

Tnfrsf18 Cas9-CKO Strategy

Designer: Jinling Wang

Design Date: 2019-7-19

Project Overview

Project Name

Tnfrsf18

Project type

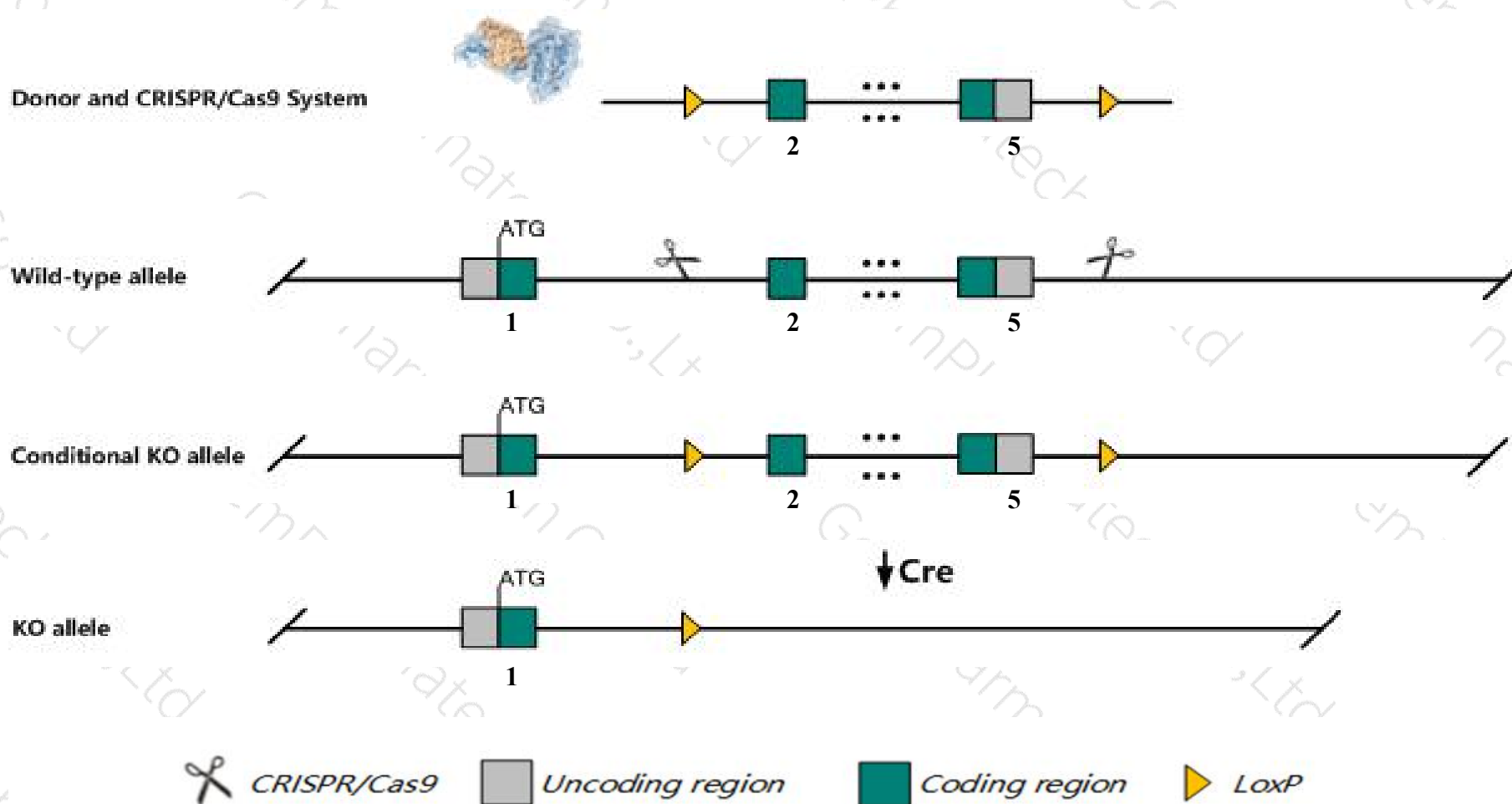
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Tnfrsf18* gene. The schematic diagram is as follows:



