9-CKO Strategy**

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Project Overview

Project Name

Rnf41

Project type

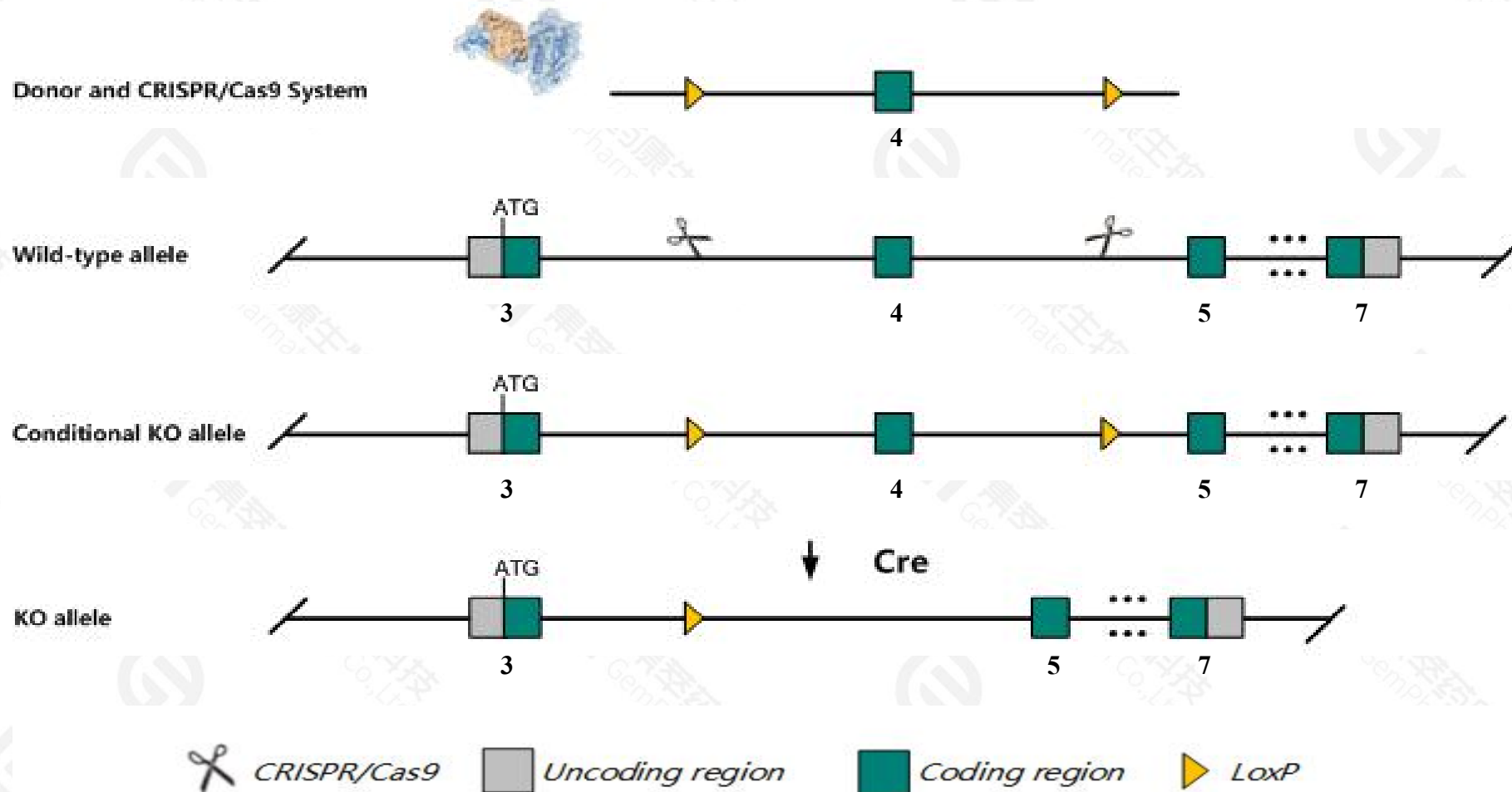
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rnf41* gene. The schematic diagram is as follows:



