

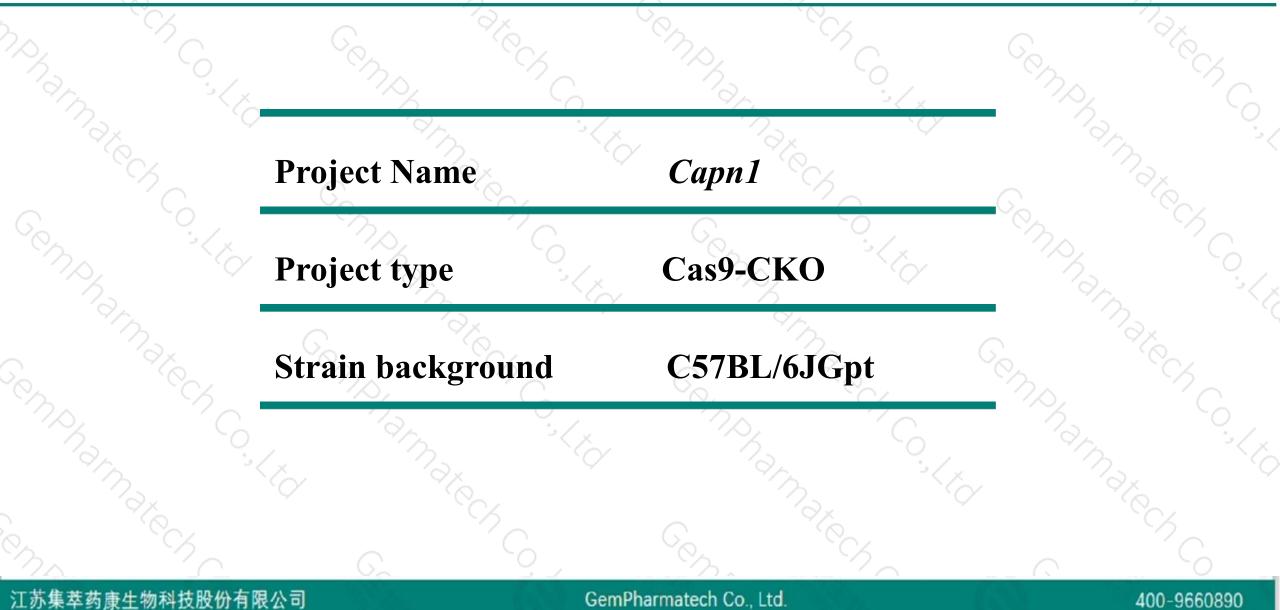
# Capn1 Cas9-CKO Strategy Annak Cherry

Cemphamatech, Cemphamatech, Designer:Lixin LYU Design Date:2019-8-12

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# **Project Overview**



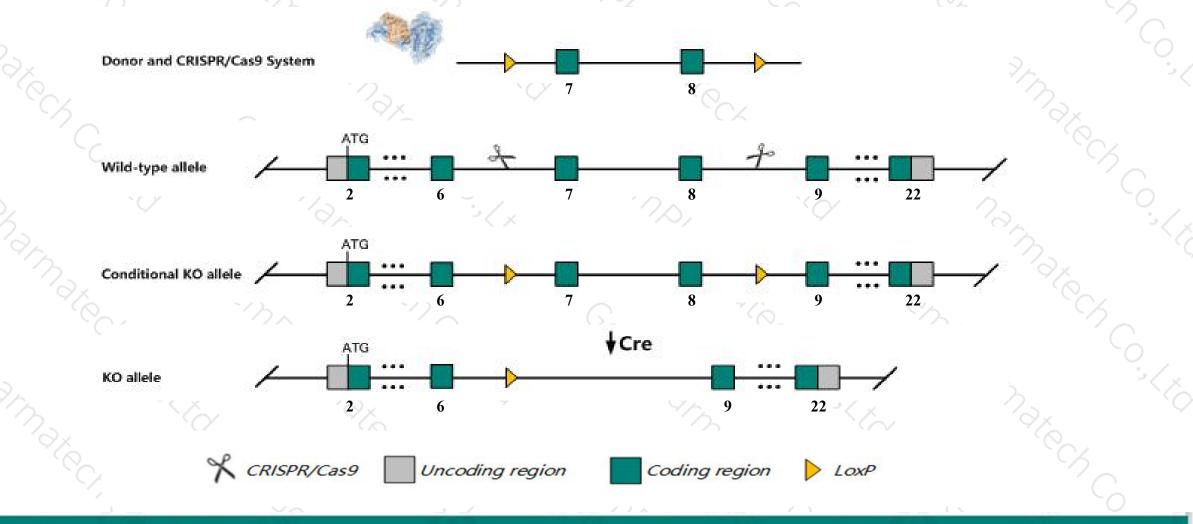


# **Conditional Knockout strategy**



400-9660890

This model will use CRISPR/Cas9 technology to edit the Capn1 gene. The schematic diagram is as follows:



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The Capn1 gene has 6 transcripts. According to the structure of Capn1 gene, exon7-exon8 of Capn1-201 (ENSMUST00000025891.9) transcript is recommended as the knockout region. The region contains 170bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Capn1* 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.

## Notice



- According to the existing MGI data, Animals homozygous for a mutation of this gene exhibit decreased platelet aggregation and defective clot retraction although bleeding times remain similar to wild-type.
- > The influence of *Capn1*-203&204&206 is unknown.

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The N-terminal of *Capn1* gene will remain 281aa,it may remain the partial function of *Capn1* gene.
The *Capn1* gene is located on the Chr19. 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.

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# Gene information (NCBI)



