

***Tdrd12* Cas9-CKO Strategy**

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Project Overview

Project Name

Tdrd12

Project type

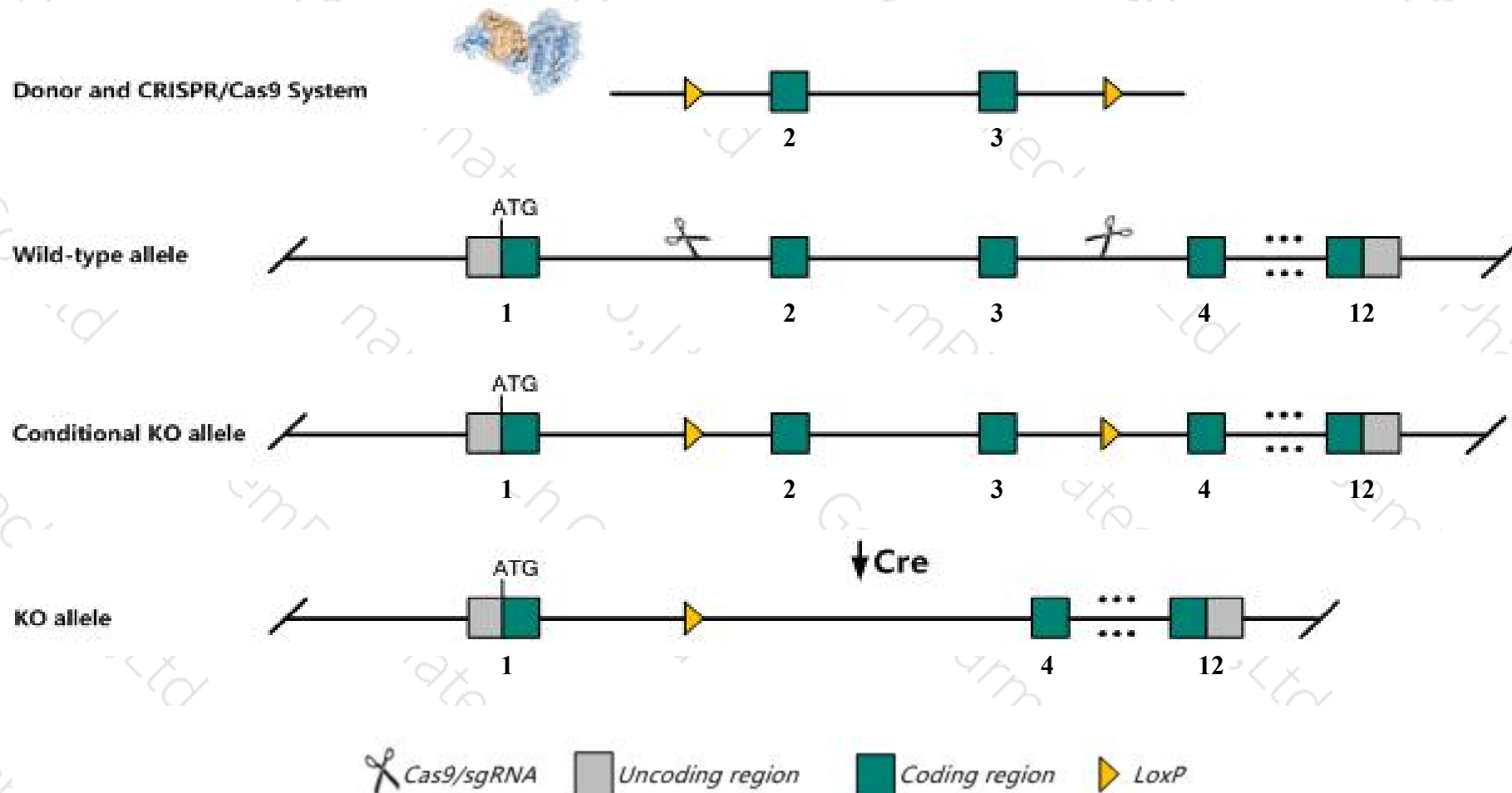
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Tdrd12* gene. The schematic diagram is as follows:



- The *Tdrd12* gene has 6 transcripts. According to the structure of *Tdrd12* gene, exon2-exon3 of *Tdrd12-201*(ENSMUST00000032701.13) transcript is recommended as the knockout region. The region contains 296bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Tdrd12* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA 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 was 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, homozygous males are infertile with small testes. Spermatogenesis is arrested predominantly at the pachytene spermatocyte stage. Retrotransposon hopping is derepressed in germ cells.
- The *Tdrd12* 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)

Tdrd12 tudor domain containing 12 [Mus musculus (house mouse)]

Gene ID: 71981, updated on 13-Mar-2020

Summary



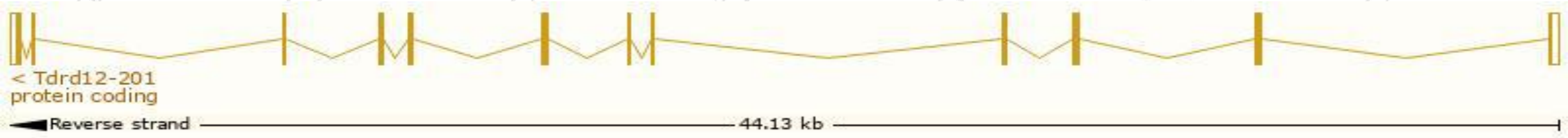
Official Symbol	Tdrd12 provided by MGI
Official Full Name	tudor domain containing 12 provided by MGI
Primary source	MGI:MGI:1919231
See related	Ensembl:ENSMUSG00000030491
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2410004F06Rik, 2410070K17Rik, ECAT8, EG434165, G1-476-14, repro23
Expression	Biased expression in testis adult (RPKM 4.7), kidney adult (RPKM 0.5) and 2 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

