

Pacsin2 Cas9-CKO Strategy

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Project Overview

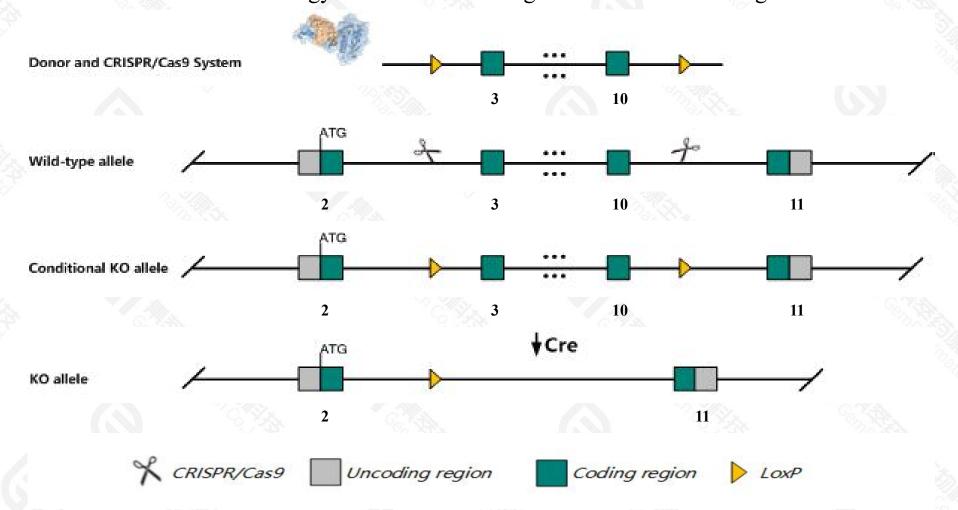


Project Name	Pacsin2			
Project type	Cas9-CKO			
Strain background	C57BL/6JGpt			

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Pacsin2* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Pacsin2* gene has 11 transcripts. According to the structure of *Pacsin2* gene, exon3-exon10 of *Pacsin2-203*(ENSMUST00000171436.8) transcript is recommended as the knockout region. The region contains 1288bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Pacsin2* 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.

Notice



- > According to the existing MGI data, mice homozygous for a null allele exhibit reduced running endurance, distance, and speed with impaired fetal cardiomyocyte electrophysiology.
- > The *Pacsin2* gene is located on the Chr15. 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)



Pacsin2 protein kinase C and casein kinase substrate in neurons 2 [Mus musculus (house mouse)]

Gene ID: 23970, updated on 17-Dec-2020

Summary



Official Symbol Pacsin2 provided by MGI

Official Full Name protein kinase C and casein kinase substrate in neurons 2 provided by MGI

Primary source MGI:MGI:1345153

See related Ensembl: ENSMUSG00000016664

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 Al197433, Sdpll

Expression Ubiquitous expression in heart adult (RPKM 34.6), kidney adult (RPKM 33.9) and 28 other tissuesSee more

Orthologs <u>human</u> all

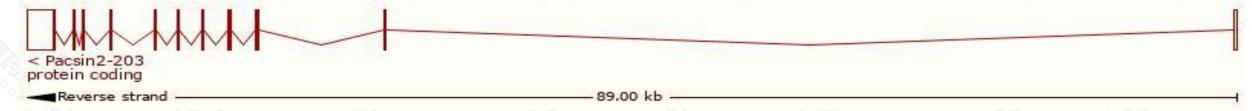
Transcript information (Ensembl)