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#### Capn1 calpain 1 [Mus musculus (house mouse)]

Gene ID: 12333, updated on 31-Jan-2019

#### Summary

Official Symbol	Const
Official Symbol	Capn1 provided by MGI
Official Full Name	calpain 1 provided by MGI
Primary source	MGI:MGI:88263
See related	Ensembl:ENSMUSG00000024942
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	Capa-1, Capa1, mu-calpin
Expression	Ubiquitous expression in large intestine adult (RPKM 48.9), bladder adult (RPKM 44.0) and 24 other tissues See more
Orthologs	human all

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# **Transcript information (Ensembl)**



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

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Capn1-201	ENSMUST00000025891.9	3060	<u>713aa</u>	Protein coding	CCDS37893	<u>035350 Q3T107</u>	TSL:1 GENCODE basic APPRIS P1
Capn1-202	ENSMUST00000164843.7	3048	<u>713aa</u>	Protein coding	CCDS37893	<u>035350 Q3T107</u>	TSL:5 GENCODE basic APPRIS P1
Capn1-205	ENSMUST00000236798.1	2260	<u>699aa</u>	Protein coding	-	-	CDS 3' incomplete
Capn1-206	ENSMUST00000237519.1	679	<u>196aa</u>	Protein coding	-	2	CDS 3' incomplete
Capn1-204	ENSMUST00000236537.1	409	<u>79aa</u>	Protein coding	5		CDS 3' incomplete
Capn1-203	ENSMUST00000235138.1	326	<u>79aa</u>	Protein coding	-	-8	CDS 3' incomplete

The strategy is based on the design of Capn1-201 transcript, The transcription is shown below

