Ppp4c Cas9-KO Strategy

Designer: Rui Xiong

Reviewer: Lingyan Wu

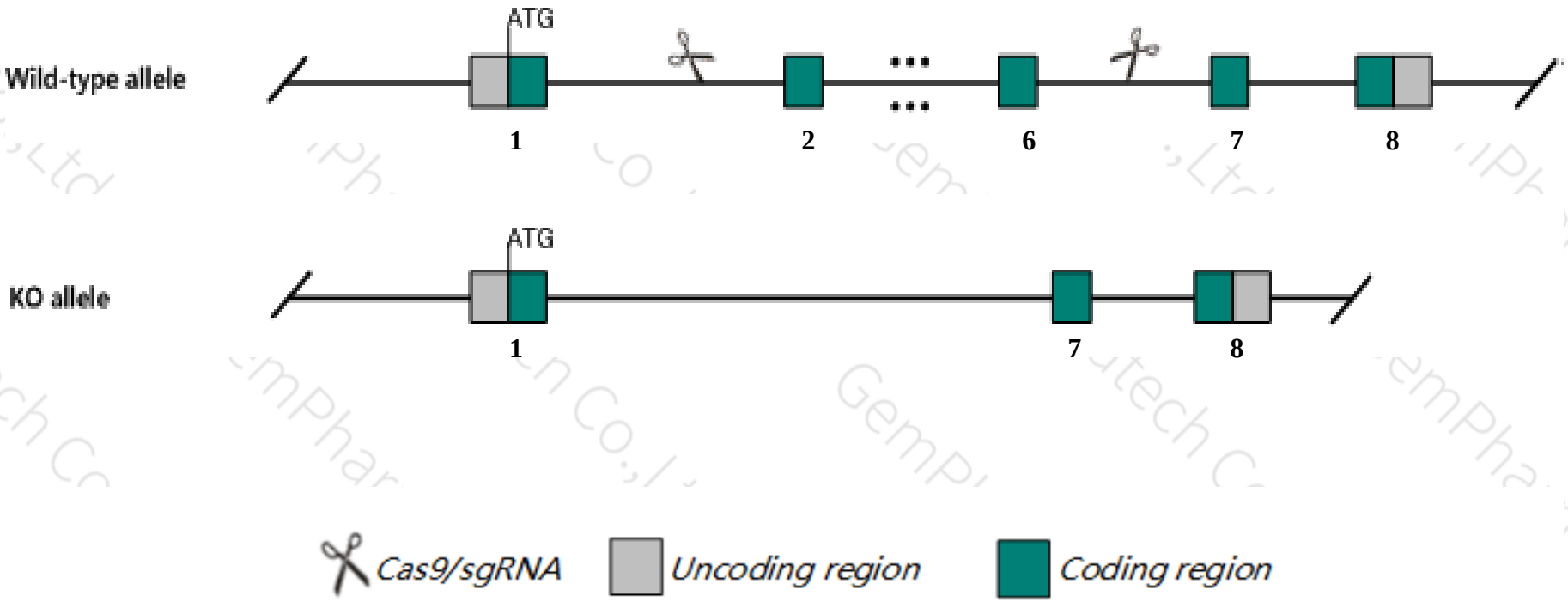
Design Date: 2021-1-8

Project Overview

Project Name	***Ppp4c***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ppp4c* gene. The schematic diagram is as follows:



Technical routes

➢ The *Ppp4c* gene has 11 transcripts. According to the structure of *Ppp4c* gene, exon2-exon6 of *Ppp4c-211*(ENSMUST00000206570.1) transcript is recommended as the knockout region.The region contains 506bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Ppp4c* 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,embryonic Ppp4c-deficiency leads to early embryonic lethality.

➢ The *Ppp4c* gene is located on the Chr7. 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

Ppp4c protein phosphatase 4, catalytic subunit [Mus musculus (house mouse)]

Gene ID: 56420, updated on 13-Mar-2020

Summary

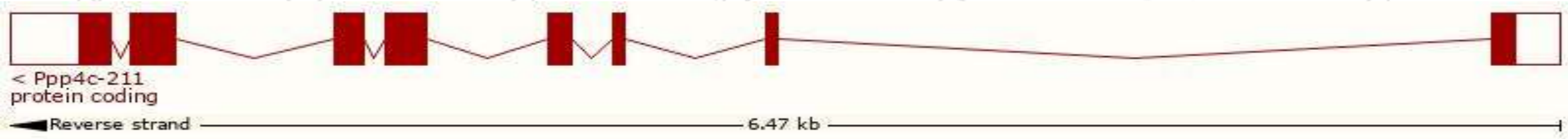
Official Symbol	Ppp4c provided by MGI
Official Full Name	protein phosphatase 4, catalytic subunit provided byMGI
Primary source	MGI:MGI:1891763
See related	Ensembl:ENSMUSG00000030697
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1110002D08Rik, AU016079, PP-X, PP4C, Ppx, pp4
Expression	Ubiquitous expression in thymus adult (RPKM 94.8), CNS E11.5 (RPKM 74.7) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

