

# Plce1 Cas9-CKO Strategy

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Reviewer: Huimin Su

**Design Date:** 2020-2-24

## **Project Overview**



**Project Name** 

Plce1

**Project type** 

Cas9-CKO

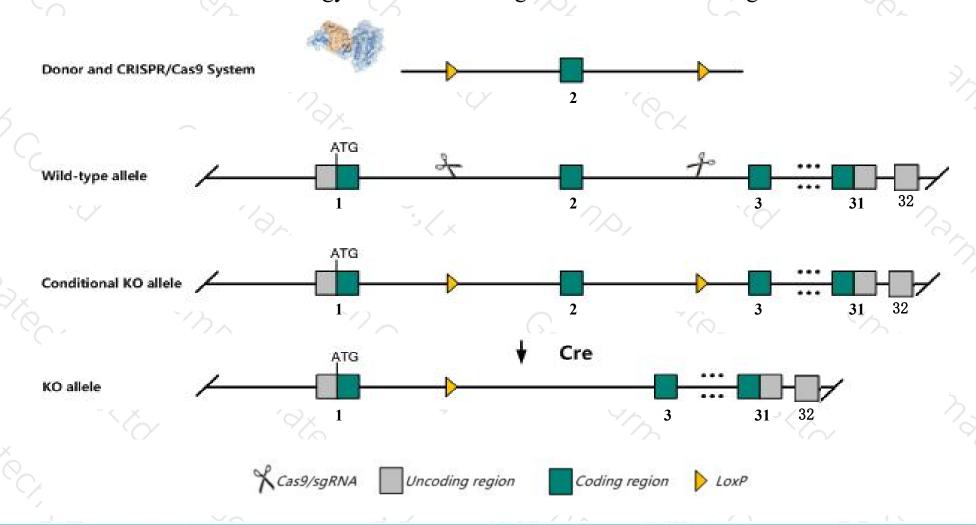
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Plce1* gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Plce1* gene has 7 transcripts. According to the structure of *Plce1* gene, exon2 of *Plce1-201*(ENSMUST00000169713.8) transcript is recommended as the knockout region. The region contains 289bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Plce1* 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, homozygous mutation of this gene results in a congenital semilunar valvulogenesis defect which causes regurgitation and stenosis, and decreased incidence of induced skin tumors. Another mutant exhibits decreased cardiac contraction and increased hypertrophy in response to chronic stress.
- > The *Plce1* 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)



#### Pice1 phospholipase C, epsilon 1 [ Mus musculus (house mouse) ]

Gene ID: 74055, updated on 10-Oct-2019

#### Summary



Official Symbol Plce1 provided by MGI

Official Full Name phospholipase C, epsilon 1 provided by MGI

Primary source MGI:MGI:1921305

See related Ensembl: ENSMUSG00000024998

Gene type protein coding
RefSeq status VALIDATED
Organism Mus musculus

Organism Mus musculus

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae;

Murinae; Mus; Mus

Also known as Plce; mKIAA1516; PLCepsilon; 4933403A21Rik

Expression Ubiquitous expression in bladder adult (RPKM 5.0), limb E14.5 (RPKM 3.2) and 26 other tissues See more

Orthologs human all

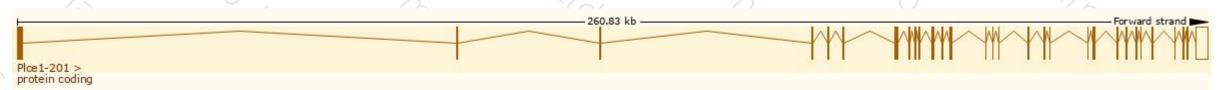
## Transcript information (Ensembl)



