Ptcd3 Cas9-CKO Strategy

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Reviewer: Huimin Su

Design Date: 2019-9-10

Project Overview



Project Name

Ptcd3

Project type

Cas9-CKO

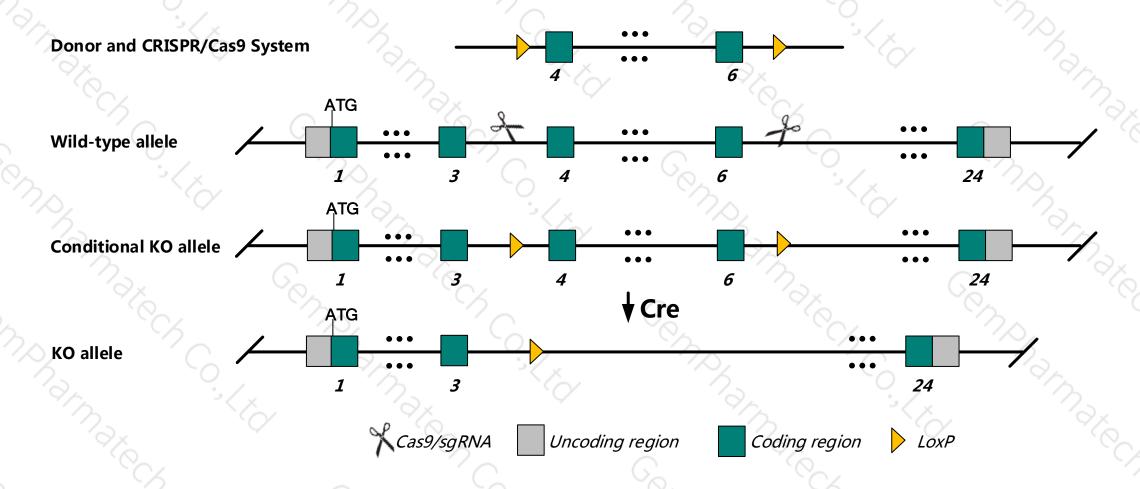
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Ptcd3* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Ptcd3* gene has 11 transcript. According to the structure of *Ptcd3* gene, exon4-6 of *Ptcd3*-201 (ENSMUST00000082094.4) transcript is recommended as the knockout region. The region contains 220bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ptcd3* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues or cell types.

Notice



- The *Ptcd3* gene is located on the Chr6. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)



Ptcd3 pentatricopeptide repeat domain 3 [Mus musculus (house mouse)]

Gene ID: 69956, updated on 12-Aug-2019

Summary



Official Symbol Ptcd3 provided by MGI

Official Full Name pentatricopeptide repeat domain 3 provided by MGI

Primary source MGI:MGI:1917206

See related Ensembl:ENSMUSG00000063884

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 MRP-S39; AA589622; AU045708; 2610034F17Rik; 2810422B04Rik

Expression Ubiquitous expression in CNS E11.5 (RPKM 15.2), liver E14 (RPKM 12.3) and 24 other tissues See more

Orthologs human all

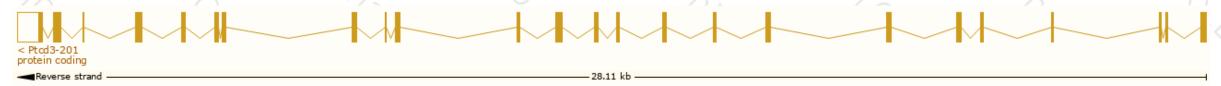
Transcript information (Ensembl)



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

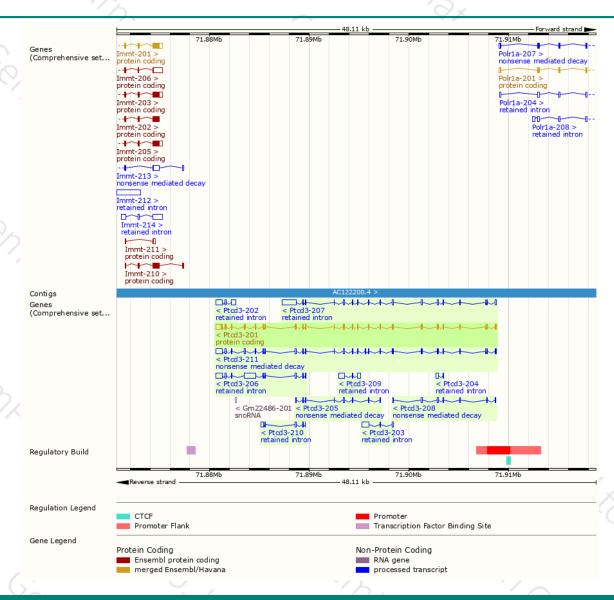
F 7%							
Name	Transcript ID	bp 🛊	Protein	Biotype	CCDS	UniProt	Flags
Ptcd3-201	ENSMUST00000082094.4	2581	<u>685aa</u>	Protein coding	CCDS20235®	<u>Q14C51</u> ₽	TSL:1 GENCODE basic APPRIS P1
Ptcd3-211	ENSMUST00000206879.1	2666	284aa	Nonsense mediated decay	-	<u>A0A0U1RQ61</u> ₽	TSL:1
Ptcd3-205	ENSMUST00000205556.1	588	<u>51aa</u>	Nonsense mediated decay	-	<u>A0A0U1RP36</u> ₽	CDS 5' incomplete TSL:5
Ptcd3-208	ENSMUST00000206284.1	545	<u>59aa</u>	Nonsense mediated decay	-	<u>A0A0U1RNH5</u> ₽	CDS 5' incomplete TSL:3
Ptcd3-207	ENSMUST00000205761.1	2680	No protein	Retained intron	-	-	TSL:1
Ptcd3-206	ENSMUST00000205691.1	2364	No protein	Retained intron	-	-	TSL:2
Ptcd3-202	ENSMUST00000205269.1	1161	No protein	Retained intron	-	-	TSL:1
Ptcd3-209	ENSMUST00000206631.1	949	No protein	Retained intron	-	-	TSL:2
Ptcd3-203	ENSMUST00000205293.1	853	No protein	Retained intron	-	-	TSL:2
Ptcd3-210	ENSMUST00000206762.1	521	No protein	Retained intron	-	-	TSL:5
Ptcd3-204	ENSMUST00000205420.1	320	No protein	Retained intron	-	-	TSL:2

The strategy is based on the design of *Ptcd3*-201 transcript, The transcription is shown below:



Genomic location distribution





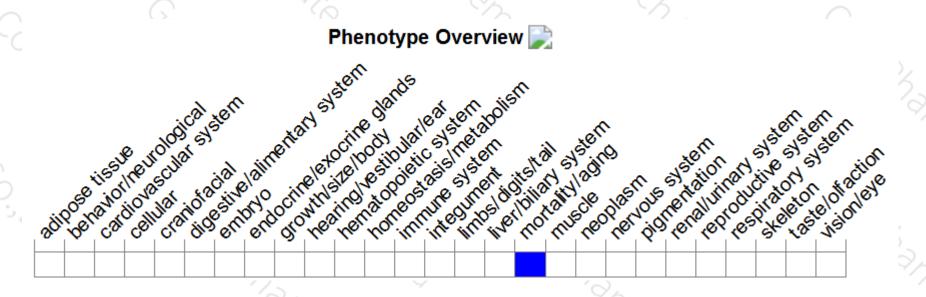
Protein domain





Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

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