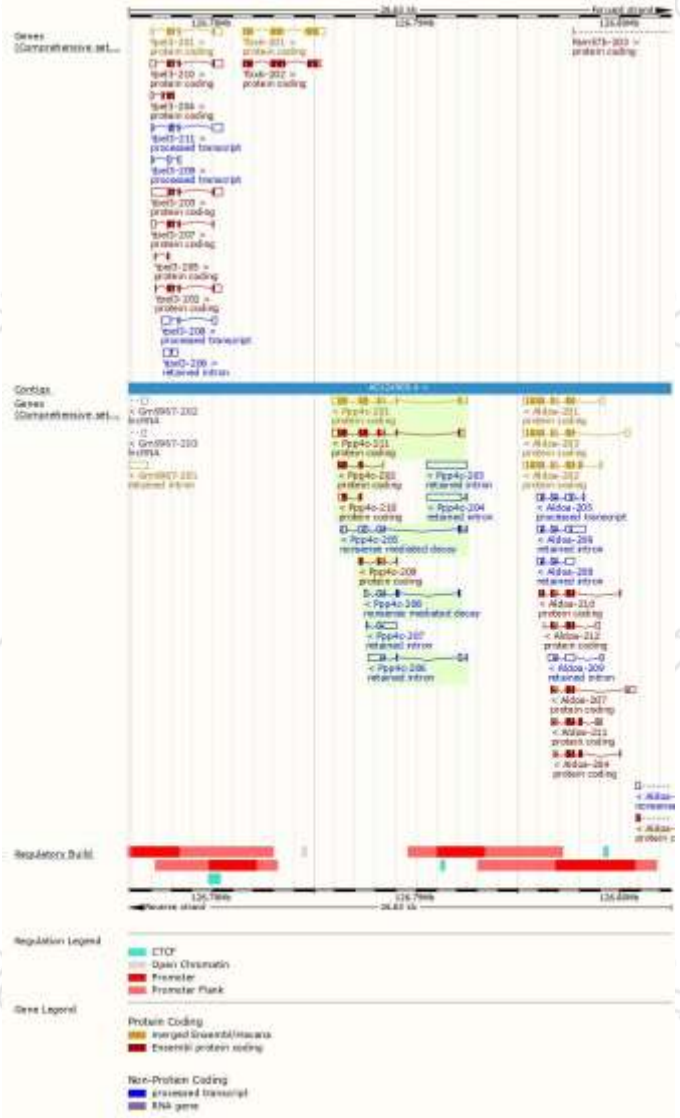
The gene has 11 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ppp4c-211	ENSMUST00000206570.1	1400	307aa	Protein coding	CCDS21844	P97470 Q3UNF2	TSL:1 GENCODE basic APPRIS P1
Ppp4c-201	ENSMUST00000032936.7	1359	307aa	Protein coding	CCDS21844	P97470 Q3UNF2	TSL:1 GENCODE basic APPRIS P1
Ppp4c-202	ENSMUST00000205786.1	477	157aa	Protein coding	-	A0A0U1RQ12	CDS 5' incomplete TSL:3
Ppp4c-210	ENSMUST00000206477.1	453	104aa	Protein coding	-	A0A0U1RPK2	CDS 5' incomplete TSL:3
Ppp4c-209	ENSMUST00000206353.1	301	100aa	Protein coding	-	A0A0U1RPR5	CDS 5' and 3' incomplete TSL:5
Ppp4c-205	ENSMUST00000205935.1	881	50aa	Nonsense mediated decay	-	A0A0U1RQ43	TSL:5
Ppp4c-208	ENSMUST00000206334.1	494	53aa	Nonsense mediated decay	-	A0A0U1RPZ1	CDS 5' incomplete TSL:3
Ppp4c-203	ENSMUST00000205862.1	1984	No protein	Retained intron	-	-	TSL:NA
Ppp4c-204	ENSMUST00000205879.1	1747	No protein	Retained intron	-	-	TSL:1
Ppp4c-206	ENSMUST00000205950.1	865	No protein	Retained intron	-	-	TSL:2
Ppp4c-207	ENSMUST00000206261.1	776	No protein	Retained intron	-	-	TSL:3