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

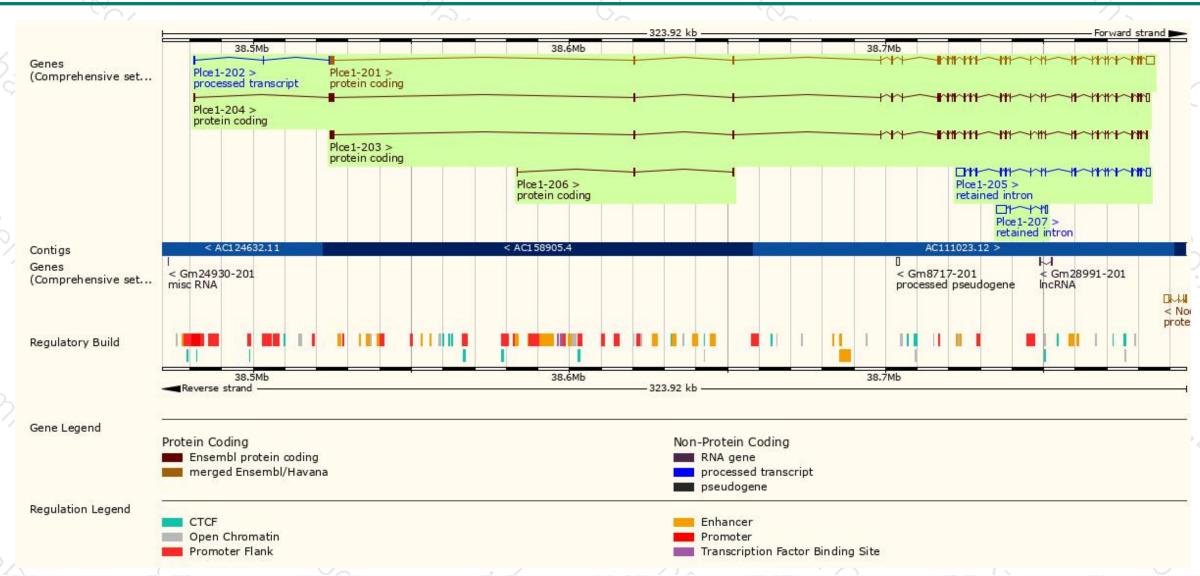
1 1				7 /		796	7
Name	Transcript ID 🝦	bp 🛊	Protein 🍦	Biotype 🖕	CCDS 🍦	UniProt	Flags
Pice1-201	ENSMUST00000169713.8	9439	2282aa	Protein coding	CCDS37973 ₽	Q8K4S1₽	TSL:1 GENCODE basic APPRIS P2
PIce1-204	ENSMUST00000182481.7	8357	2282aa	Protein coding	CCDS37973&	<u>Q8K4S1</u> 函	TSL:5 GENCODE basic APPRIS P2
Pice1-203	ENSMUST00000182267.1	7234	2296aa	Protein coding		<u>S4R1Q8</u> 률	TSL:1 GENCODE basic APPRIS ALT2
Plce1-206	ENSMUST00000182999.1	619	<u>74aa</u>	Protein coding	-	S4R168&	CDS 3' incomplete TSL:3
PIce1-202	ENSMUST00000181994.1	473	No protein	Processed transcript	12	(27)	TSL:2
Pice1-205	ENSMUST00000182589.7	6769	No protein	Retained intron	-	-	TSL:2
Pice1-207	ENSMUST00000183131.1	4220	No protein	Retained intron	=	-	TSL:2

The strategy is based on the design of *Plce1-201* transcript, the transcription is shown below:



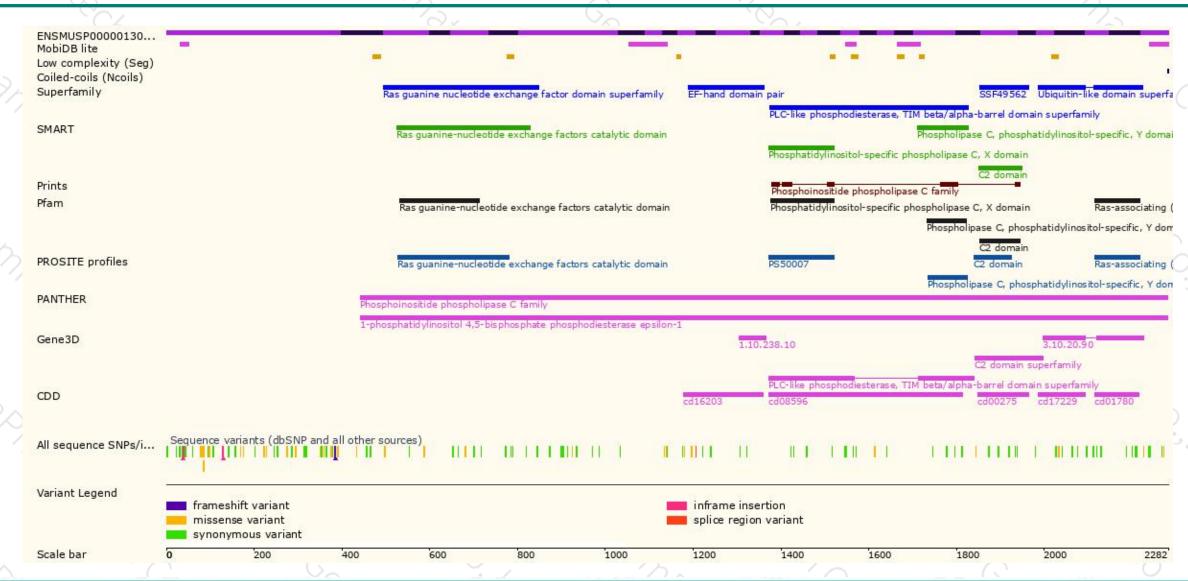
## Genomic location distribution





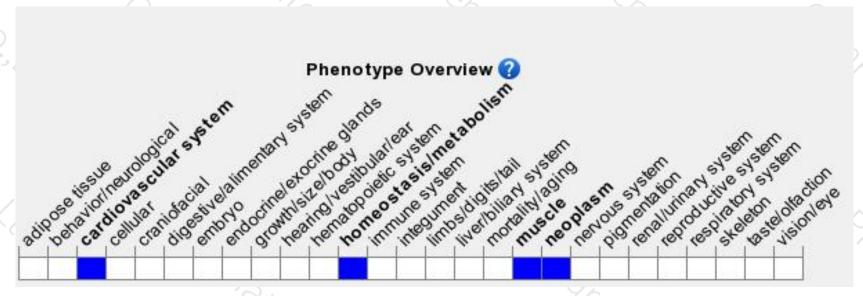
### Protein domain





## Mouse phenotype description(MGI)





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

According to the existing MGI data, homozygous mutation of this gene results in a congenital semilunar valvulogenesis defect which causes regurgitation and stenosis, and decreased incidence of induced skin tumors. Another mutant exhibits decreased cardiac contraction and increased hypertrophy in response to chronic stress.



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





