Cmtm1 Cas9-CKO Strategy

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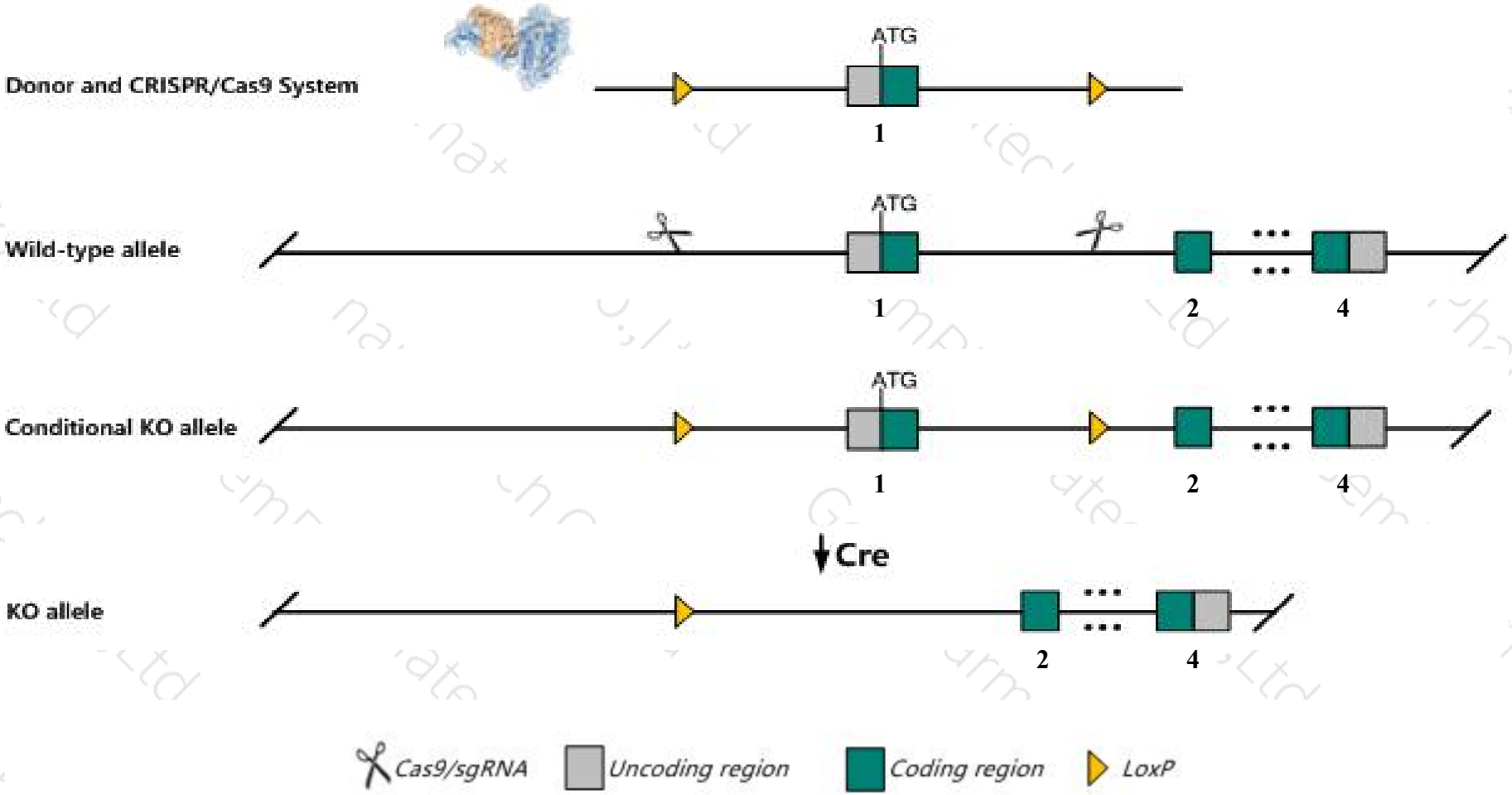
Project Overview

Project Name	***Cmtm1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cmtm1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Cmtm1* gene has 3 transcripts. According to the structure of *Cmtm1* gene, exon1 of *Cmtm1-201*(ENSMUST00000159039.1) transcript is recommended as the knockout region.The region contains start codon ATG.Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Cmtm1* 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.

Notice

➢ According to the existing MGI data,mice homozygous for a knock-out allele exhibit normal male spermatozoa and fertility.

➢ The KO region contains functional region of the *Gm45710 and Gm45711* gene.Knockout the region may affect the function of *Gm45710 and Gm45711* gene.

