

Ppp2r5c Cas9-KO Strategy

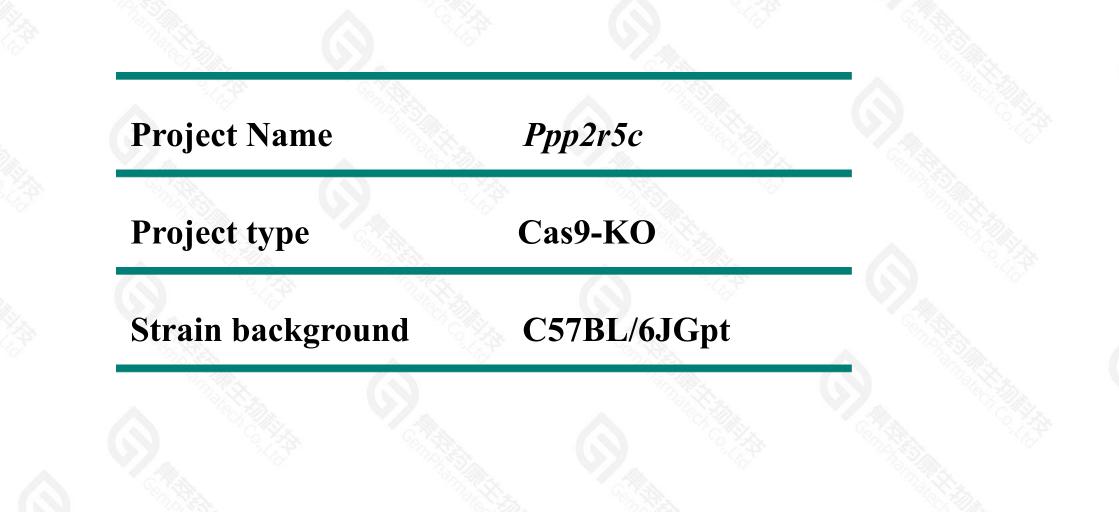
Designer: Longyun Hu

Reviewer: Rui Xiong

Design Date: 2021-3-12

Project Overview





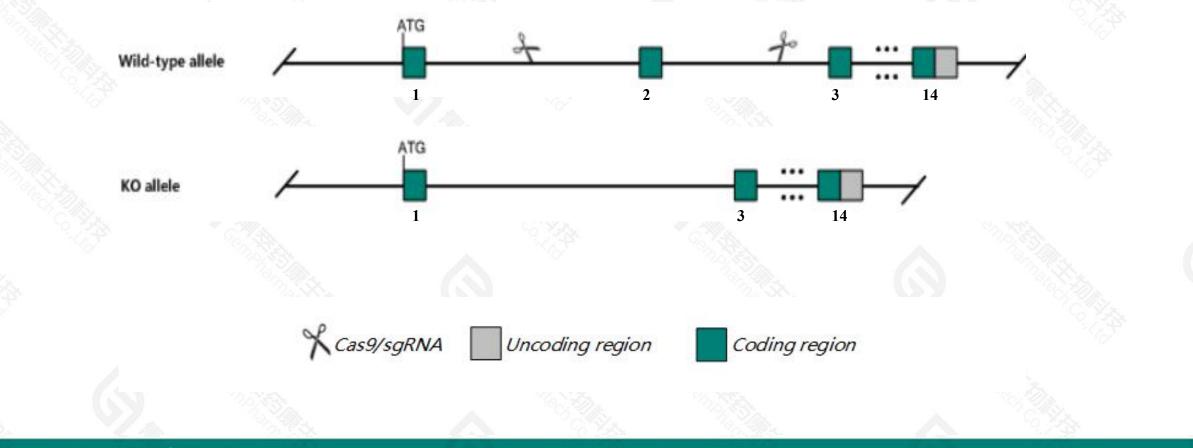
江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Ppp2r5c* gene. The schematic diagram is as follows:



江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.



> The *Ppp2r5c* gene has 7 transcripts. According to the structure of *Ppp2r5c* gene, exon2 of *Ppp2r5c*-201(ENSMUST00000084985.10) transcript is recommended as the knockout region. The region contains 200bp coding sequence. Knock out the region will result in disruption of protein function.

> In this project we use CRISPR/Cas9 technology to modify Ppp2r5c 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.



According to the existing MGI data, mice homozygous for a gene-trapped allele show partial neonatal lethality, hypoactivity, and abnormal ventricular septum formation associated with increased fetal cardiomyocyte apoptosis.
Surviving homozygotes develop obesity and show an abnormal gait, decreased grip strength, and impaired balance.
Transcript *Ppp2r5c-206* may not be affected.

- > The Ppp2r5c gene is located on the Chr12. 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)



☆ ?

