

Capn2 Cas9-CKO Strategy

Designer:

Bingxuan Li

Design Date:

2019-9-5

Project Overview

Project Name

Capn2

Project type

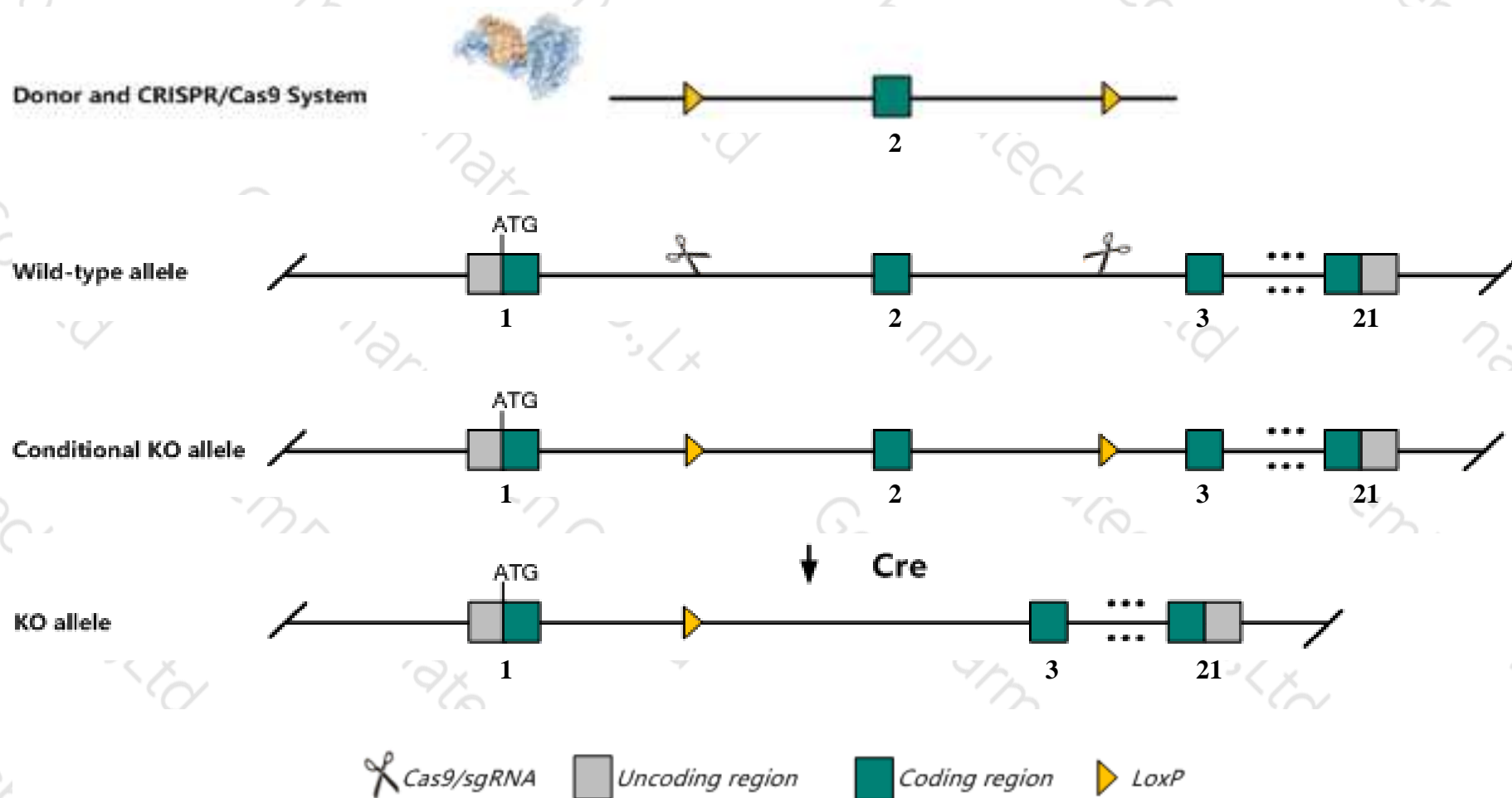
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Capn2* gene. The schematic diagram is as follows:



Technical routes

- The *Capn2* gene has 5 transcripts. According to the structure of *Capn2* gene, exon2 of *Capn2-201* (ENSMUST00000068505.9) transcript is recommended as the knockout region. The region contains 70bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Capn2* 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.

