

Ppp2r5b Cas9-CKO Strategy

Designer:

Reviewer:

Design Date:

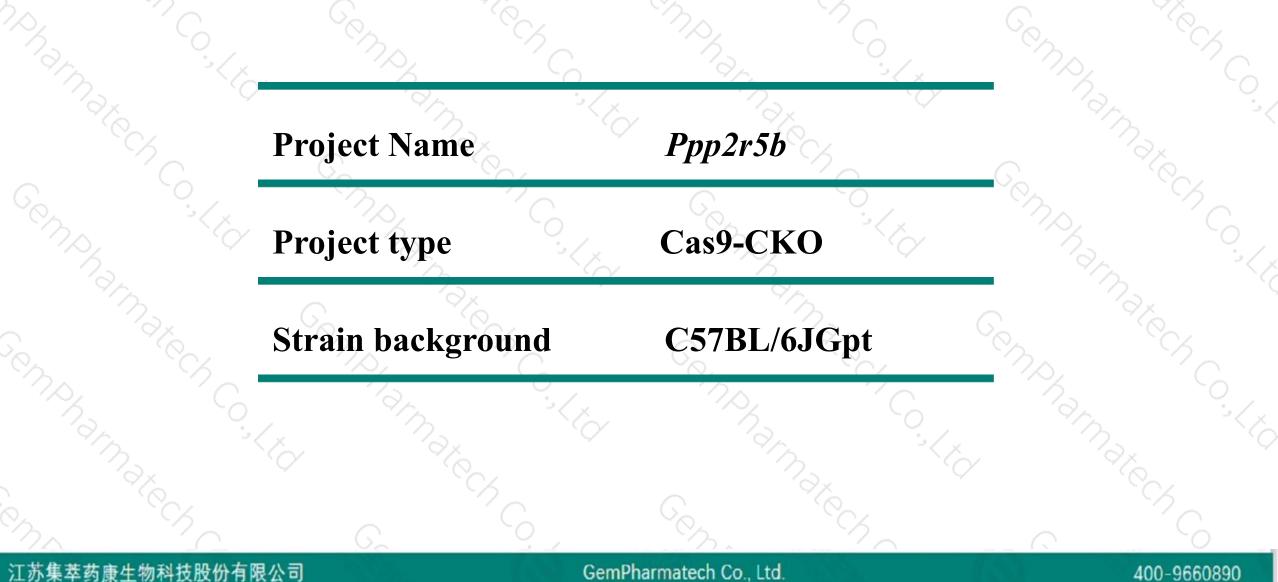
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2020-4-20

Project Overview





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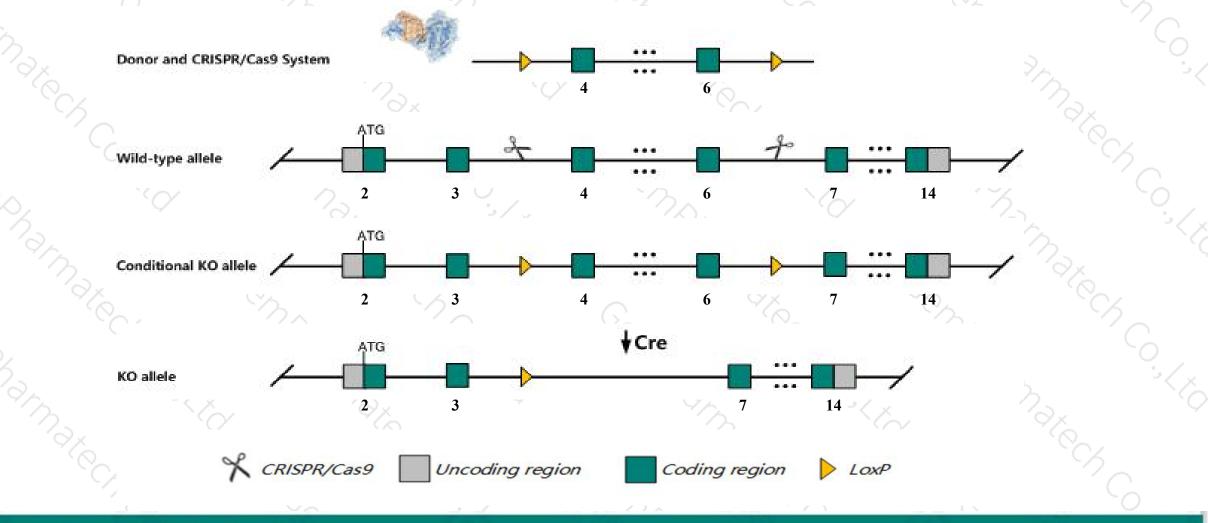
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Conditional Knockout strategy