Technical routes

- The *Rnf41* gene has 4 transcripts. According to the structure of *Rnf41* gene, exon4 of *Rnf41-202*(ENSMUST00000171342.3) transcript is recommended as the knockout region. The region contains 272bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rnf41* 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, mice homozygous for a transgenic gene disruption exhibit male infertility. In contrast, mice homozygous for a gene trapped allele showed no overt phenotypes but show an increase in activation of naive CD8⁺ T cells before TCR engagement.
- *Gm26347* gene will be destroyed.
- The *Rnf41* gene is located on the Chr10. 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)

Rnf41 ring finger protein 41 [Mus musculus (house mouse)]

Gene ID: 67588, updated on 22-Nov-2020

Summary



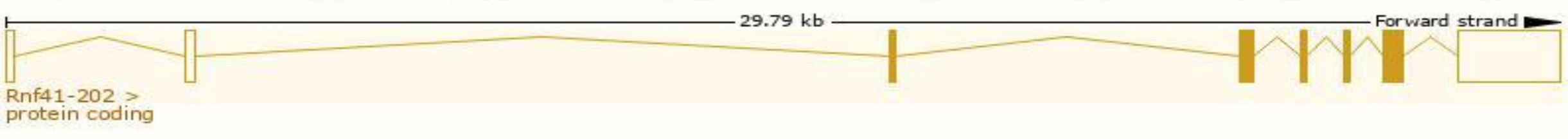
Official Symbol	Rnf41 provided by MGI
Official Full Name	ring finger protein 41 provided by MGI
Primary source	MGI:MGI:1914838
See related	Ensembl:ENSMUSG00000025373
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2210404G21Rik, 4930511A05Rik, 4933415P08Rik, D10Ertd722, D10Ertd722e, FL, FLRF, Nr, Nrdp1
Expression	Ubiquitous expression in testis adult (RPKM 22.7), CNS E18 (RPKM 16.8) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

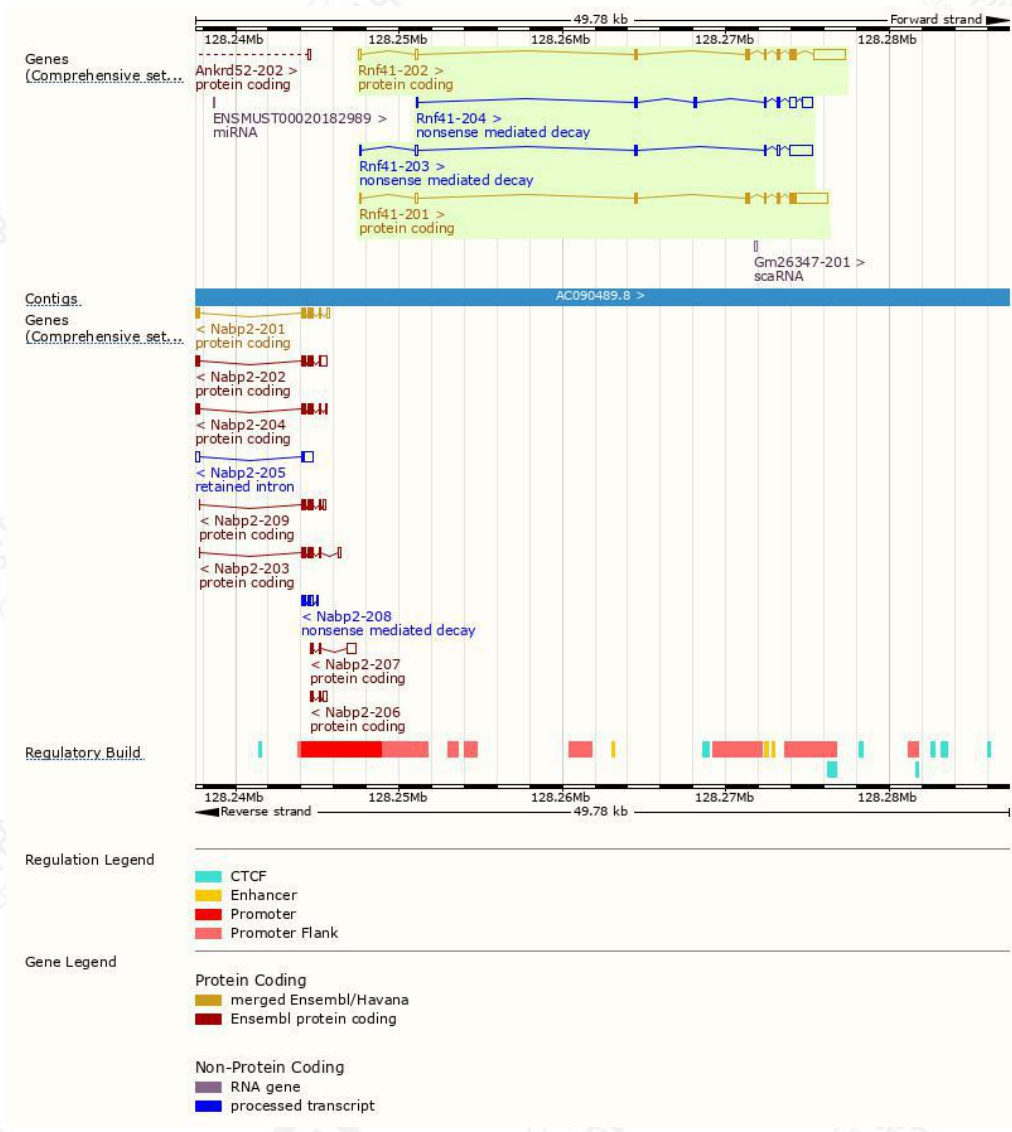
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rnf41-202	ENSMUST00000171342.3	3283	317aa	Protein coding	CCDS24277		TSL:1 , GENCODE basic , APPRIS P1 ,
Rnf41-201	ENSMUST00000096386.13	3144	317aa	Protein coding	CCDS24277		TSL:1 , GENCODE basic , APPRIS P1 ,
Rnf41-203	ENSMUST00000217826.2	1984	36aa	Nonsense mediated decay	-		TSL:1 ,
Rnf41-204	ENSMUST00000218371.2	1594	122aa	Nonsense mediated decay	-		TSL:1 ,

