Frmd7 Cas9-CKO *Strategy*

Designer: **Daohua Xu**
Reviewer： **Huimin Su**

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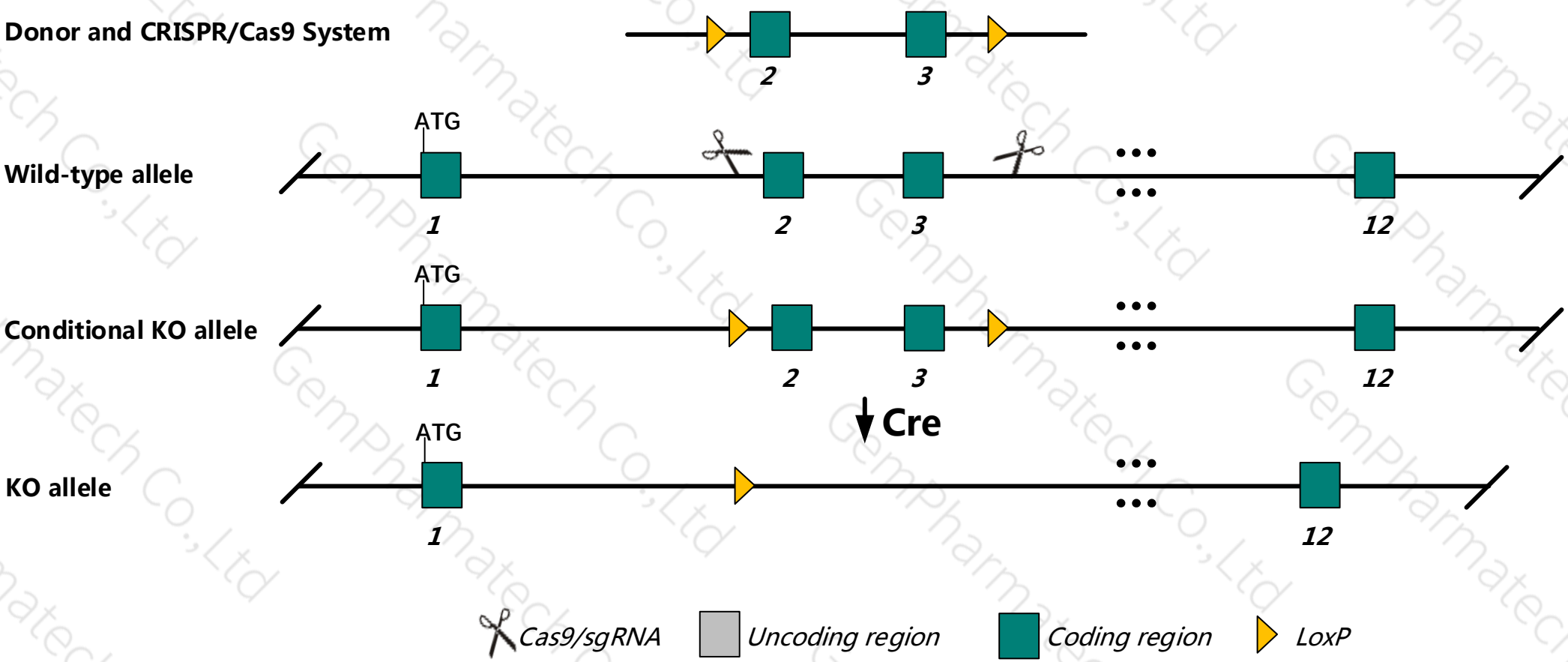
Project Overview

Project Name	***Frmd7***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Frmd7* gene. The schematic diagram is as follows:



Technical routes

- The *Frmd7* gene has 2 transcripts.According to the structure of *Frmd7* gene, exon2-exon3 of *Frmd7*-201 (
- ENSMUST00000060650.6) transcript is recommended as the knockout region.The region contains 148bp coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Frmd7* 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 or cell types.