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This model will use CRISPR/Cas9 technology to edit the *Ppp2r5b* gene. The schematic diagram is as follows:



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The Ppp2r5b gene has 4 transcripts. According to the structure of Ppp2r5b gene, exon4-exon6 of Ppp2r5b-201 (ENSMUST00000025695.9) transcript is recommended as the knockout region. The region contains 326bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Ppp2r5b* gene. The brief process is as follows:CRISPR/Cas9 system 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 will be 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.



- > The *Ppp2r5b* gene is located on the Chr19. 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)



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Ppp2r5b protein phosphatase 2, regulatory subunit B', beta [Mus musculus (house mouse)]

Gene ID: 225849, updated on 13-Mar-2020

Summary

Official Symbol	Ppp2r5b provided by MGI	
Official Full Name	protein phosphatase 2, regulatory subunit B', beta provided by MGI	
Primary source	MGI:MGI:2388480	
See related	Ensembl:ENSMUSG00000024777	
Gene type	protein coding	
RefSeq status	VALIDATED	
Organism	Mus musculus	
	Muroidea; Muridae; Murinae; Mus; Mus	
Also known as	B'beta, BC026670	
Expression	Ubiquitous expression in adrenal adult (RPKM 89.9), cerebellum adult (RPKM 51.2) and 23 other tissues See more	
Orthologs	human all	

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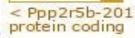
Transcript information (Ensembl)



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

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ppp2r5b-201	ENSMUST00000025695.9	2742	<u>497aa</u>	Protein coding	CCDS29499	Q6PD28	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Ppp2r5b-202	ENSMUST00000148693.1	467	No protein	Processed transcript	-	-	TSL:5
Ppp2r5b-204	ENSMUST00000236699.1	740	No protein	Retained intron	22	-	
Ppp2r5b-203	ENSMUST00000153155.1	389	No protein	Retained intron	20	2	TSL:3
	N // N /						

The strategy is based on the design of *Ppp2r5b-201* transcript, The transcription is shown below



Reverse strand -

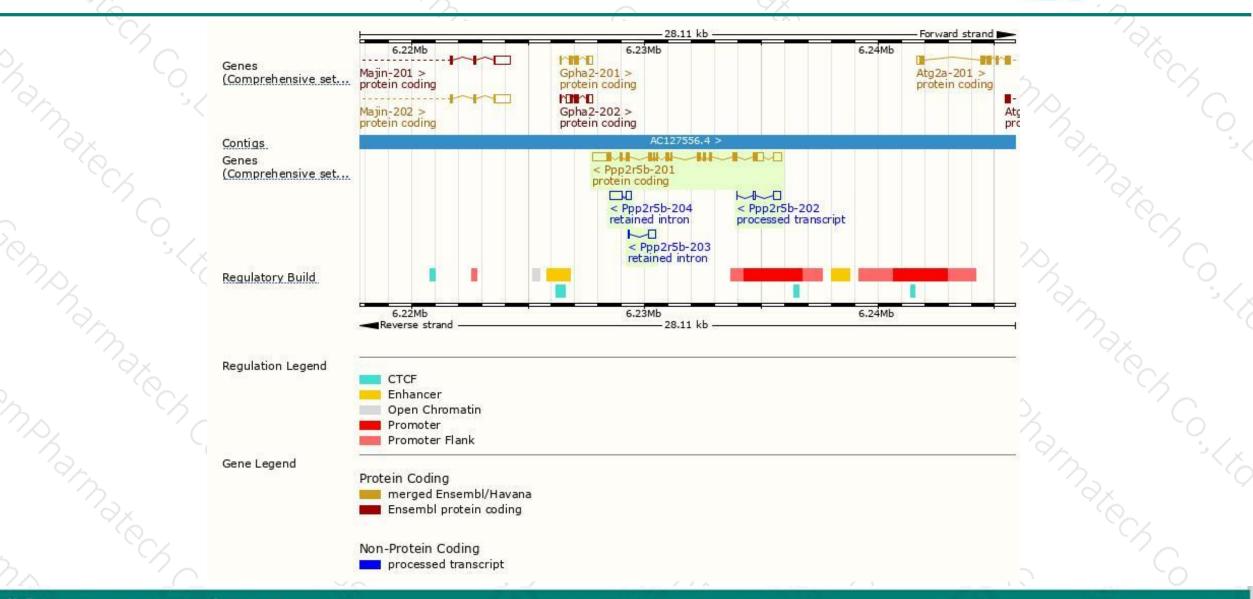
- 8.11 kb -

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Genomic location distribution