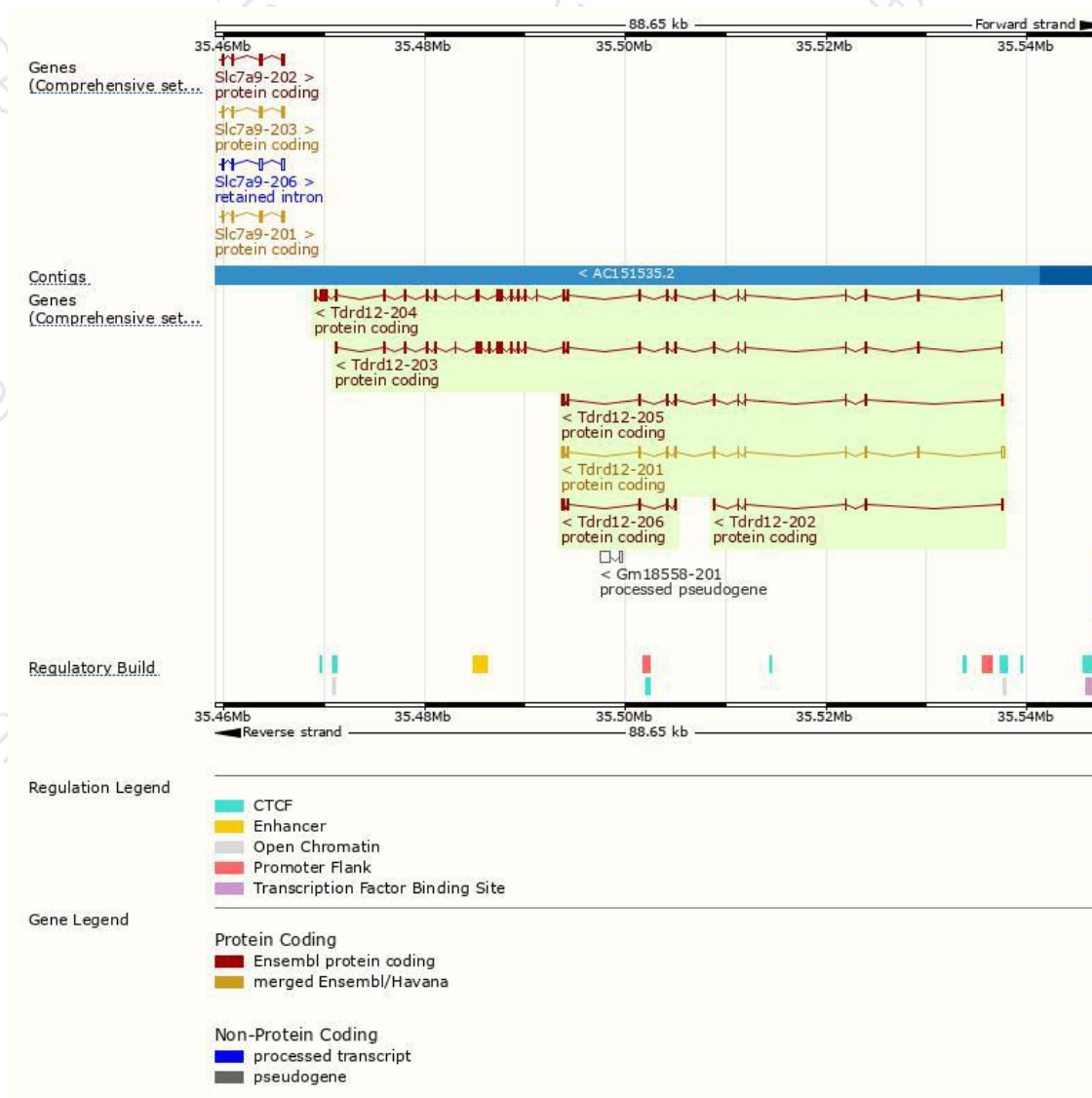
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Tdrd12-201	ENSMUST00000032701.13	1620	407aa	Protein coding	CCDS21151	Q9CWU0	TSL:1 GENCODE basic
Tdrd12-204	ENSMUST00000193633.5	3651	1216aa	Protein coding	-	A0A0A6YX17	TSL:5 GENCODE basic APPRIS P1
Tdrd12-203	ENSMUST00000187190.6	3341	1113aa	Protein coding	-	A0A087WQS9	CDS 3' incomplete TSL:5
Tdrd12-205	ENSMUST00000205407.1	1412	354aa	Protein coding	-	A0A0U1RNL0	TSL:5 GENCODE basic
Tdrd12-202	ENSMUST00000127472.2	769	204aa	Protein coding	-	D3YZI6	CDS 3' incomplete TSL:3
Tdrd12-206	ENSMUST00000206641.1	543	127aa	Protein coding	-	A0A0U1RNP6	CDS 5' incomplete TSL:3