Technical routes

- The *Tnfrsf18* gene has 6 transcripts. According to the structure of *Tnfrsf18* gene, exon2-exon5 of *Tnfrsf18-202* (ENSMUST00000103173.9) transcript is recommended as the knockout region. The region contains 536bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Tnfrsf18* 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 targeted mutation display dysregulation of T-cell receptor/CD3-driven T-cell activation and programmed cell death.
- The *Tnfrsf18* 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)

Tnfrsf18 tumor necrosis factor receptor superfamily, member 18 [Mus musculus (house mouse)]

Gene ID: 21936, updated on 31-Jan-2019

Summary

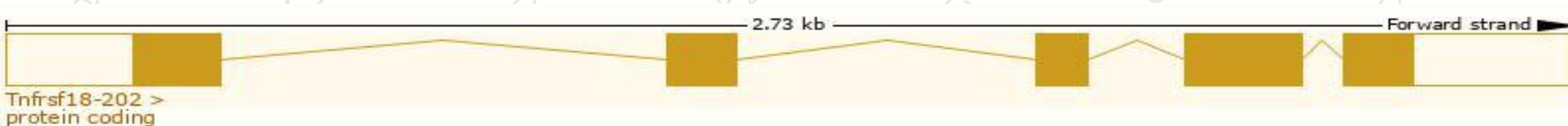
Official Symbol	Tnfrsf18 provided by MGI
Official Full Name	tumor necrosis factor receptor superfamily, member 18 provided by MGI
Primary source	MGI:MGI:894675
See related	Ensembl:ENSMUSG00000041954
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AITR, Gitr
Expression	Ubiquitous expression in thymus adult (RPKM 23.3), mammary gland adult (RPKM 14.4) and 25 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

