

F2rl3 Cas9-KO Strategy

Designer:

Shilei Zhu

Design Date:

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Project Overview

Project Name

F2rl3

Project type

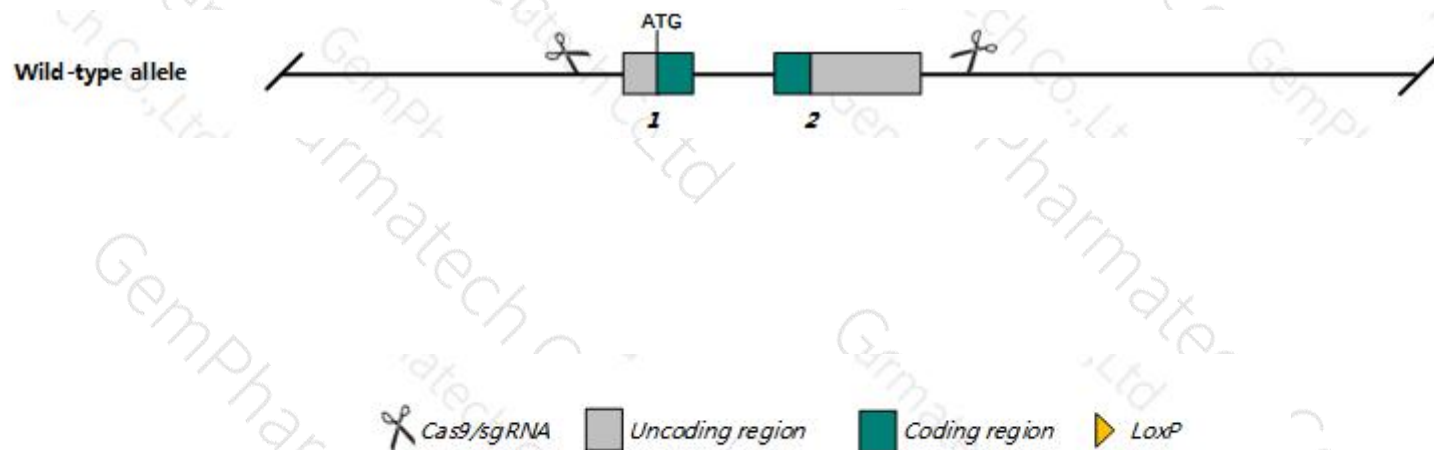
Cas9-KO

Strain background

C57BL/6J

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *F2rl3* gene. The schematic diagram is as follows:



Technical routes

- The *F2rl3* gene has 2 transcripts. According to the structure of *F2rl3* gene, exon1-2 of *F2rl3*-201 (ENSMUST00000058099.8) transcript is recommended as the knockout region. The region contains all coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *F2rl3* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed. Cas9, gRNA and Donor were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.

- According to the existing MGI data , Homozygous mutation of this gene results in prolonged bleeding time and protection against thrombosis.
- The *F2rl3* gene is located on the Chr8. 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 the loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)

F2rl3 coagulation factor II (thrombin) receptor-like 3 [*Mus musculus* (house mouse)]

Gene ID: 14065, updated on 25-Jun-2019

Summary

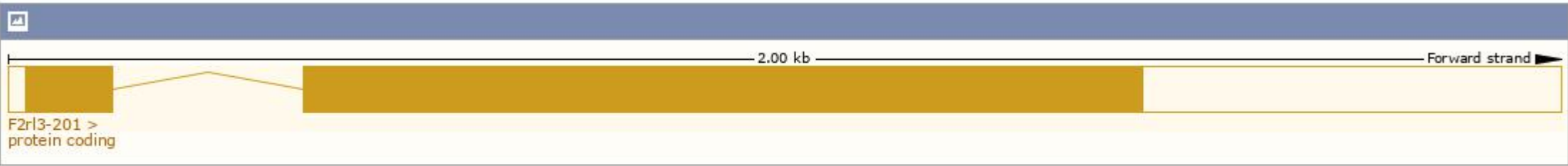
Official Symbol	F2rl3 provided by MGI
Official Full Name	coagulation factor II (thrombin) receptor-like 3 provided by MGI
Primary source	MGI:MGI:1298207
See related	Ensembl:ENSMUSG00000050147
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	PAR4
Summary	This gene encodes a member of the protease-activated receptor subfamily, part of the G-protein coupled receptor 1 family of proteins. The encoded receptor is proteolytically processed to reveal an extracellular N-terminal tethered ligand that binds to and activates the receptor. This receptor plays a role in blood coagulation, inflammation and response to pain. Mice lacking a functional copy of this gene exhibit impaired platelet activation and prolonged bleeding times. [provided by RefSeq, Sep 2016]
Expression	Broad expression in spleen adult (RPKM 9.1), thymus adult (RPKM 4.3) and 20 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