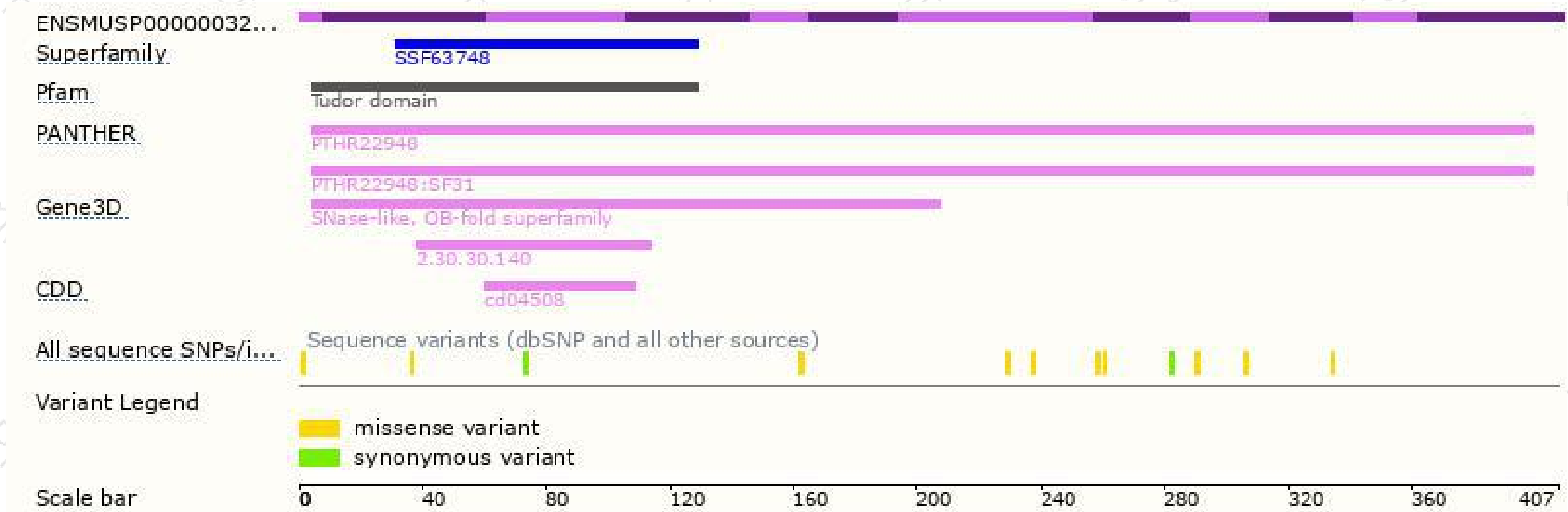
The strategy is based on the design of *Tdrd12-201* transcript,the transcription is shown below:



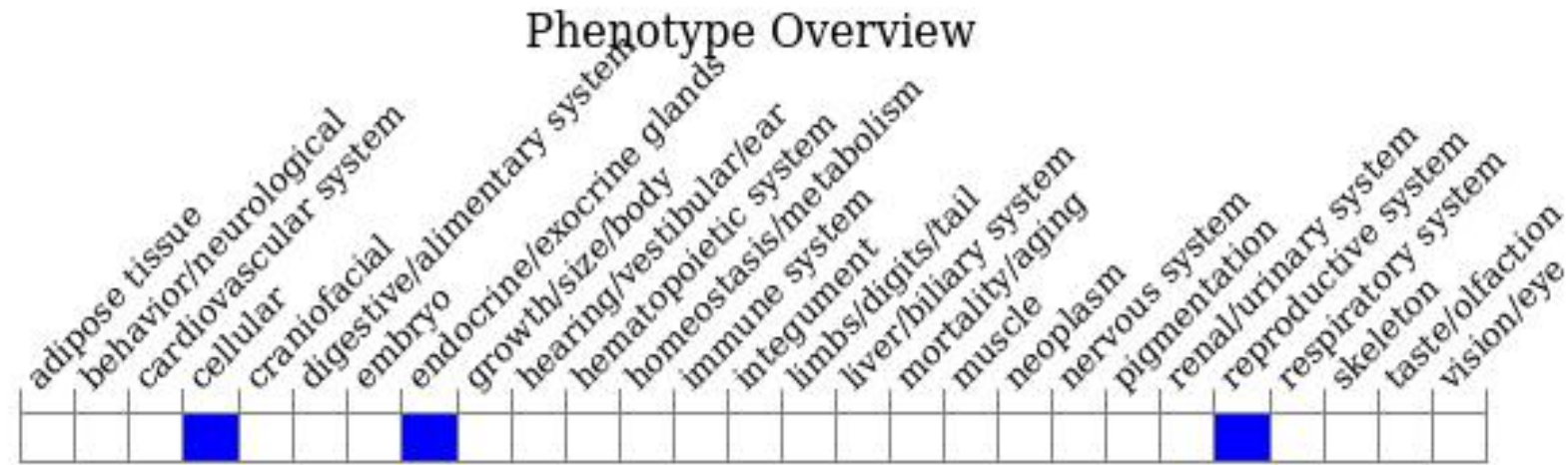
Genomic location distribution



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, homozygous males are infertile with small testes. Spermatogenesis is arrested predominantly at the pachytene spermatocyte stage. Retrotransposon hopping is derepressed in germ cells.

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

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