

# Plaa Cas9-CKO Strategy

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## **Project Overview**



**Project Name** 

Project type

Strain background

Cas9-CKO

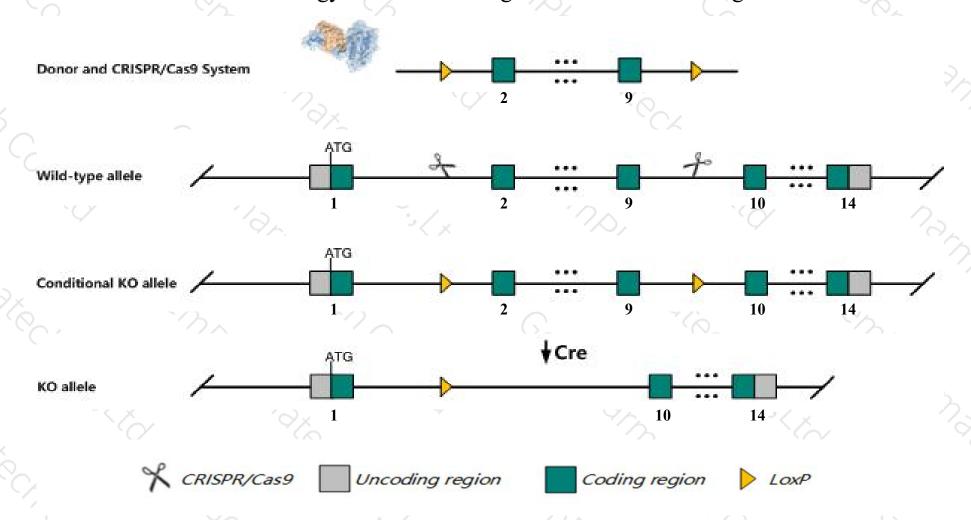
Plaa

C57BL/6JGpt

## Conditional Knockout strategy



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



### Technical routes



- ➤ The *Plaa* gene has 6 transcripts. According to the structure of *Plaa* gene, exon2-exon9 of *Plaa-201*(ENSMUST00000107107.8) transcript is recommended as the knockout region. The region contains 1268bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Plaa* 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 KO is embryonic lethal. A hypomorphic homozygous point mutation affects neuromuscular junctions and Purkinje cell development, causing early-onset neurodysfunction.
- The *Plaa* gene is located on the Chr4. 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)



#### Plaa phospholipase A2, activating protein [Mus musculus (house mouse)]

Gene ID: 18786, updated on 31-Jan-2019

#### Summary

☆ ?

Official Symbol Plaa provided by MGI

Official Full Name phospholipase A2, activating protein provided by MGI

Primary source MGI:MGI:104810

See related Ensembl: ENSMUSG00000028577

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 2410007N06, Al536418, AU018445, AW208417, D4Ertd618e, PLA2P, PLAP, Ufd3

Expression Ubiquitous expression in CNS E11.5 (RPKM 9.7), placenta adult (RPKM 8.4) and 28 other tissuesSee more

Orthologs <u>human</u> all

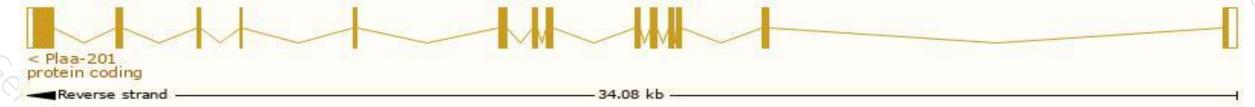
## Transcript information (Ensembl)



The gene has 6 transcripts, all transcripts are shown below:

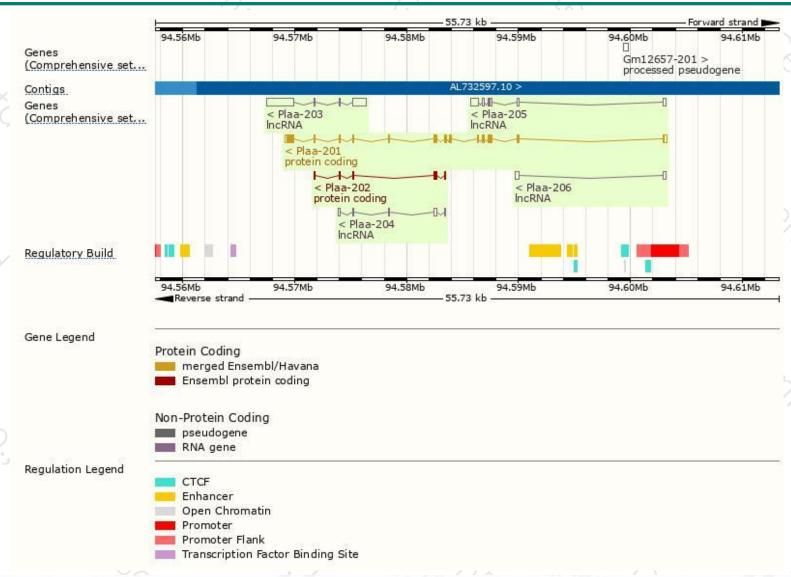
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Plaa-201	ENSMUST00000107107.8	2784	<u>794aa</u>	Protein coding	CCDS18359	P27612	TSL:1 GENCODE basic APPRIS P1
Plaa-202	ENSMUST00000127656.2	641	<u>214aa</u>	Protein coding	-	F7D1R5	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:2
Plaa-203	ENSMUST00000129748.1	3886	No protein	IncRNA	ų.	94	TSL:2
Plaa-205	ENSMUST00000136669.1	1514	No protein	IncRNA	-	(4)	TSL:1
Plaa-206	ENSMUST00000146118.1	693	No protein	IncRNA		65	TSL:2
Plaa-204	ENSMUST00000135696.1	645	No protein	IncRNA		19 <del>0</del>	TSL:3

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



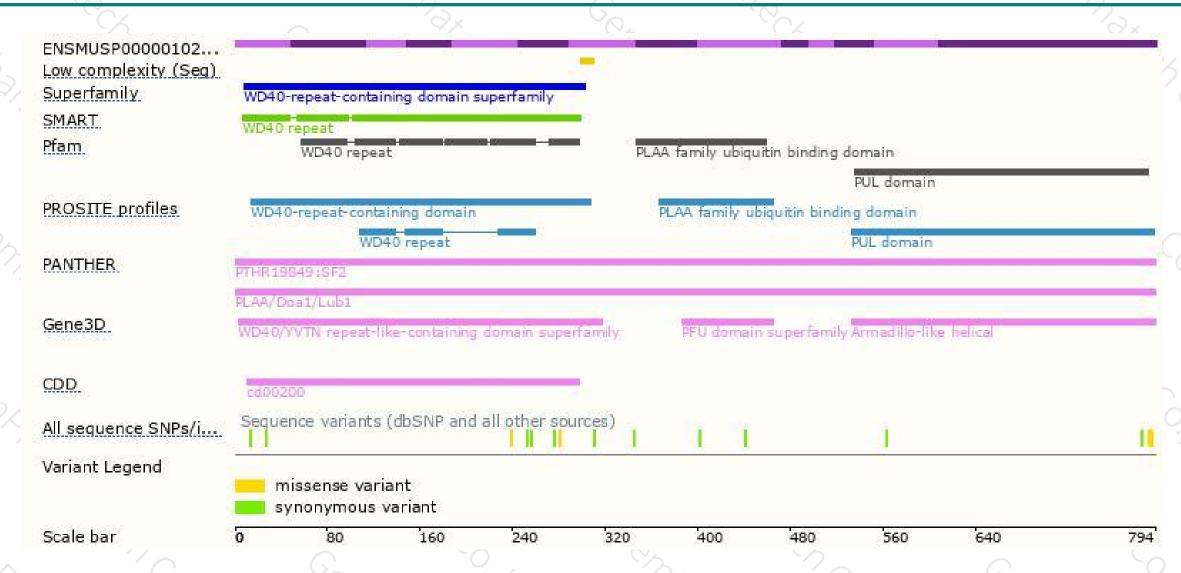
### Genomic location distribution





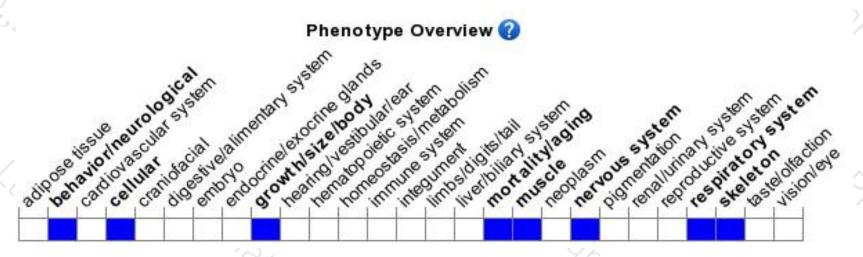
### Protein domain





## 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 KO is embryonic lethal. A hypomorphic homozygous point mutation affects neuromuscular junctions and Purkinje cell development, causing early-onset neurodysfunction.



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





