

Ctcf1 Cas9-CKO Strategy

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Reviewer:

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Design Date:

2020-2-19

Project Overview

Project Name

Ctcf1

Project type

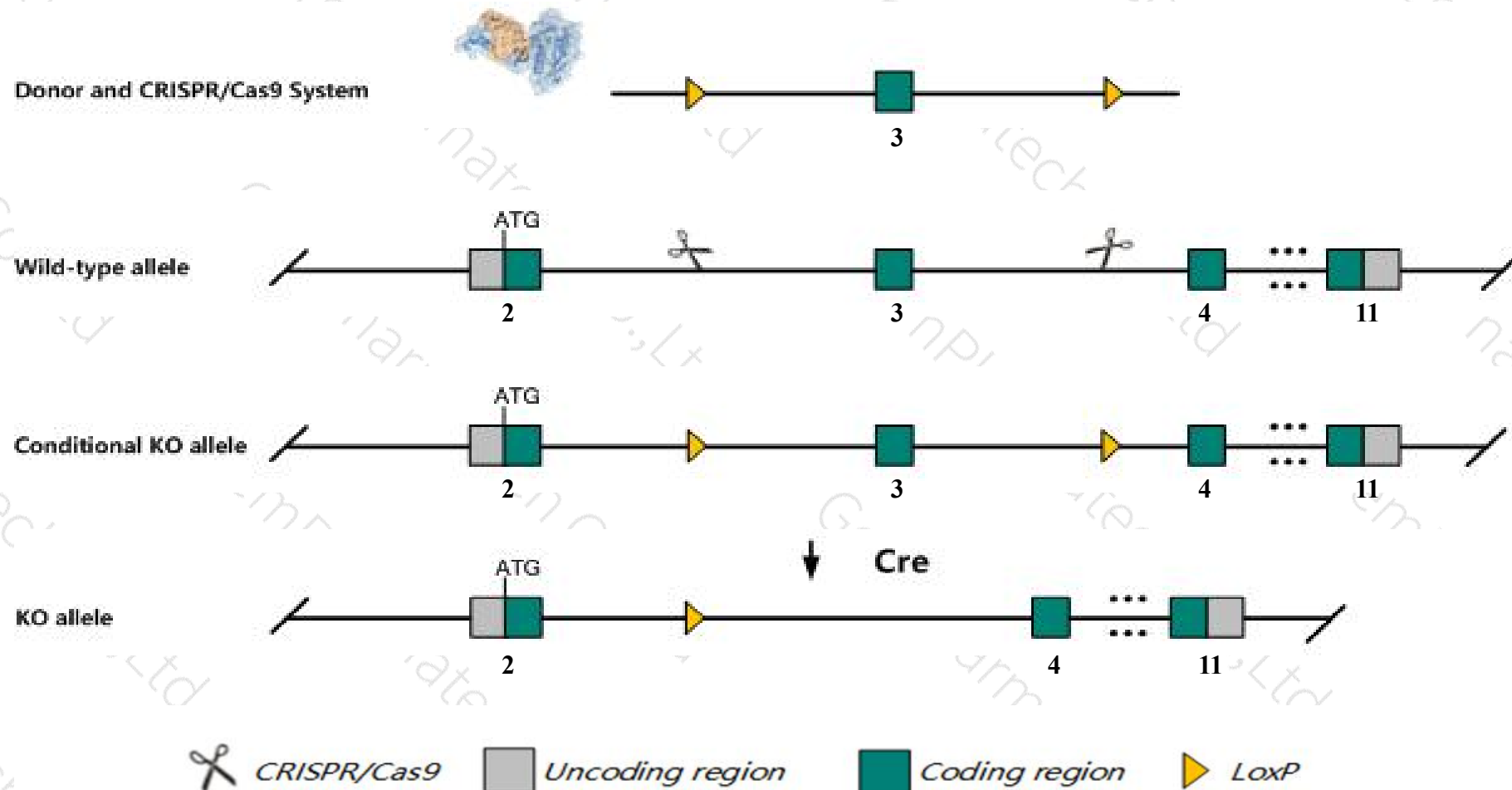
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Ctcfl* gene has 2 transcripts. According to the structure of *Ctcfl* gene, exon3 of *Ctcfl*-201 (ENSMUST00000094287.9) transcript is recommended as the knockout region. The region contains 226bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ctcfl* 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 knock-out allele exhibit small testes, delayed spermatid development, and increased male germ cell apoptosis without affecting fertility.
- The *Ctcf1* gene is located on the Chr2. 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)

Ctcf1 CCCTC-binding factor (zinc finger protein)-like [*Mus musculus* (house mouse)]

