

# Cmtm3 Cas9-CKO Strategy

**Designer:** 

**Reviewer:** 

**Design Date:** 

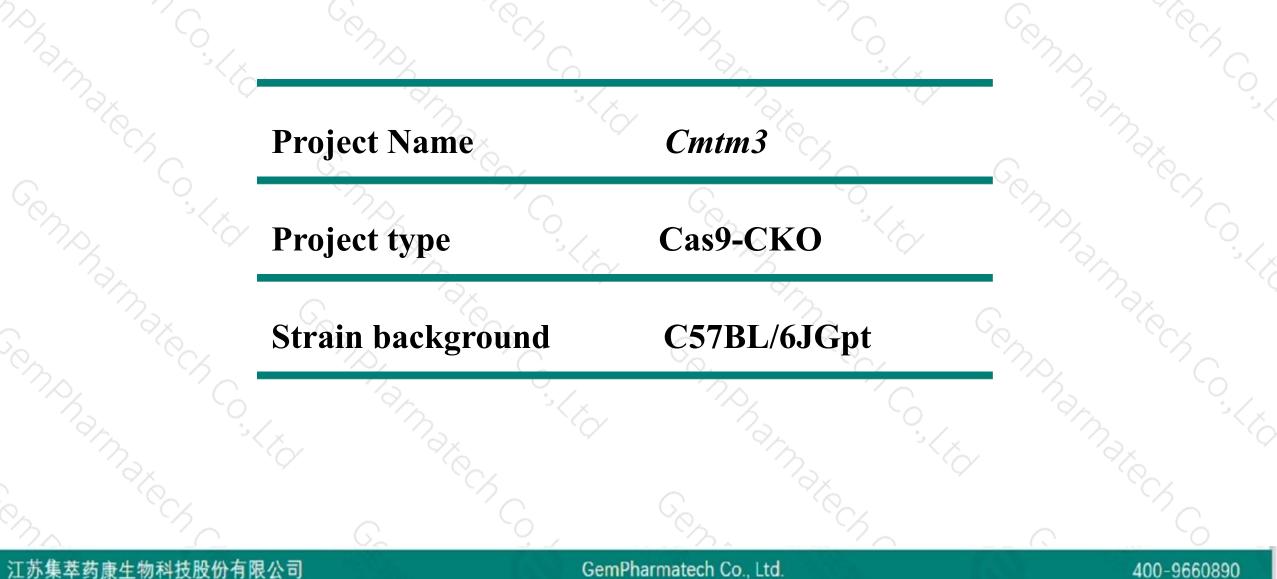
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2020-5-7

## **Project Overview**



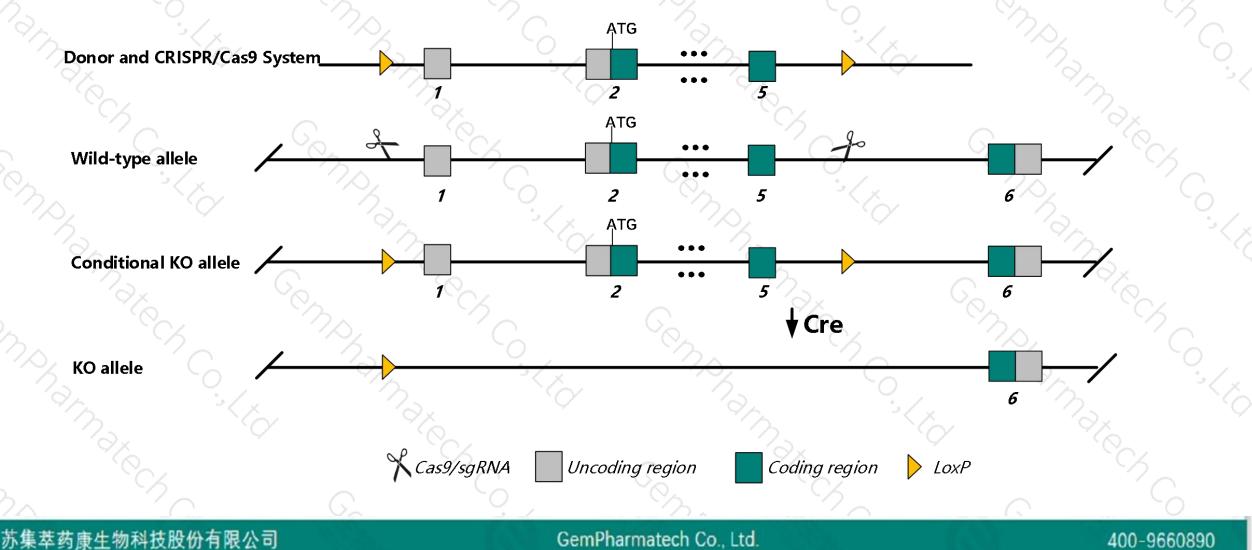


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### **Conditional Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the Cmtm3 gene. The schematic diagram is as follows:





The Cmtm3 gene has 8 transcripts. According to the structure of Cmtm3 gene, exon1-exon5 of Cmtm3-204 (ENSMUST00000212081.1) 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 *Cmtm3* 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.



