Capn2 Cas9-KO *Strategy*

Designer: **Bingxuan Li**

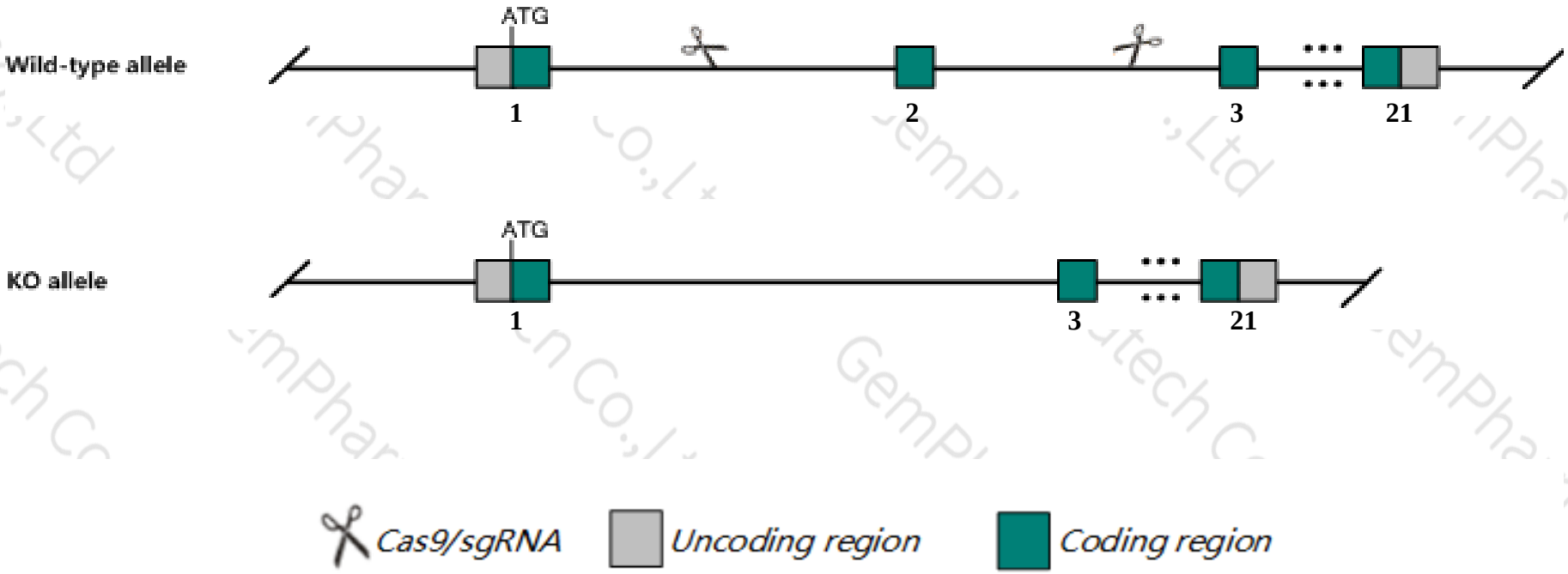
Design Date: **2019-9-5**

Project Overview

Project Name	***Capn2***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

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.Cas9 and sgRNA 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.

Notice

- 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information （NCBI）

集萃药康 GemPharmatech

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:ENSMUSG00000026509
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; AI326419; 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