Gene ID: 664799, updated on 10-Oct-2019

Summary

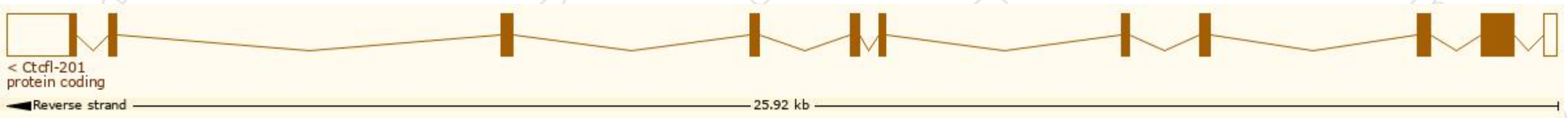
Official Symbol	Ctcf1 provided by MGI
Official Full Name	CCCTC-binding factor (zinc finger protein)-like provided by MGI
Primary source	MGI:MGI:3652571
See related	Ensembl:ENSMUSG00000070495
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Boris
Expression	Biased expression in subcutaneous fat pad adult (RPKM 1.3), genital fat pad adult (RPKM 1.1) and 11 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

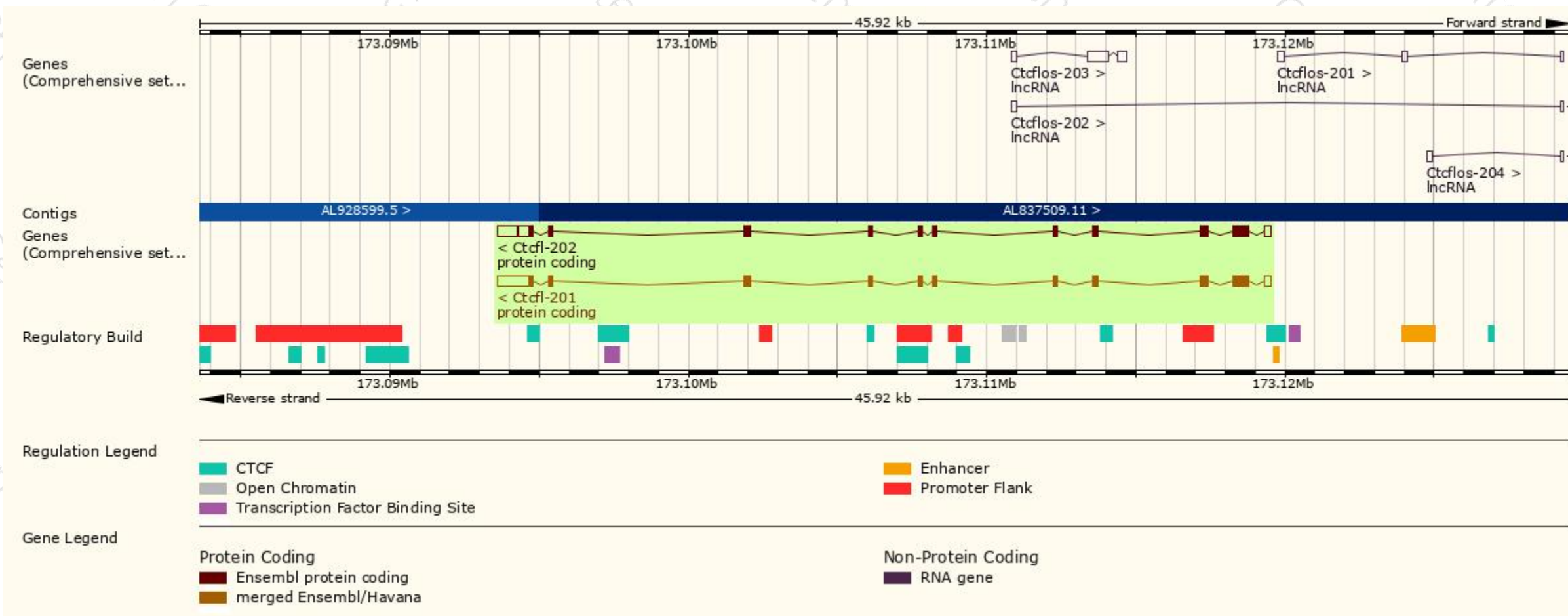
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ctcf1-201	ENSMUST00000094287.9	3217	636aa	Protein coding	CCDS38349	A2APF3	TSL:5 Gencode basic APPRIS P1
Ctcf1-202	ENSMUST00000179693.1	3174	636aa	Protein coding	CCDS38349	A2APF3	TSL:5 Gencode basic APPRIS P1