< Capn1-201 protein coding

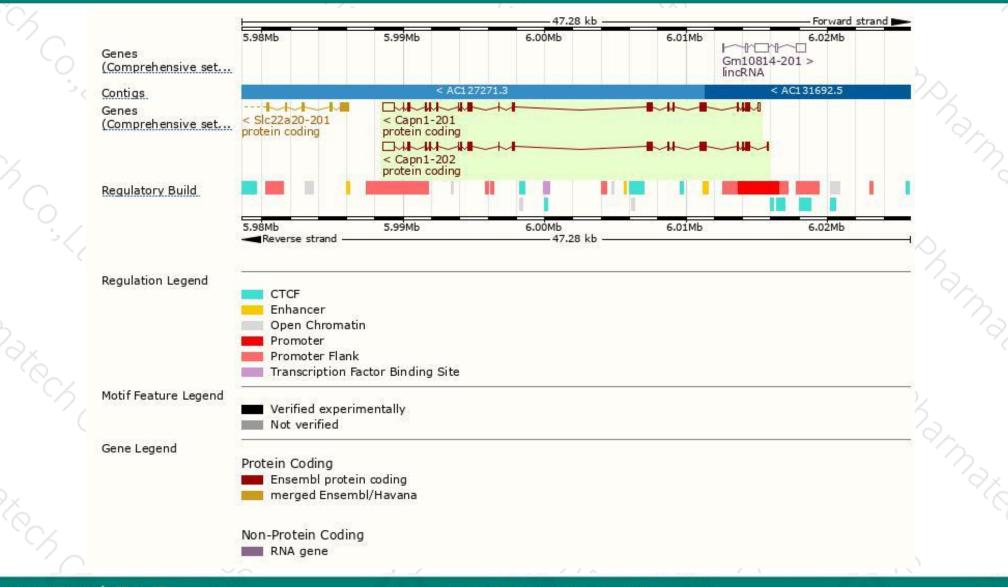
Reverse strand

\_\_\_\_\_ 26.66 kb

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## **Genomic location distribution**



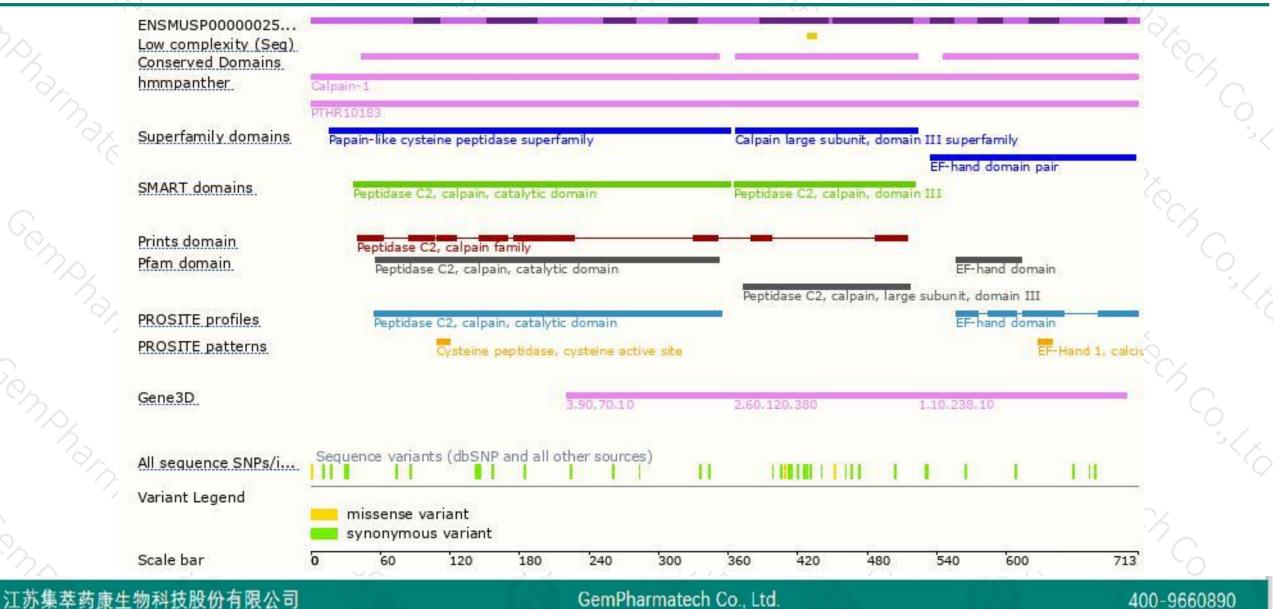


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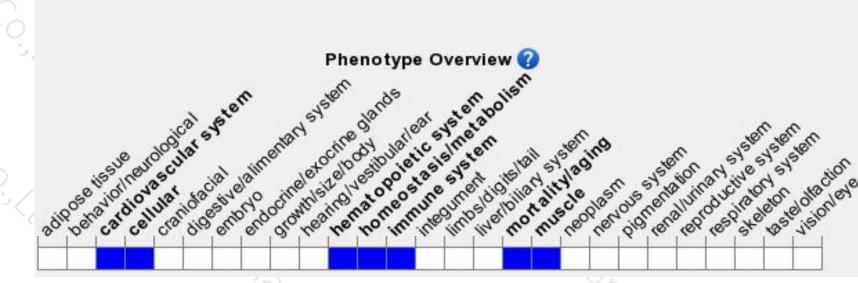
## **Protein domain**





# 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, Animals homozygous for a mutation of this gene exhibit decreased platelet aggregation and defective clot retraction although bleeding times remain similar to wild-type.

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