Notice

- The *Frmd7* gene is located on the ChrX. 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）

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Frmd7 FERM domain containing 7 [*Mus musculus* (house mouse)]

Gene ID: 385354, updated on 13-Mar-2020

Summary

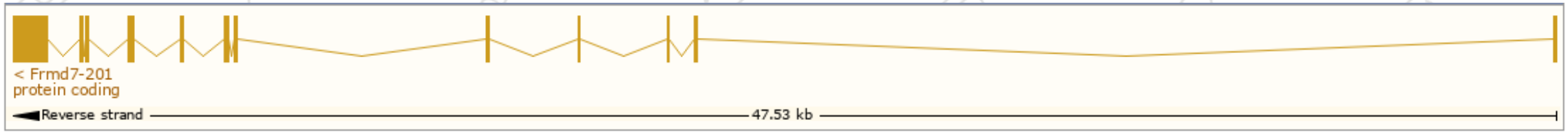
Official Symbol	Frmd7 provided by MGI
Official Full Name	FERM domain containing 7 provided by MGI
Primary source	MGI:MGI:2686379
See related	Ensembl:ENSMUSG00000036131
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Gm1533; EG665849
Expression	Biased expression in frontal lobe adult (RPKM 9.1), limb E14.5 (RPKM 0.9) and 2 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

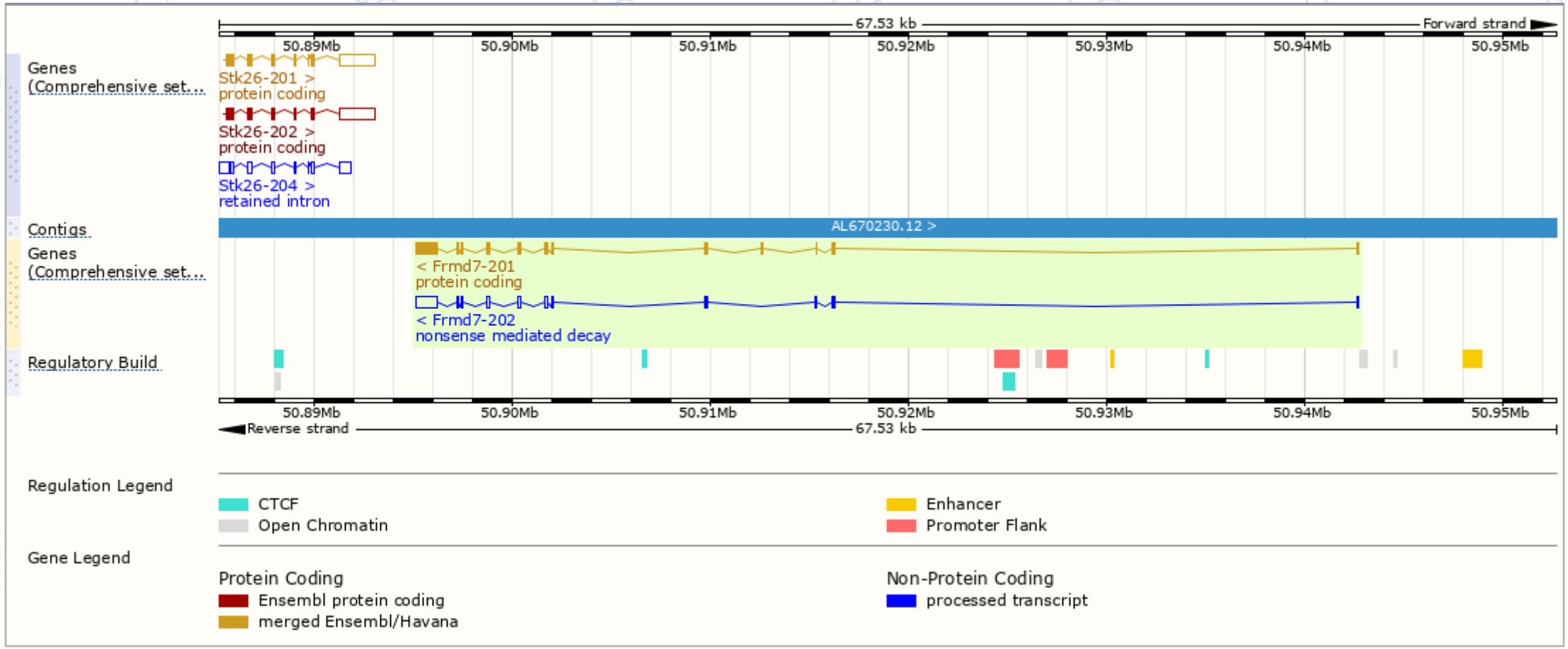
The gene has 2 transcripts, and all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Frmd7-201	ENSMUST00000060650.6	2112	703aa	Protein coding	CCDS53067	A2AD83	TSL:5 GENCODE basic APPRIS P1
Frmd7-202	ENSMUST00000164311.6	2063	85aa	Nonsense mediated decay	-	E9PXK8	TSL:5