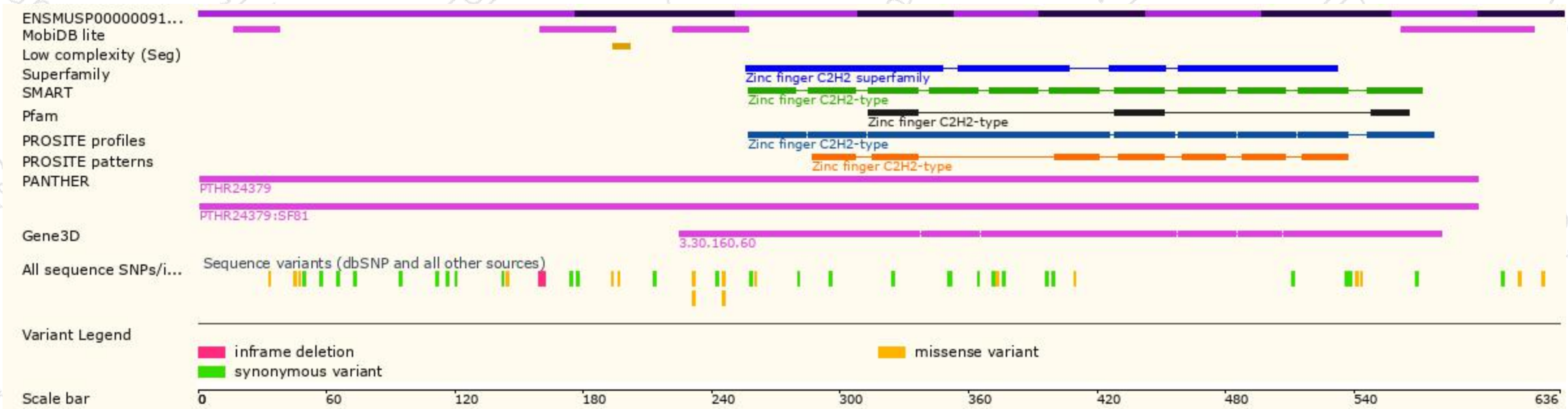
The strategy is based on the design of *Ctcf1-201* transcript, the transcription is shown below:



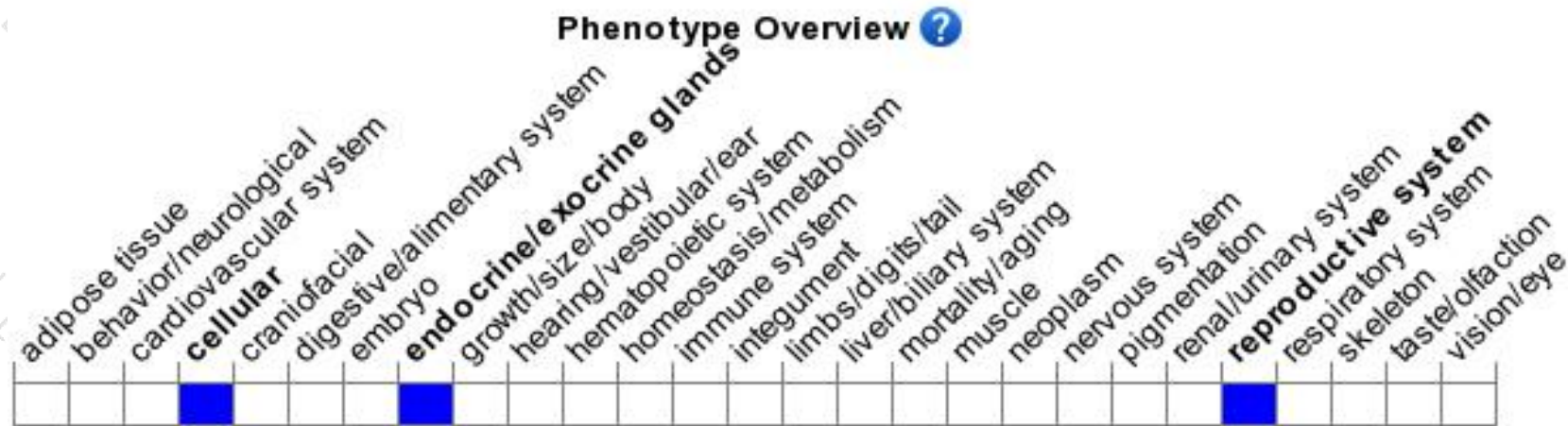
Genomic location distribution



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 homozygous for a knock-out allele exhibit small testes, delayed spermatid development, and increased male germ cell apoptosis without affecting fertility.

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

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