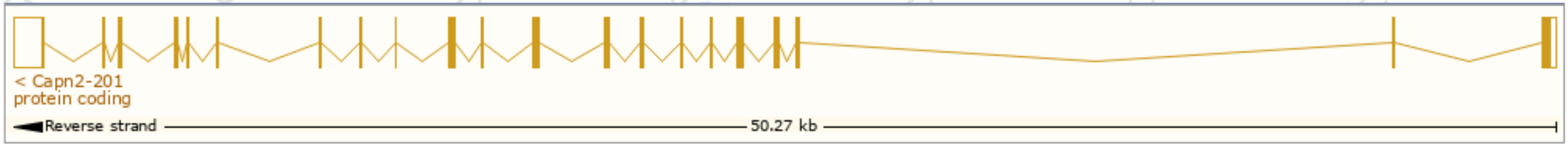
Transcript information（Ensembl）

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

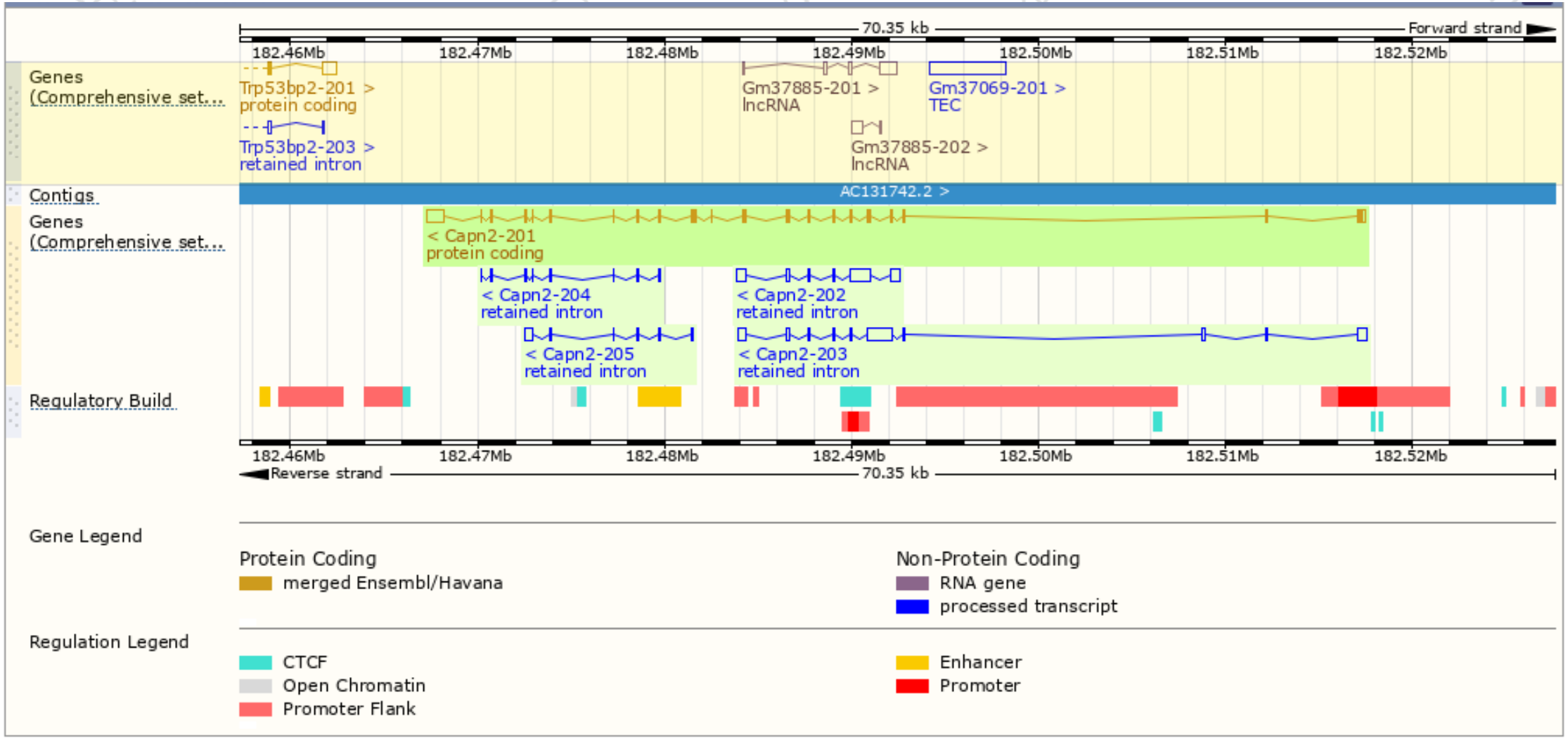
Show/hide columns (1 hidden) Filter

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

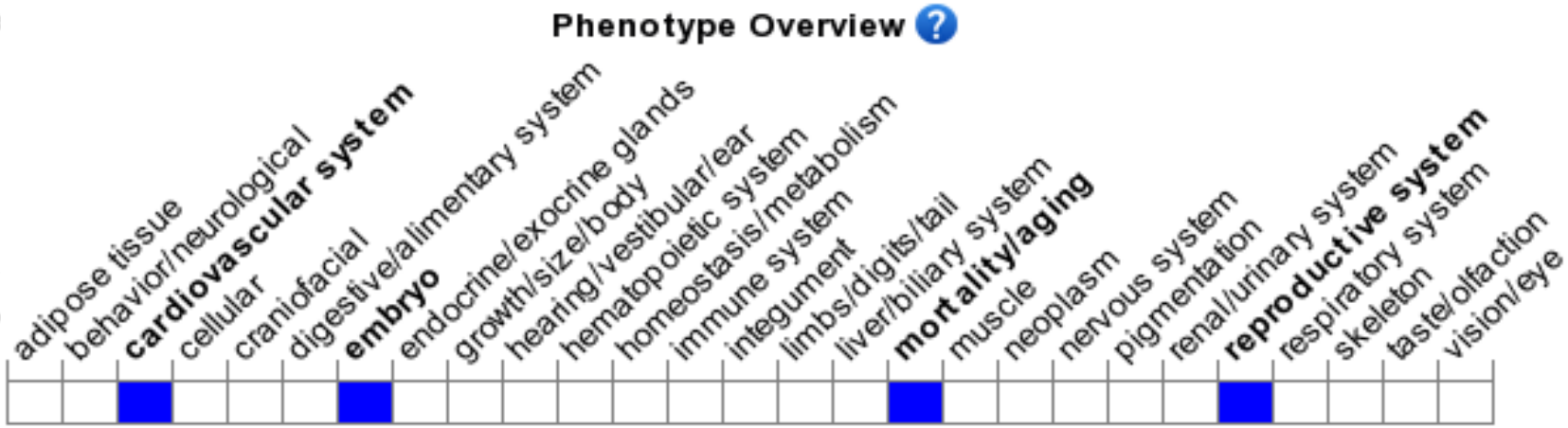


Genomic location distribution



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/).

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

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