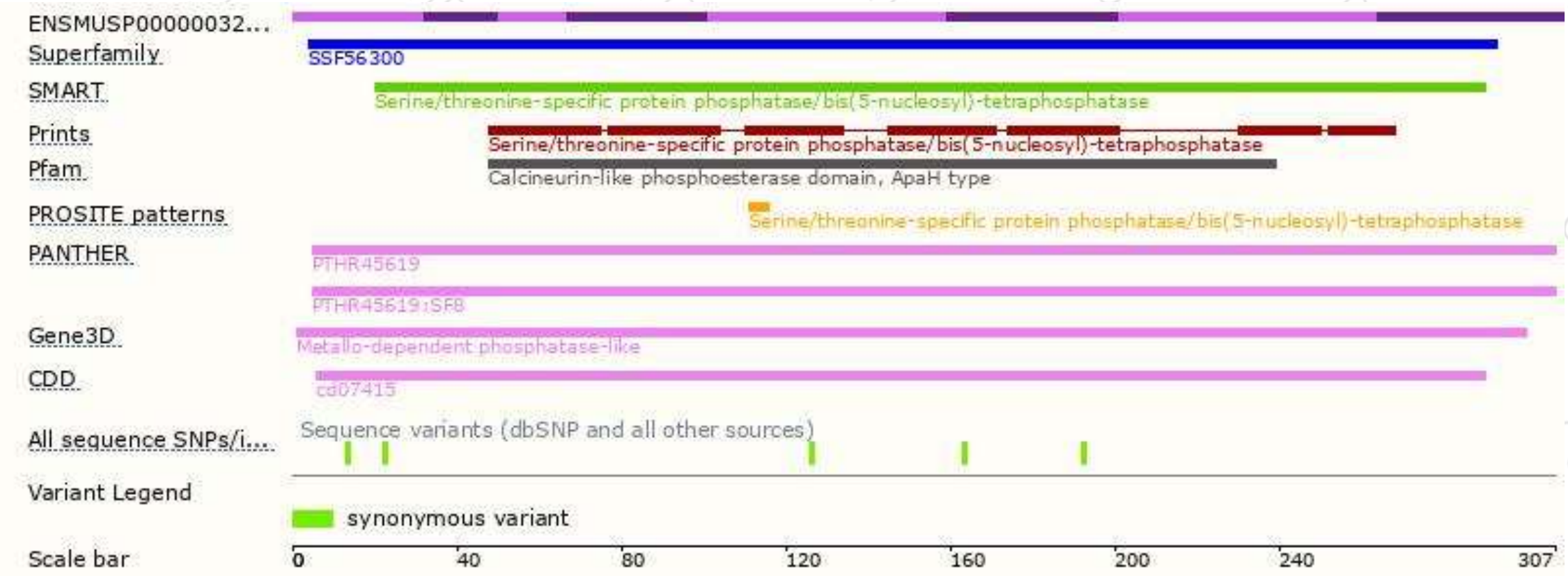
The strategy is based on the design of *Ppp4c-211* transcript,the transcription is shown below:



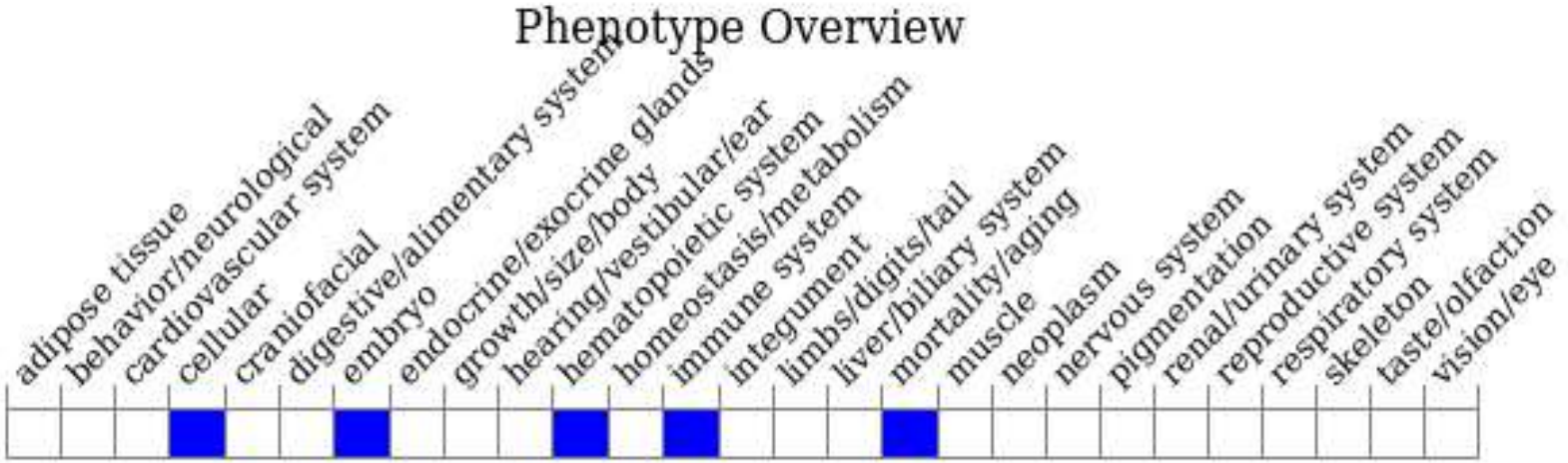
Genomic location distribution



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,embryonic Ppp4c-deficiency leads to early embryonic lethality.

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

Tel: 025-5864 1534

集萃药康生物科技
GemPharmatech Co.,Ltd