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Protein domain



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ENSMUSP00000025 MobiDB lite Low complexity (Seg) Superfamily		Armadillo-ty	pe fold							0		
Pfam	Protein phosphatase 2A, regulatory B subunit, B56											
PIRSF												
PANTHER	Protein phospha	tase 2A, regulat	ory B subunit, (856						0		
	PTHR10257:SF4									~<		
Gene3D		Armad illo-like	helical						2			
All sequence SNPs/i	Sequence vari	iants (dbSNP a	nd all other so	ources)	10 U	1			1			
Variant Legend												
Scale bar	0	60	120	180	240	300	360	420	497			
A Ch						N BY CH			C. C.			
	MobiDB lite Low complexity (Seg) Superfamily Pfam PIRSF PANTHER Gene3D All sequence SNPs/i Variant Legend	MobiDB lite Low complexity (Seg) Superfamily Pfam PIRSF PANTHER Protein phospha Protein phospha Protein phospha Protein phospha Protein phospha Protein phospha Protein phospha Protein phospha Sequence var Variant Legend Main Sequence var Sequence var Sequence var Sequence var Sequence var Sequence var Sequence var	MobiDB lite Low complexity (Seg) Superfamily Pfam Pfam Protein phosphatase 2A, regulat PRSF PANTHER PANTHER Gene3D Armadillo-like Armadillo-like Sequence variants (dbSNP a Variant Legend missense variant synonymous variant	MobiDB lite Low complexity (Seq) Superfamily Pfam Pfam PIRSF PANTHER Potein phosphatase 2A, regulatory B subunit, Protein phosphatase 2B, Armadillo-like helical Sequence variants (dbSNP and all other so Protein phosphatase variant Protein phosphat	MobiDB lite Low complexity (Seg) Superfamily Pfam Pfam Protein phosphatase 2A, regulatory B subunit, B56 PANTHER Protein phosphatase 2A, regulatory B subunit, B56 Protein phosphatase 2A, regulatory B subunit, B56	MobiDB lite Low complexity (Seg) Superfamily Pfam Pfam PRSF PANTHER Protein phosphatase 2A, regulatory B subunit, B56 PTHR10257:SF4 Gene3D Armadillo-like helical Variant Legend missense variant synonymous variant	MobiDB lite Armadillo-type fold Superfamily Armadillo-type fold Pfam. Protein phosphatase 2A, regulatory B subunit, B56 PIRSF. Protein phosphatase 2A, regulatory B subunit, B56 PANTHER. Protein phosphatase 2A, regulatory B subunit, B56 PERSF. Protein phosphatase 2A, regulatory B subunit, B56 PANTHER. Protein phosphatase 2A, regulatory B subunit, B56 Gene3D. Armadillo-like helical All sequence SNPs/i Sequence variants (dbSNP and all other sources) Variant Legend missense variant	MobiDB lite Armadillo-type fold Superfamily Armadillo-type fold Pfam Protein phosphatase 2A, regulatory B subunit, B56 PIRSE Protein phosphatase 2A, regulatory B subunit, B56 PANTHER Protein phosphatase 2A, regulatory B subunit, B56 PTHR10257:SF4 Armadillo-like helical Sequence SNPs/i Sequence variants (dbSNP and all other sources) Variant Legend missense variant	MobiDB lite Low complexity (Seg) Superfamily Pfam. Pfam. Protein phosphatase 2A, regulatory B subunit, B56 PIRSF. Protein phosphatase 2A, regulatory B subunit, B56 PANTHER. Protein phosphatase 2A, regulatory B subunit, B56 PANTHER. PTHR10257:SF4 Gene3D. Armadillo-like helical Sequence SNPs/i Variant Legend missense variant synonymous variant	MobiDB lite Low complexity. (Seq) Superfamily Pfam. Protein phosphatase 2A, regulatory B subunit, B56 PANTHER. Protein phosphatase 2A, regulatory B subunit, B56 Pantilio-like halical Sequence SNPs/i Variant Legend missense variant synonymous variant		

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If you have any questions, you are welcome to inquire. Tel: 400-9660890



