

# Capn7 Cas9-CKO Strategy

Designer: Yun Li

**Reviewer: Shuang Zhang** 

**Design Date: 2021-7-15** 

## **Project Overview**

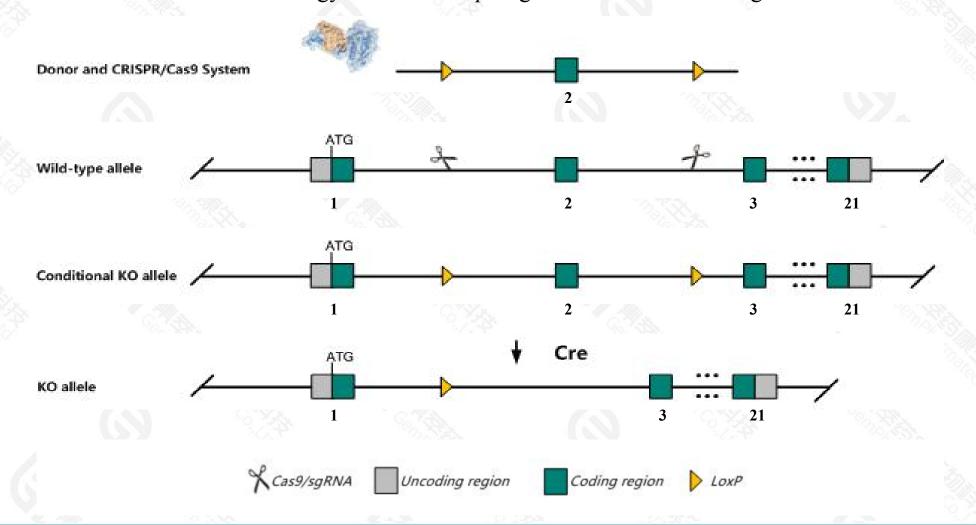


Project Name	Capn 7		
Project type	Cas9-CKO		
Strain background	C57BL/6JGpt		

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Capn7* gene. The schematic diagram is as follows:



### **Technical routes**



- > The *Capn7* gene has 4 transcripts. According to the structure of *Capn7* gene, exon2 of *Capn7*201(ENSMUST00000022451.14) transcript is recommended as the knockout region. The region contains 109bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Capn7* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor 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 disruptions in this gene frequently die before weaning. Survivors display reduced body weight.
- > The Capn7 gene is located on the Chr14. 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)



#### Capn7 calpain 7 [Mus musculus (house mouse)]

Gene ID: 12339, updated on 17-Dec-2020

#### Summary

☆ ?

Official Symbol Capn7 provided by MGI
Official Full Name calpain 7 provided by MGI

Primary source MGI:MGI:1338030

See related Ensembl: ENSMUSG00000021893

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 AU022319, Pa, PalBH

Expression Ubiquitous expression in CNS E18 (RPKM 22.2), CNS E14 (RPKM 19.2) and 28 other tissuesSee more

Orthologs <u>human all</u>

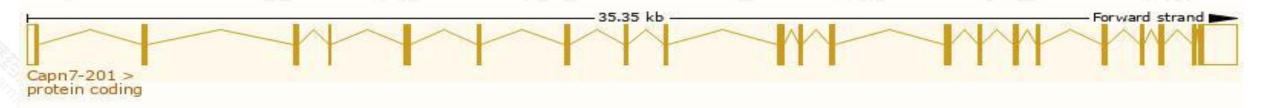
## Transcript information (Ensembl)



The gene has 4 transcripts, all transcripts are shown below:

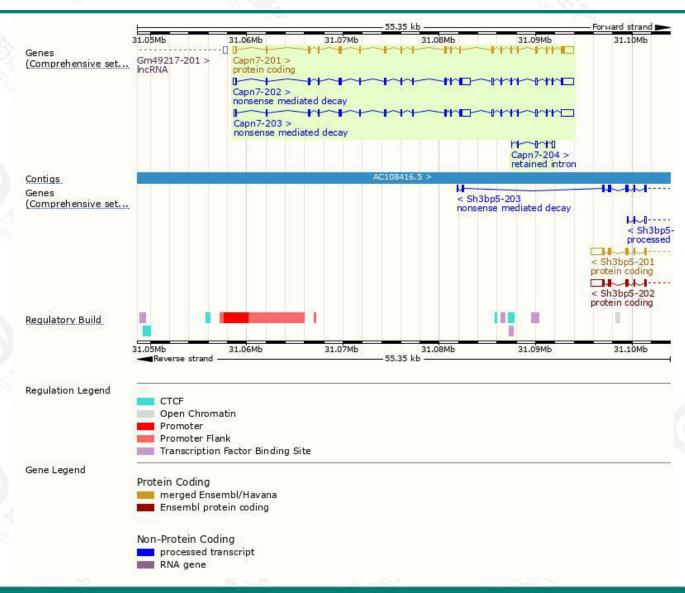
Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags	
ENSMUST00000022451.14	3648	<u>813aa</u>	Protein coding	CCDS26911		TSL:1, GENCODE basic, APPRIS P1	
ENSMUST00000152182.2	4662	<u>508aa</u>	Nonsense mediated decay			TSL:1,	
ENSMUST00000143472.8	4613	<u>508aa</u>	Nonsense mediated decay	2		TSL:5,	
ENSMUST00000228237.2	614	No protein	Retained intron	-			
	ENSMUST000000152182.2 ENSMUST00000143472.8	ENSMUST00000022451.14 3648 ENSMUST00000152182.2 4662 ENSMUST00000143472.8 4613	ENSMUST00000022451.14 3648 813aa  ENSMUST00000152182.2 4662 508aa  ENSMUST00000143472.8 4613 508aa	ENSMUST00000022451.14         3648         813aa         Protein coding           ENSMUST00000152182.2         4662         508aa         Nonsense mediated decay           ENSMUST00000143472.8         4613         508aa         Nonsense mediated decay	ENSMUST00000022451.14         3648         813aa         Protein coding         CCDS26911           ENSMUST00000152182.2         4662         508aa         Nonsense mediated decay         -           ENSMUST00000143472.8         4613         508aa         Nonsense mediated decay         -	ENSMUST00000022451.14         3648         813aa         Protein coding         CCDS26911           ENSMUST00000152182.2         4662         508aa         Nonsense mediated decay         -           ENSMUST00000143472.8         4613         508aa         Nonsense mediated decay         -	

The strategy is based on the design of *Capn7-201* transcript, the transcription is shown below:



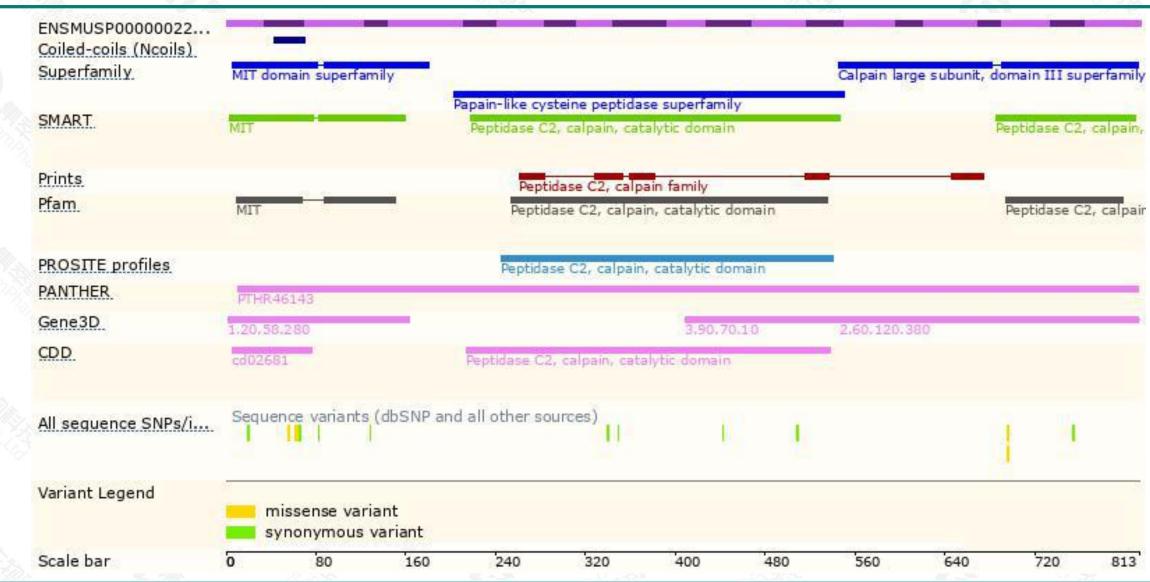
### Genomic location distribution





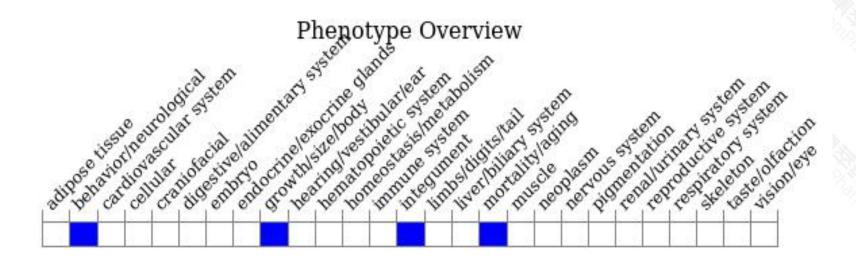
### Protein domain





## Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

According to the existing MGI data,mice homozygous for disruptions in this gene frequently die before weaning. Survivors display reduced body weight.



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

Tel: 025-5864 1534