- According to the existing MGI data, Homozygous inactivation of this gene leads to complete prenatal lethality. Mice homozygous for one null allele display placental dysfunction, thin ventricular walls, and peripheral vessel failure.
- The *Capn2* gene is located on the Chr1. 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)

Capn2 calpain 2 [*Mus musculus* (house mouse)]









Gene ID: 12334, updated on 12-Aug-2019

Summary

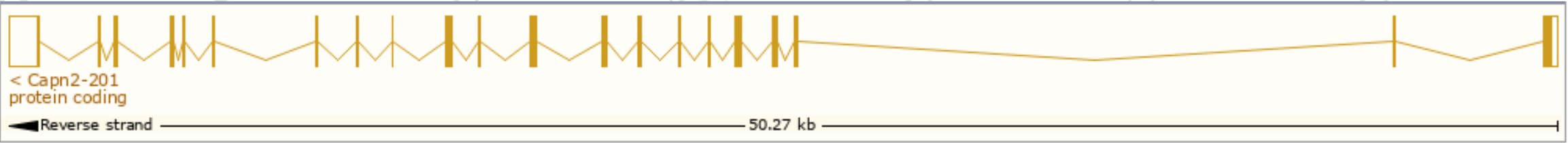
Official Symbol	Capn2 provided by MGI
Official Full Name	calpain 2 provided by MGI
Primary source	MGI:MGI:88264
See related	Ensembl:ENSMUSG000000026509
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Capa2; CALP80; Capa-2; A1326419; m-calpin; m-calpain
Expression	Ubiquitous expression in bladder adult (RPKM 35.3), subcutaneous fat pad adult (RPKM 29.4) and 27 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

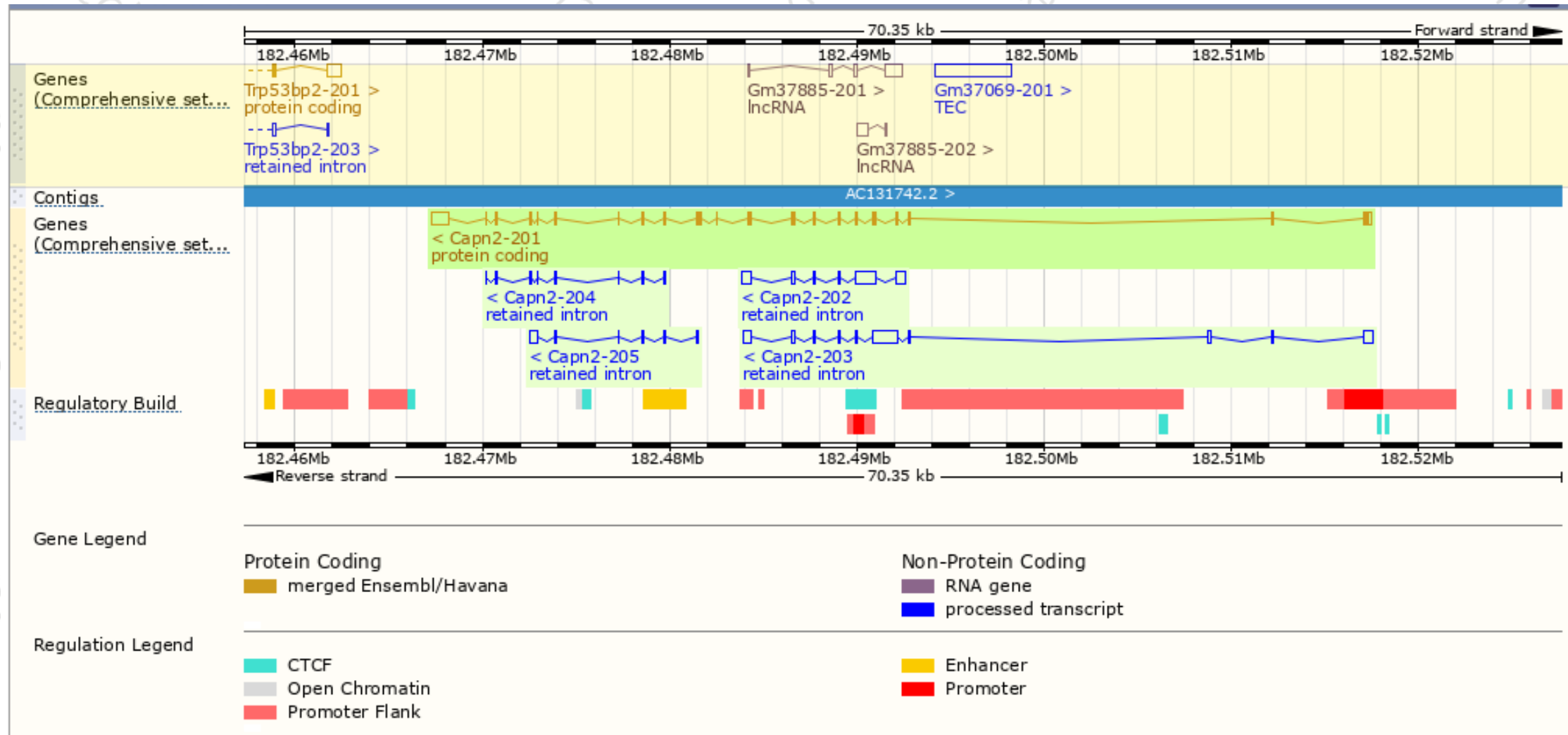
The gene has 5 transcripts,all transcripts are shown below:

Show/hide columns (1 hidden)							Filter			
Name ▾	Transcript ID ▲	bp ▾	Protein ▾	Biotype ▾	CCDS ▾	UniProt ▾	Flags ▾			
Capn2-201	ENSMUST00000068505.9	3225	700aa	 Protein coding	CCDS35813 	O08529 	TSL:1	GENCODE basic	APPRIS P1	
Capn2-202	ENSMUST00000192230.5	2366	No protein	 Retained intron	-	-	TSL:1			
Capn2-203	ENSMUST00000192483.1	3017	No protein	 Retained intron	-	-	TSL:1			
Capn2-204	ENSMUST00000194940.5	580	No protein	 Retained intron	-	-	TSL:3			
Capn2-205	ENSMUST00000194961.1	728	No protein	 Retained intron	-	-	TSL:1			

The strategy is based on the design of *Capn2-201* transcript,The transcription is shown below



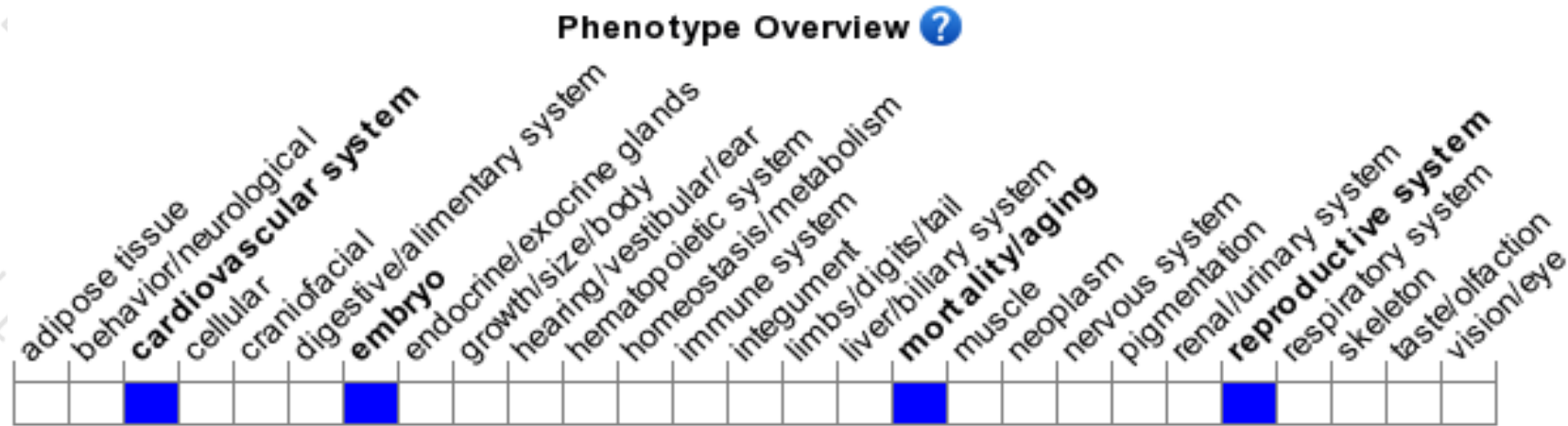
Genomic location distribution



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, Homozygous inactivation of this gene leads to complete prenatal lethality. Mice homozygous for one null allele display placental dysfunction, thin ventricular walls, and peripheral vessel failure.

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

Tel: 025-5864 1534