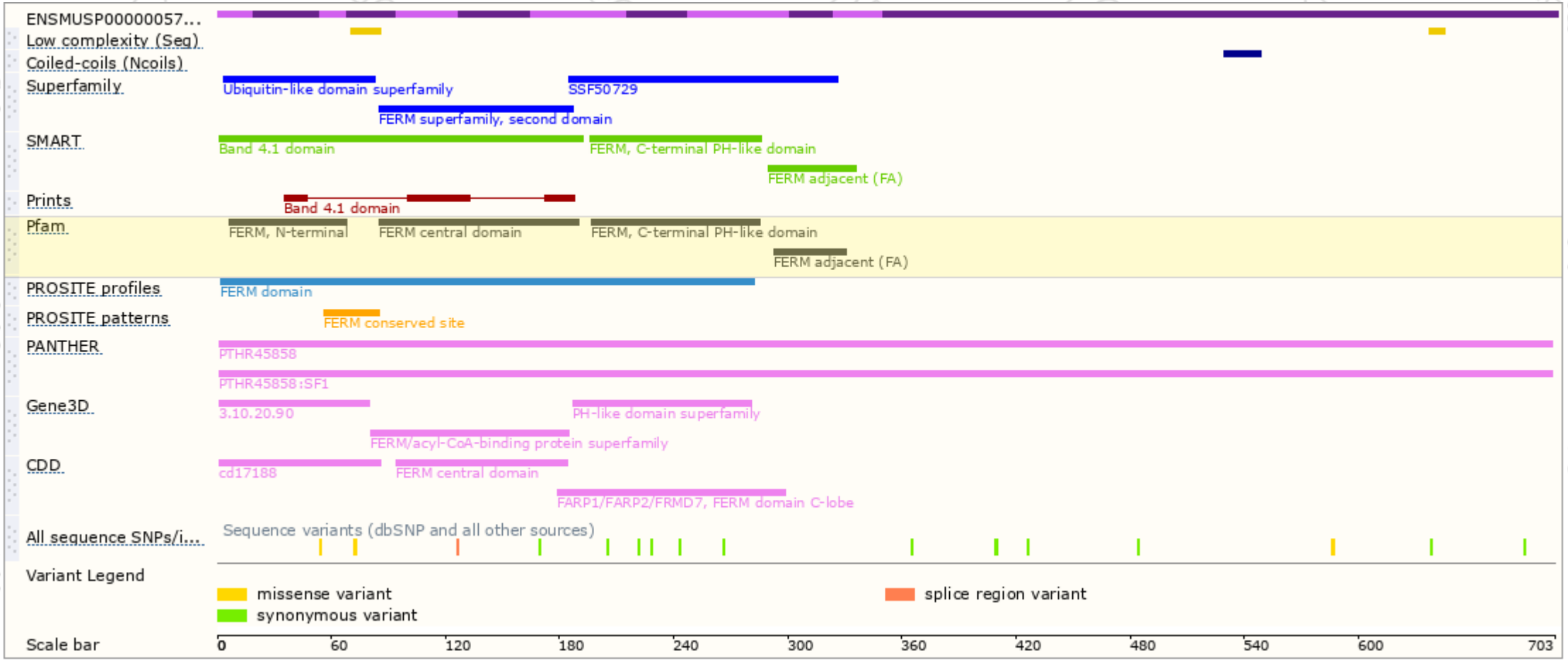
The strategy is based on the design of *Frmd7*-201 transcript,The transcription is shown below



Genomic location distribution



Protein domain



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

Tel: 025-5864 1534

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GemPharmatech Co.,Ltd