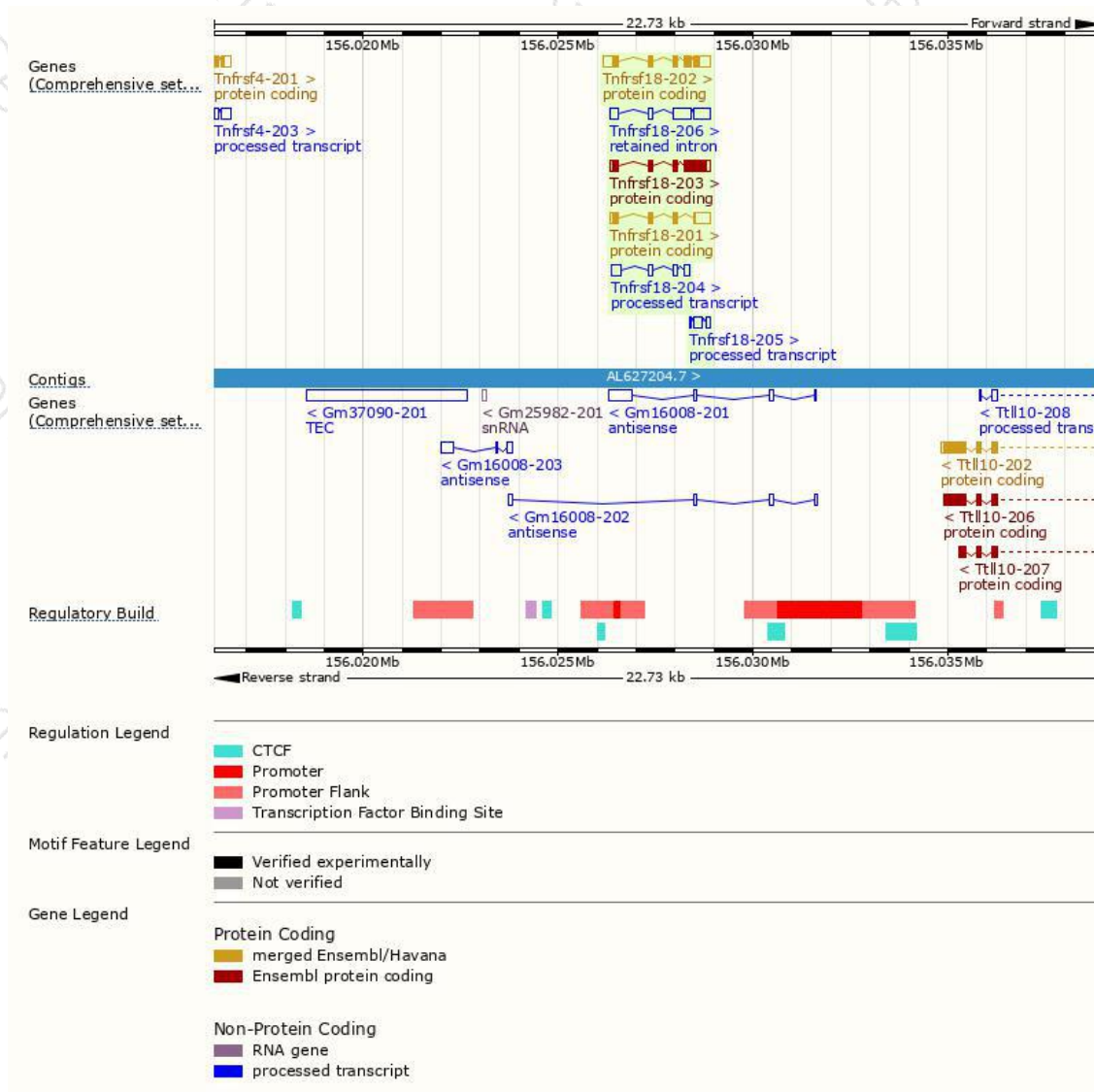
The gene has 6 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Tnfrsf18-202	ENSMUST00000103173.9	1189	228aa	Protein coding	CCDS19056	O35714 Q540M6	TSL:1 GENCODE basic APPRIS P2
Tnfrsf18-201	ENSMUST00000040274.12	804	132aa	Protein coding	CCDS19057	O35714	TSL:1 GENCODE basic
Tnfrsf18-203	ENSMUST00000122001.2	1078	314aa	Protein coding	-	B1ASL6	TSL:1 GENCODE basic APPRIS ALT2
Tnfrsf18-204	ENSMUST00000124339.1	615	No protein	Processed transcript	-	-	TSL:3
Tnfrsf18-205	ENSMUST00000125100.1	328	No protein	Processed transcript	-	-	TSL:5
Tnfrsf18-206	ENSMUST00000141260.7	1188	No protein	Retained intron	-	-	TSL:5