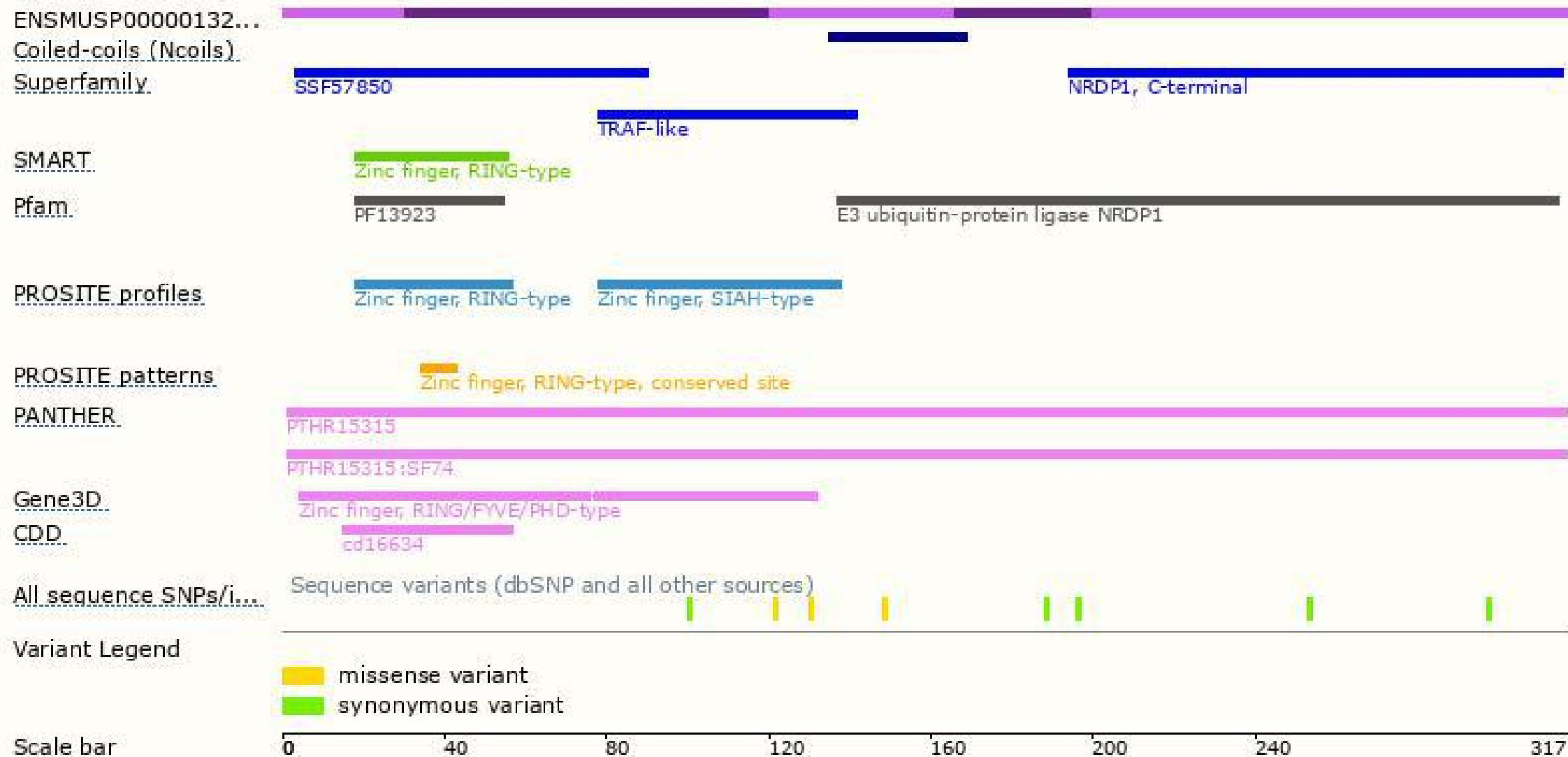
The strategy is based on the design of *Rnf41-202* transcript,the transcription is shown below:



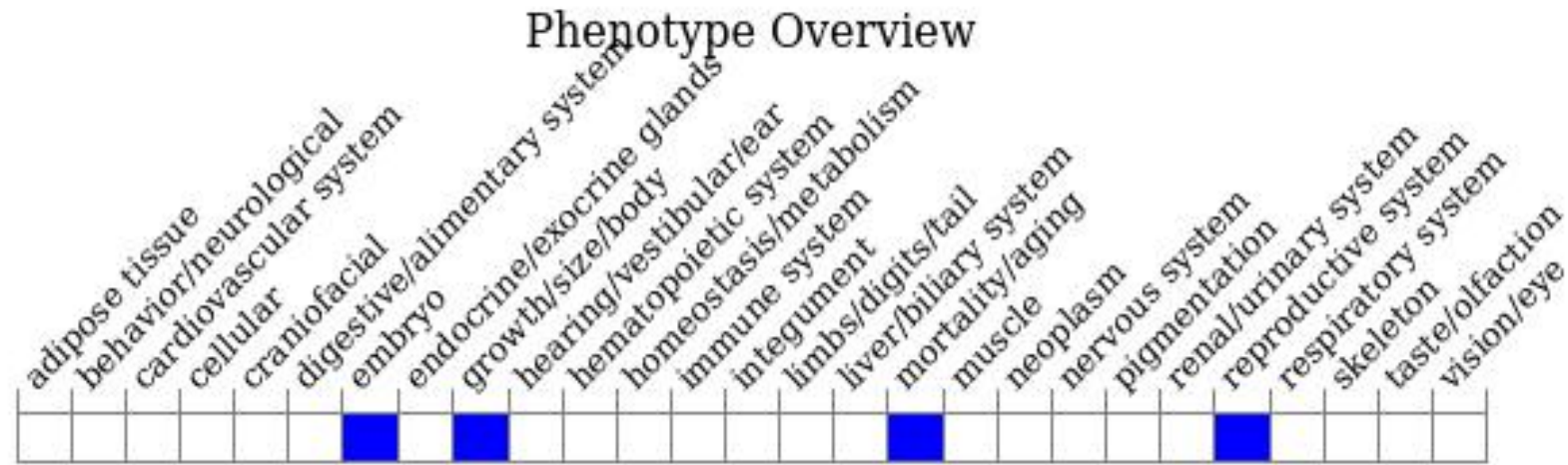
Genomic location distribution



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 homozygous for a transgenic gene disruption exhibit male infertility. In contrast, mice homozygous for a gene trapped allele showed no overt phenotypes but show an increase in activation of naive CD8⁺ T cells before TCR engagement.

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