- > The *Cmtm3* 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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#### Cmtm3 CKLF-like MARVEL transmembrane domain containing 3 [Mus musculus (house mouse)]

Gene ID: 68119, updated on 13-Mar-2020

#### Summary

Official Symbol	Cmtm3 provided by MGI
Official Full Name	CKLF-like MARVEL transmembrane domain containing 3 provided by MGI
Primary source	MGI:MGI:2447162
See related	Ensembl:ENSMUSG0000031875
Gene type	protein coding
<b>RefSeq status</b>	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	9430096L06Rik, AI413895, BNAS2, Cklfsf3
Expression	Broad expression in ovary adult (RPKM 59.8), adrenal adult (RPKM 43.1) and 22 other tissues See more
Orthologs	human all

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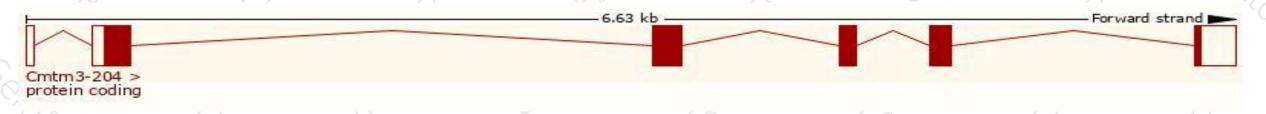
# **Transcript information (Ensembl)**



#### The gene has 8 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cmtm3-201	ENSMUST0000034343.4	1616	<u>184aa</u>	Protein coding	CCDS22577	Q99LJ5	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P
Cmtm3-204	ENSMUST00000212081.1	850	<u>184aa</u>	Protein coding	CCDS22577	<u>Q99LJ5</u>	TSL:5 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P
Cmtm3-208	ENSMUST00000212948.1	523	<u>142aa</u>	Protein coding	647	A0A1D5RM50	CDS 3' incomplete TSL:3
Cmtm3-205	ENSMUST00000212139.1	502	<u>118aa</u>	Protein coding	222	A0A1D5RLE8	CDS 3' incomplete TSL:5
Cmtm3-202	ENSMUST00000211885.1	465	<u>82aa</u>	Protein coding	1271	A0A1D5RLU9	CDS 3' incomplete TSL:3
Cmtm3-207	ENSMUST00000212734.1	867	No protein	Retained intron	10.		TSL:2
Cmtm3-206	ENSMUST00000212399.1	790	No protein	Retained intron	640	-	TSL:2
Cmtm3-203	ENSMUST00000211996.1	630	No protein	Retained intron	1224	2	TSL:3
		1	C		1 1	- A-	

The strategy is based on the design of *Cmtm3-204* transcript, the transcription is shown below:



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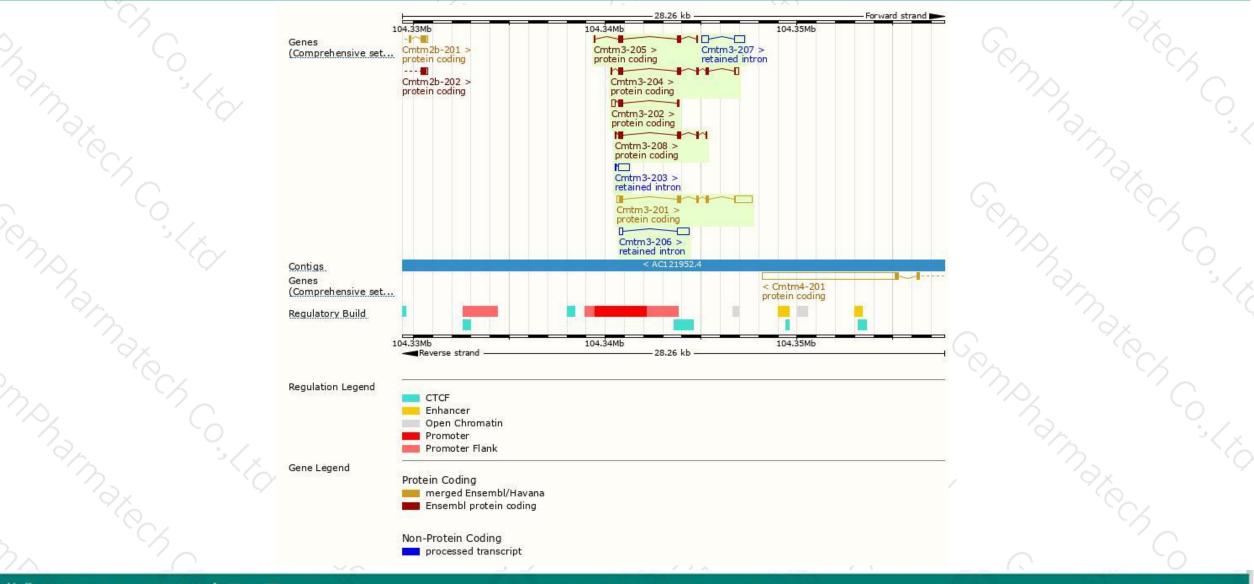
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### **Genomic location distribution**



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### **Protein domain**







If you have any questions, you are welcome to inquire. Tel: 400-9660890