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

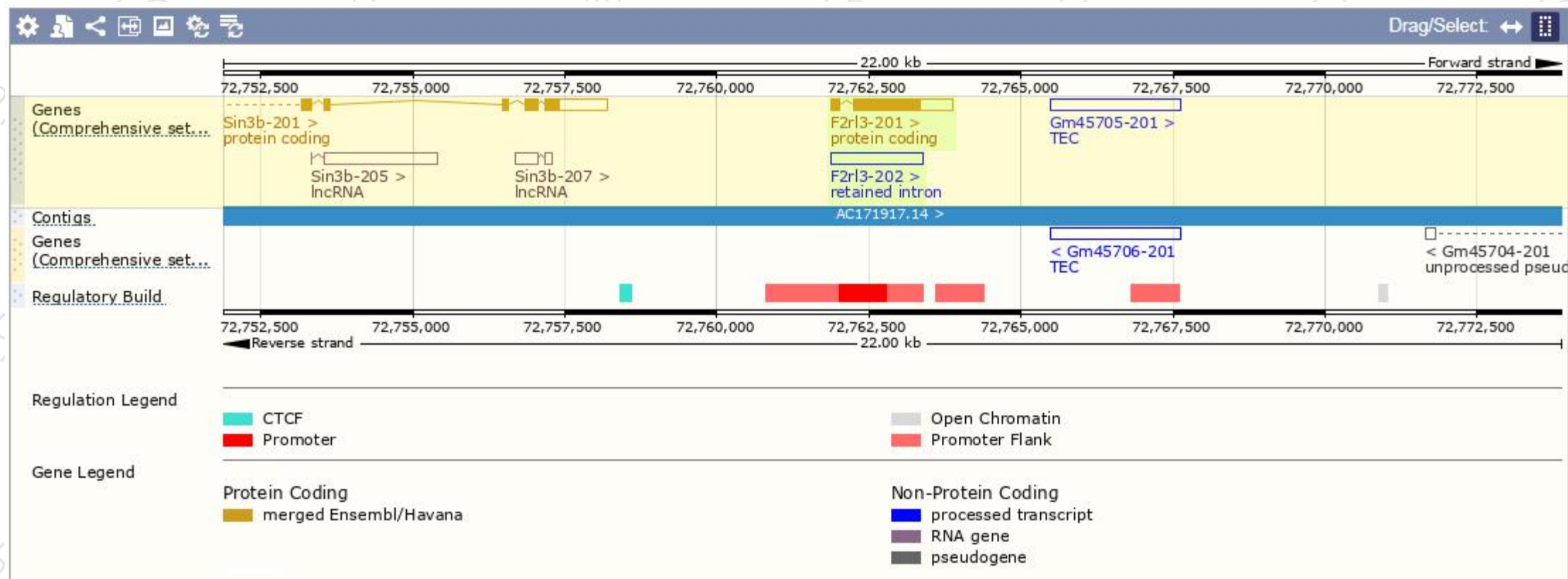
Show/hide columns (1 hidden)		Filter					
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
F2rl3-201	ENSMUST00000058099.8	1751	396aa	Protein coding	CCDS22419	O88634	TSL:1 Gencode basic APPRIS P1
F2rl3-202	ENSMUST00000212755.1	1504	No protein	Retained intron	-	-	TSL:NA

The strategy is based on the design of *F2rl3*-201 transcript, The transcription is shown below

