

# Plcd4 Cas9-KO Strategy

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# **Project Overview**



**Project Name** 

Project type Cas9-KO

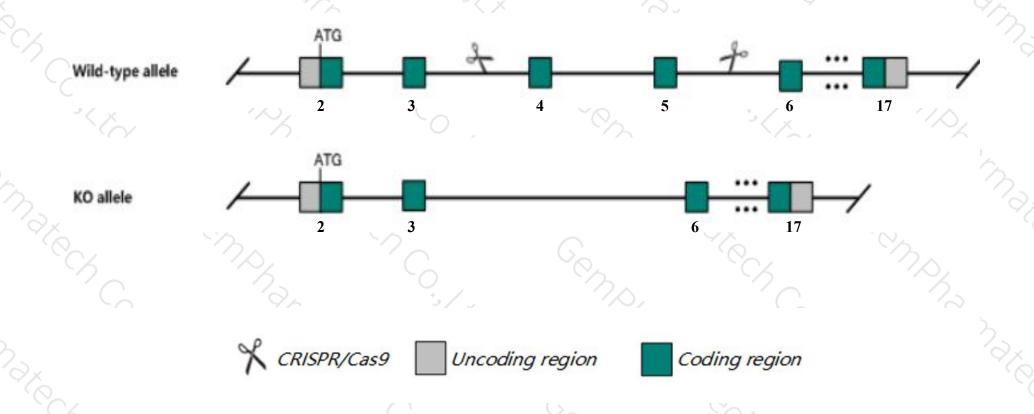
Strain background C57BL/6JGpt

Plcd4

# **Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Plcd4* gene. The schematic diagram is as follows:



#### **Technical routes**



- ➤ The *Plcd4* gene has 11 transcripts. According to the structure of *Plcd4* gene, exon4-exon5 of *Plcd4*-201(ENSMUST00000027362.13) transcript is recommended as the knockout region. The region contains 359bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Plcd4* gene. The brief process is as follows: CRISPR/Cas9 system 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.

#### **Notice**



- > According to the existing MGI data, males homozygous for a targeted null mutation are subfertile or sterile. Sperm from mutant males fail to initiate the acrosome reaction.
- > The *Plcd4* gene is located on the Chr1. 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 gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

## Gene information (NCBI)



Plcd4 phospholipase C, delta 4 [Mus musculus (house mouse)]

Gene ID: 18802, updated on 13-Mar-2020





Official Symbol Plcd4 provided by MGI

Official Full Name phospholipase C, delta 4 provided by MGI

Primary source MGI:MGI:107469

See related Ensembl: ENSMUSG000000026173

Gene type protein coding

RefSeq status VALIDATED

Organism <u>Mus musculus</u>

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

Muroidea; Muridae; Murinae; Mus; Mus

Also known as 4921507K24Rik

Expression Biased expression in testis adult (RPKM 22.6), cerebellum adult (RPKM 4.2) and 3 other tissuesSee more

Orthologs <u>human</u> <u>all</u>

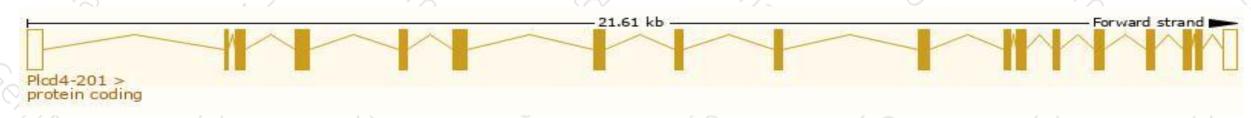
# Transcript information (Ensembl)