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

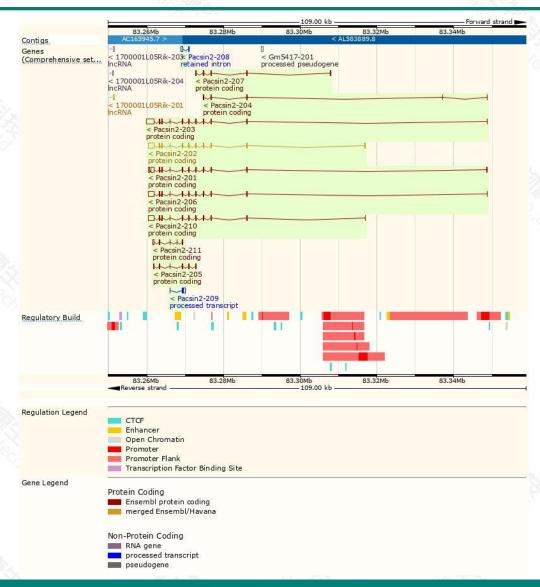
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pacsin2-203	ENSMUST00000171436.8	3693	486aa	Protein coding	CCDS27701		TSL:5 , GENCODE basic , APPRIS P1
Pacsin2-202	ENSMUST00000165095.9	3186	486aa	Protein coding	CCDS27701		TSL:1 , GENCODE basic , APPRIS P1
Pacsin2-210	ENSMUST00000231184.2	3103	486aa	Protein coding	CCDS27701		GENCODE basic , APPRIS P1 ,
Pacsin2-206	ENSMUST00000230679.2	3083	486aa	Protein coding	CCDS27701		GENCODE basic , APPRIS P1 ,
Pacsin2-201	ENSMUST00000056177.7	2795	<u>486aa</u>	Protein coding	CCDS27701		TSL:1 , GENCODE basic , APPRIS P1
Pacsin2-205	ENSMUST00000230030.2	857	272aa	Protein coding	-		CDS 5' incomplete ,
Pacsin2-211	ENSMUST00000231946.2	823	<u>180aa</u>	Protein coding	-		CDS 5' incomplete ,
Pacsin2-207	ENSMUST00000230816.2	765	<u>191aa</u>	Protein coding			CDS 3' incomplete ,
Pacsin2-204	ENSMUST00000229337.2	640	<u>124aa</u>	Protein coding	17.		CDS 3' incomplete ,
Pacsin2-209	ENSMUST00000231043.2	635	No protein	Processed transcript	-		
Pacsin2-208	ENSMUST00000230960.2	639	No protein	Retained intron	2		

The strategy is based on the design of *Pacsin2-203* transcript, the transcription is shown below:



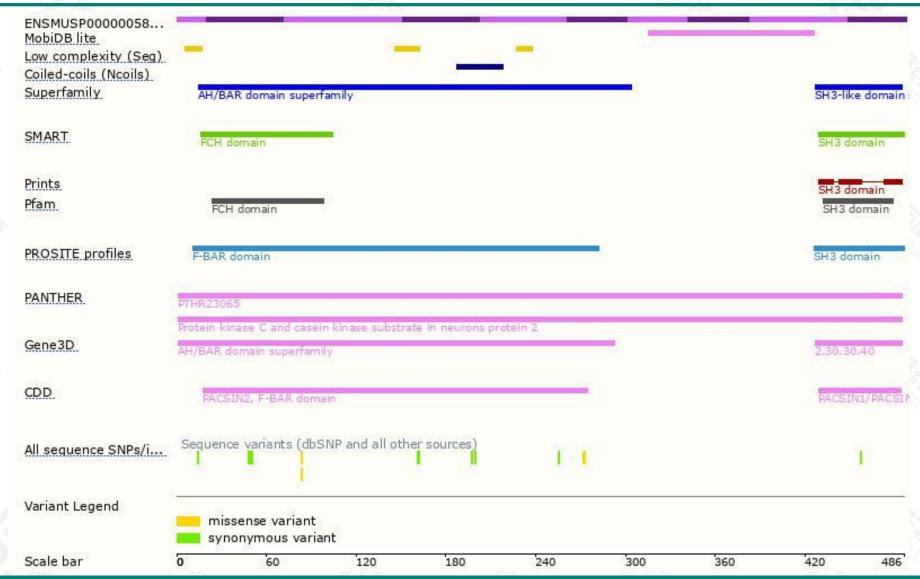
Genomic location distribution





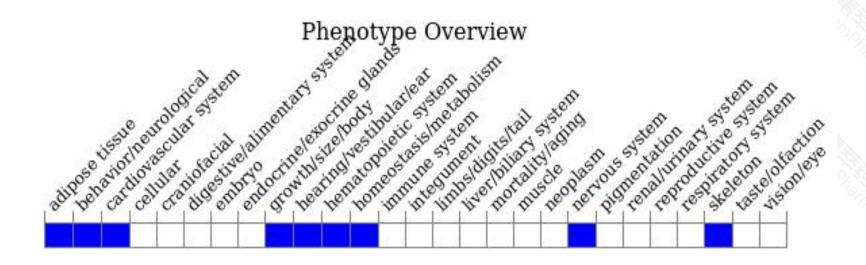
Protein domain





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, mice homozygous for a null allele exhibit reduced running endurance, distance, and speed with impaired fetal cardiomyocyte electrophysiology.



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

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