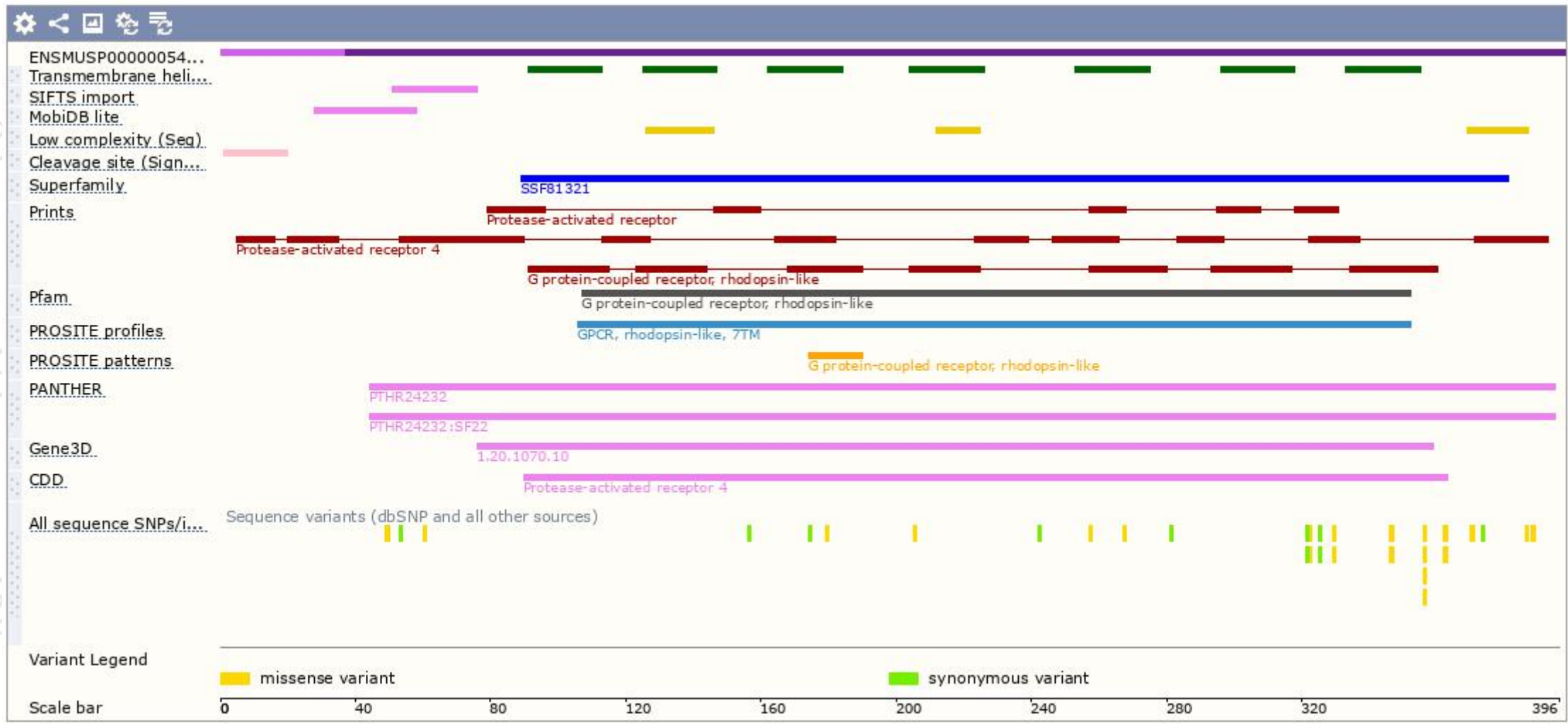


Statistics Exons: 2, Coding exons: 2, Transcript length: 1,751 bps, Translation length: 396 residues

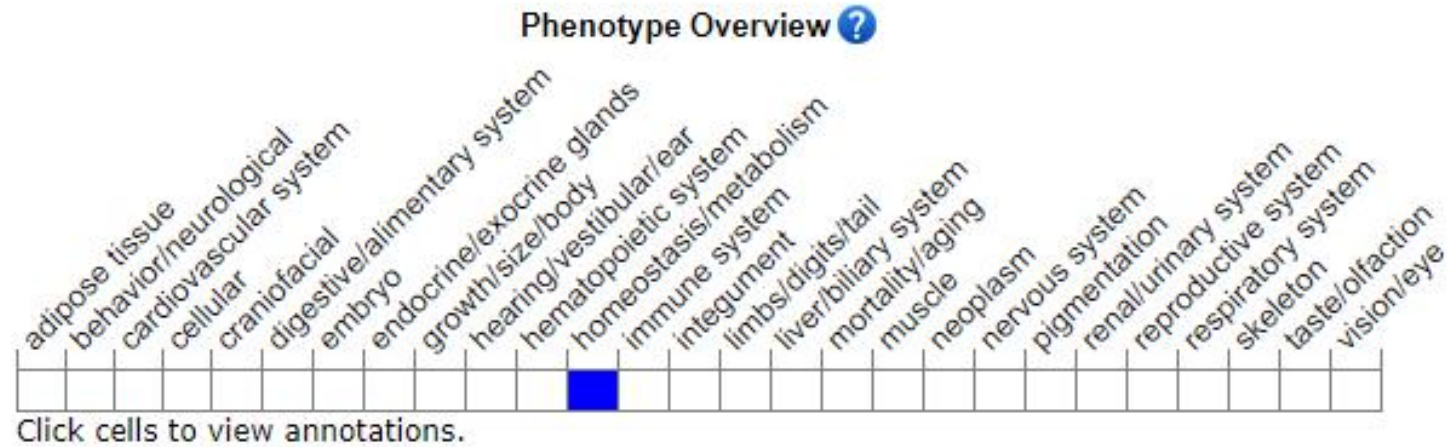
Genomic location distribution



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/>) .

Homozygous mutation of this gene results in prolonged bleeding time and protection against thrombosis.

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

Tel: 400-9660890



集萃药康生物科技

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