Ppp2r5c protein phosphatase 2, regulatory subunit B', gamma [Mus musculus (house mouse)]

Gene ID: 26931, updated on 13-Mar-2020

Summary

Official Symbol	Ppp2r5c provided by MGI
Official Full Name	protein phosphatase 2, regulatory subunit B', gamma provided by MGI
Primary source	MGI:MGI:1349475
See related	Ensembl:ENSMUSG0000017843
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	2610043M05Rik, 2700063L20Rik, Al060890, AW545884, C85228, D12Bwg0916e
Expression	Ubiquitous expression in testis adult (RPKM 41.8), cortex adult (RPKM 18.2) and 27 other tissuesSee more
Orthologs	human all

江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

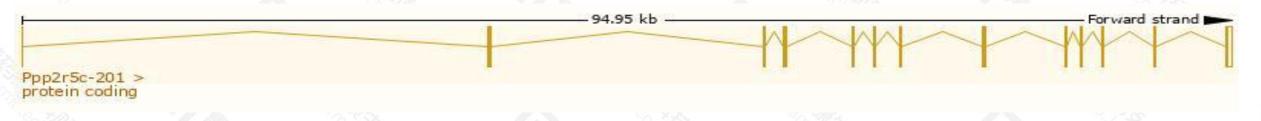
Transcript information (Ensembl)



The gene has 7 transcripts, all transcripts are shown below:

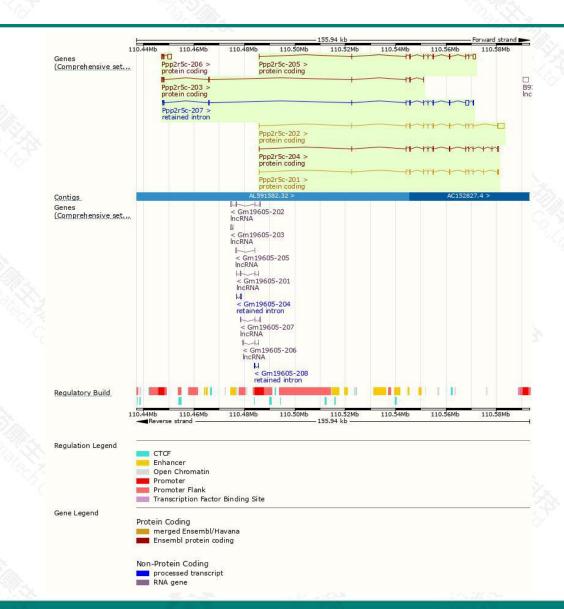
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags		
Ppp2r5c-202	ENSMUST00000109832.2	4108	<u>485aa</u>	Protein coding	CCDS36558	<u>Q60996</u>	TSL:2 GENCODE basic APPRIS ALT1		
Ppp2r5c-201	ENSMUST0000084985.10	1847	<u>524aa</u>	Protein coding	CCDS36557	<u>Q60996</u>	TSL:2 GENCODE basic APPRIS P4		
Ppp2r5c-205	ENSMUST00000221715.1	2367	<u>452aa</u>	Protein coding		<u>Q60996</u>	TSL:1 GENCODE basic		
Ppp2r5c-204	ENSMUST00000221074.1	1971	<u>496aa</u>	Protein coding	8 . -9	<u>Q60996</u>	TSL:5 GENCODE basic		
Ppp2r5c-206	ENSMUST00000222276.1	1802	<u>49aa</u>	Protein coding	8 4 8	A0A1Y7VJC8	TSL:2 GENCODE basic		
Ppp2r5c-203	ENSMUST00000220509.1	956	<u>283aa</u>	Protein coding	(E)	A0A1Y7VIR0	CDS 3' incomplete TSL:5		
Ppp2r5c-207	ENSMUST00000223168.1	2790	No protein	Retained intron	-	-	TSL:2		

The strategy is based on the design of *Ppp2r5c-201* transcript, the transcription is shown below:



Genomic location distribution





江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

Protein domain

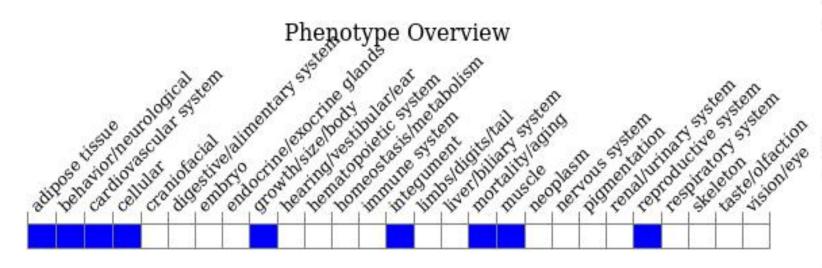


ENSMUSP00000082 MobiDB lite Low complexity (Seg) Superfamily	Armadi	lo-type fold		-			-	
Pfam	Protein ph	osphatase 2A, regu	latory B subunit, B	16				
PIRSF	Protein phosp	hatase 2A, regulato	ry Bisubunit, 856					
PANTHER	Protein phospha	itase 2A, regulatory	8 subunit, 856					
	PTHR10257:SF4	19						
Gene3D	Armadillo	r-like halical						
All sequence SNPs/i	Sequence varia	ints (dbSNP and a	Il other sources)		1 0	10 SE	1	33
Variant Legend		on variant us variant						
Scale bar	6 60	120	180	240	300	360	420	524

江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

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 a gene-trapped allele show partial neonatal lethality, hypoactivity, and abnormal ventricular septum formation associated with increased fetal cardiomyocyte apoptosis. Surviving homozygotes develop obesity and show an abnormal gait, decreased grip strength, and impaired balance.

江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.



If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