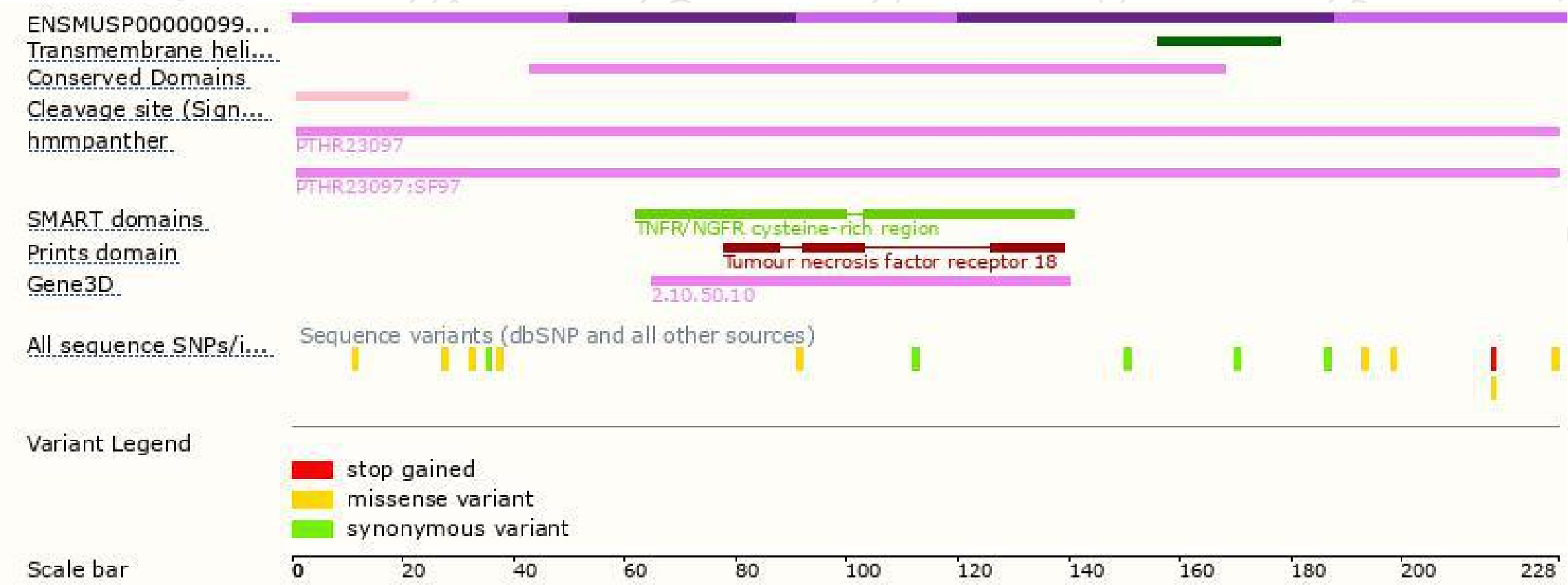
The strategy is based on the design of *Tnfrsf18-202* transcript, The transcription is shown below



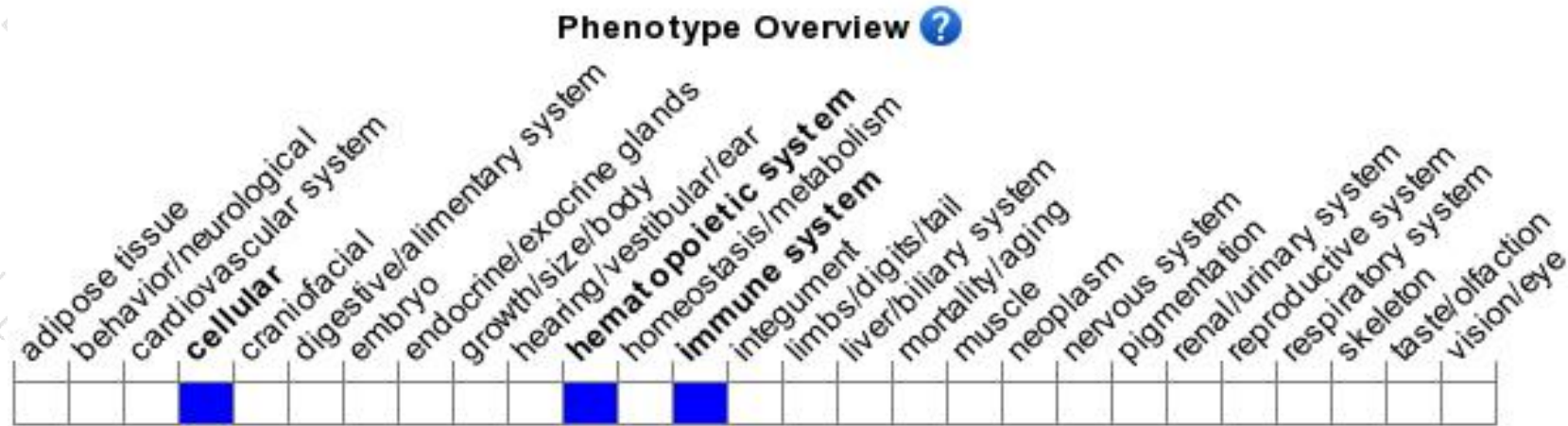
Genomic location distribution



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 targeted mutation display dysregulation of T-cell receptor/CD3-driven T-cell activation and programmed cell death.

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

Tel: 400-9660890

