

Pacsin1 Cas9-CKO Strategy

Designer: JiaYu

Reviewer: Xiaojing Li

Design Date: 2020-1-17

Project Overview



Project Name

Pacsin1

Project type

Cas9-CKO

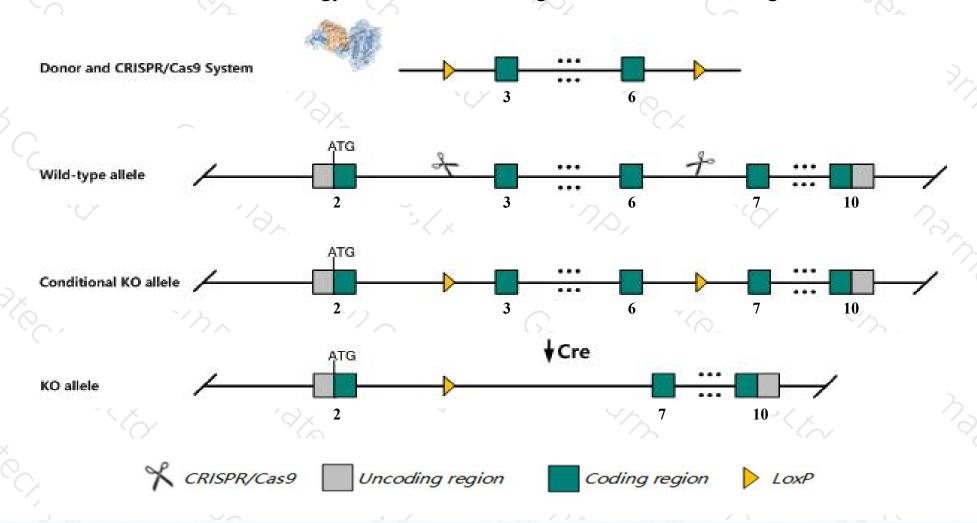
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Pacsin1* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Pacsin1* gene has 11 transcripts. According to the structure of *Pacsin1* gene, exon3-exon6 of *Pacsin1-211*(ENSMUST00000232437.1) transcript is recommended as the knockout region. The region contains 725bp coding sequence.

 Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Pacsin1* 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, Homozygotes for a gene trapped allele show altered type I interferon responses in plasmacytoid dendritic cells. Homozygotes for a null allele show impaired synaptic vesicle formation, synaptic transmission and neuronal network activity, and develop generalized seizures with tonic-clonic convulsions.
- The *Pacsin1* gene is located on the Chr17. 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)



Pacsin1 protein kinase C and casein kinase substrate in neurons 1 [Mus musculus (house mouse)]

Gene ID: 23969, updated on 7-Apr-2019

Summary



Official Symbol Pacsin1 provided by MGI

Official Full Name protein kinase C and casein kinase substrate in neurons 1 provided by MGI

Primary source MGI:MGI:1345181

See related Ensembl:ENSMUSG00000040276

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 A830061D09Rik, H74, mKIAA1379, syndapin

Expression Biased expression in frontal lobe adult (RPKM 48.4), cerebellum adult (RPKM 46.5) and 8 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
Pacsin1-211	ENSMUST00000232437.1	4566	441aa	Protein coding	CCDS28567	Q543Y7 Q61644	GENCODE basic APPRIS P1
Pacsin1-208	ENSMUST00000231669.1	4209	<u>441aa</u>	Protein coding	CCDS28567	Q543Y7 Q61644	GENCODE basic APPRIS P1
Pacsin1-201	ENSMUST00000045896.10	4176	<u>441aa</u>	Protein coding	CCDS28567	Q543Y7 Q61644	TSL:1 GENCODE basic APPRIS P1
Pacsin1-204	ENSMUST00000114873.7	1857	441aa	Protein coding	CCDS28567	Q543Y7 Q61644	TSL:1 GENCODE basic APPRIS P1
Pacsin1-202	ENSMUST00000097360.2	1800	<u>441aa</u>	Protein coding	CCDS28567	Q543Y7 Q61644	TSL:1 GENCODE basic APPRIS P1
Pacsin1-203	ENSMUST00000114872.8	972	320aa	Protein coding	-81	A0A384DVB1	CDS 5' incomplete TSL:1
Pacsin1-206	ENSMUST00000231236.1	672	109aa	Protein coding	20	A0A338P6K4	CDS 3' incomplete
Pacsin1-209	ENSMUST00000231854.1	1786	No protein	Retained intron	29	<u>.</u>	
Pacsin1-210	ENSMUST00000232225.1	697	No protein	Retained intron	-	5	
Pacsin1-207	ENSMUST00000231350.1	1368	No protein	IncRNA			
Pacsin1-205	ENSMUST00000155259.1	552	No protein	IncRNA	20	÷	TSL:2
	- / 1 /		77		7 1	V /	1 V.m.

The strategy is based on the design of *Pacsin1-211* transcript, The transcription is shown below

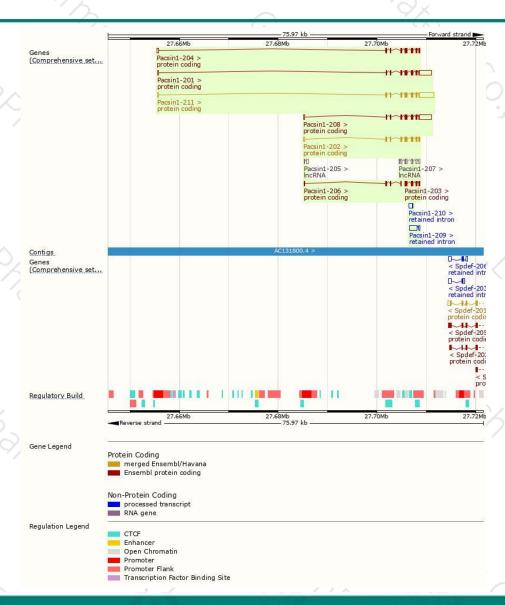
Pacsin1-211 > protein coding



55.90 kb

Genomic location distribution





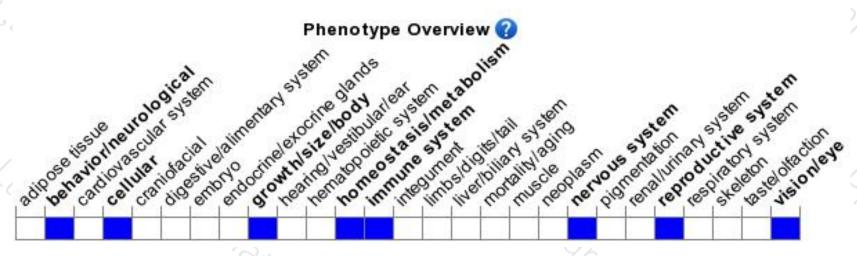
Protein domain





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, Homozygotes for a gene trapped allele show altered type I interferon responses in plasmacytoid dendritic cells. Homozygotes for a null allele show impaired synaptic vesicle formation, synaptic transmission and neuronal network activity, and develop generalized seizures with tonic-clonic convulsions.



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