➢ The *Cmtm1* 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information （NCBI）

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Cmtm1 CKLF-like MARVEL transmembrane domain containing 1 [Mus musculus (house mouse)]

Gene ID: 100504164, updated on 13-Mar-2020

Summary

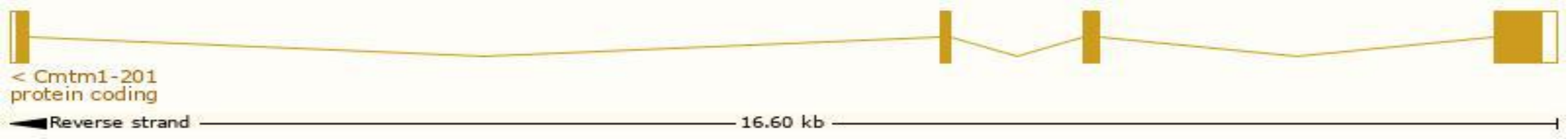
Official Symbol	Cmtm1 provided by MGI
Official Full Name	CKLF-like MARVEL transmembrane domain containing 1 provided by MGI
Primary source	MGI:MGI:2447159
See related	Ensembl:ENSMUSG00000110430
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	CHLFH1a, CKLFH, CKLFH1, Cklfsf1
Expression	Restricted expression toward testis adult (RPKM 35.0) See more
Orthologs	human all

Transcript information（Ensembl）

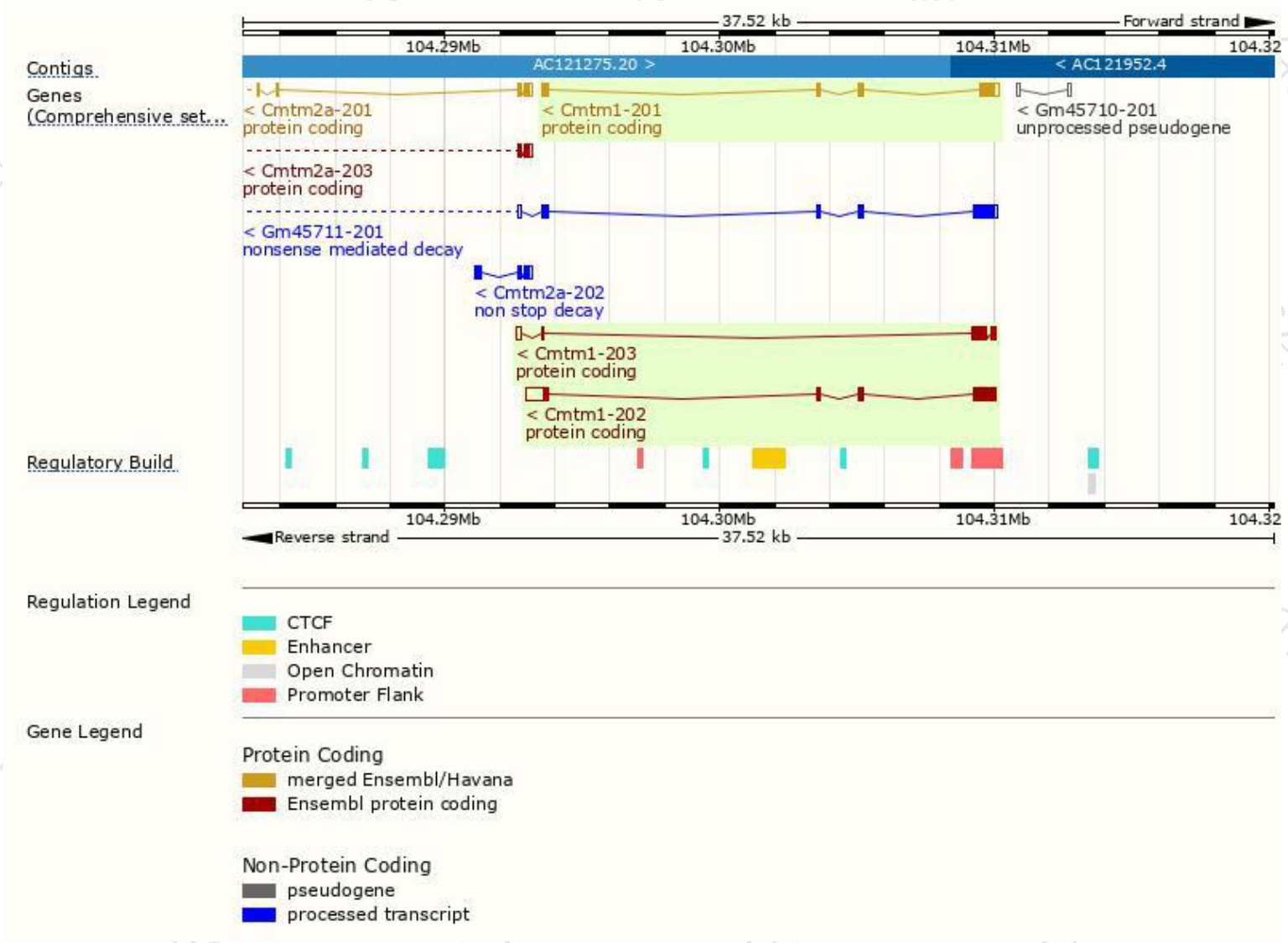
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cmtm1-201	ENSMUST00000159039.1	1130	303aa	Protein coding	CCDS57636	B7ZP21	TSL:1 GENCODE basic APPRIS P2
Cmtm1-202	ENSMUST00000160596.7	1900	391aa	Protein coding	-	E0CXA2	TSL:5 GENCODE basic APPRIS ALT2
Cmtm1-203	ENSMUST00000164175.1	979	220aa	Protein coding	-	E9PXC5	TSL:5 GENCODE basic APPRIS ALT2

The strategy is based on the design of *Cmtm1-201* transcript,the transcription is shown below:



Genomic location distribution



Protein domain

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

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