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

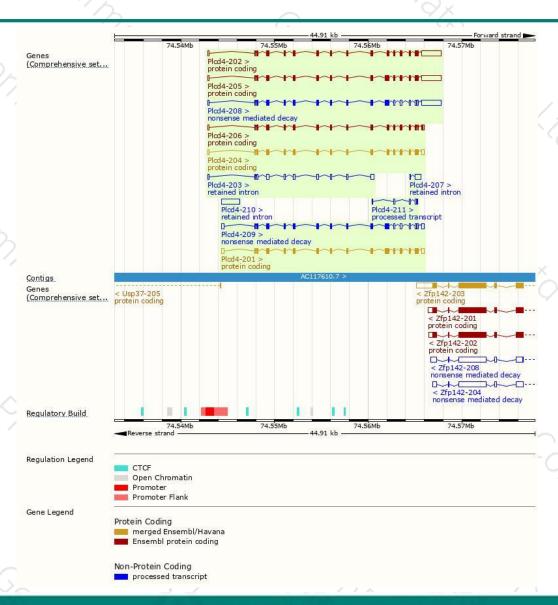
			1/3				
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Plcd4-201	ENSMUST00000027362.13	2977	807aa	Protein coding	CCDS35616	Q8K3R3	TSL:1 GENCODE basic APPRIS P4
Plcd4-204	ENSMUST00000113747.7	2720	<u>775aa</u>	Protein coding	CCDS35617	Q8K3R3	TSL:1 GENCODE basic APPRIS ALT:
Plcd4-205	ENSMUST00000113749.7	4648	807aa	Protein coding	12	Q8K3R3	TSL:5 GENCODE basic APPRIS ALT:
Plcd4-202	ENSMUST00000067916.12	4552	<u>775aa</u>	Protein coding		Q8K3R3	TSL:5 GENCODE basic APPRIS ALT
Plcd4-206	ENSMUST00000113750.7	2692	<u>798aa</u>	Protein coding	2	Q8K3R3	TSL:1 GENCODE basic APPRIS ALT
Plcd4-208	ENSMUST00000141412.7	4682	<u>574aa</u>	Nonsense mediated decay	-	D6RGN2	TSL:1
Plcd4-209	ENSMUST00000152707.7	3011	<u>574aa</u>	Nonsense mediated decay	:=	D6RGN2	TSL:5
Plcd4-211	ENSMUST00000189956.1	512	No protein	Processed transcript	2	-	TSL:3
Plcd4-210	ENSMUST00000185411.1	1912	No protein	Retained intron	=	=	TSL:NA
Plcd4-203	ENSMUST00000113745.7	1749	No protein	Retained intron	:=	#	TSL:1
Plcd4-207	ENSMUST00000135111.1	671	No protein	Retained intron	-	25	TSL:3
				/ 1			

The strategy is based on the design of *Plcd4-201* transcript, the transcription is shown below:



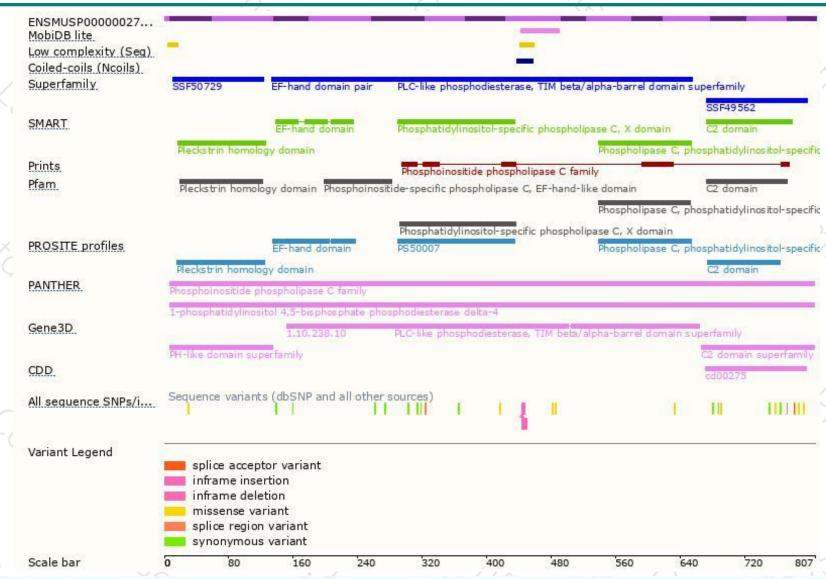
### Genomic location distribution





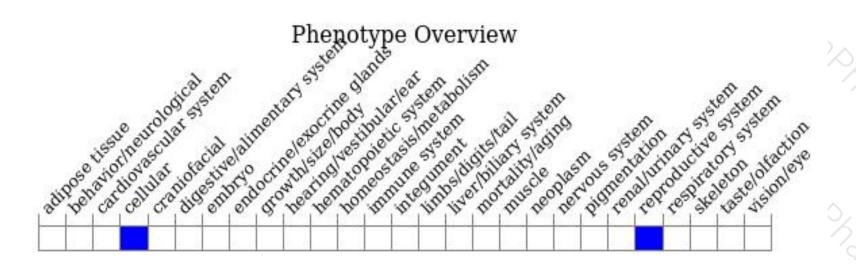
#### Protein domain





## 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, males homozygous for a targeted null mutation are subfertile or sterile. Sperm from mutant males fail to initiate the acrosome reaction.



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





