

Pde6a Cas9-CKO Strategy

Designer:Xueting Zhang

Reviewer: Yanhua Shen

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Project Overview



Project Name

Pde6a

Project type

Cas9-CKO

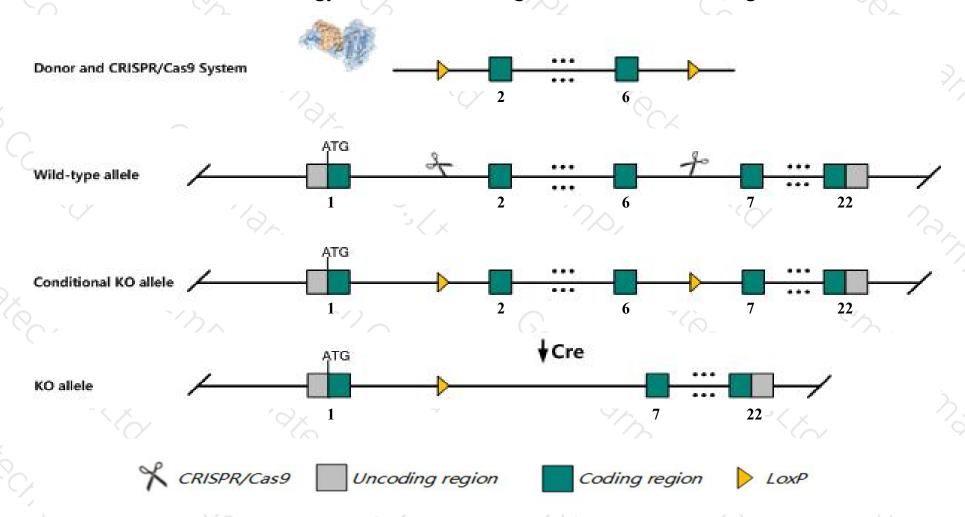
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Pde6a* gene. The schematic diagram is as follows:



Technical routes



- The *Pde6a* gene has 2 transcripts. According to the structure of *Pde6a* gene, exon2-exon6 of *Pde6a-201*(ENSMUST00000025468.14) transcript is recommended as the knockout region. The region contains 524bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Pde6a* 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 mutant mice have retinal degeneration.
- ➤ The effect on transcript *Pde6a*-202 is unknown.
- ➤ The N-terminal of *Pde6a* gene will remain 158aa,it may remain the partial function of *Pde6a* gene.
- > The *Pde6a* gene is located on the Chr18. 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)



Pde6a phosphodiesterase 6A, cGMP-specific, rod, alpha [Mus musculus (house mouse)]

Gene ID: 225600, updated on 12-Aug-2019

Summary

☆ ?

Official Symbol Pde6a provided by MGI

Official Full Name phosphodiesterase 6A, cGMP-specific, rod, alpha provided by MGI

Primary source MGI:MGI:97524

See related Ensembl: ENSMUSG00000024575

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 Pdea; nmf282

Expression Low expression observed in reference dataset <u>See more</u>

Orthologs human all

Genomic context

↑ ?

Location: 18 E1; 18 34.41 cM

See Pde6a in Genome Data Viewer

Exon count: 23

Annotation release	Status	Assembly	Chr	Location
<u>108</u>	current	GRCm38.p6 (GCF 000001635.26)	18	NC_000084.6 (6122047461289750)
Build 37.2	previous assembly	MGSCv37 (GCF 000001635.18)	18	NC_000084.5 (6138015361449404)

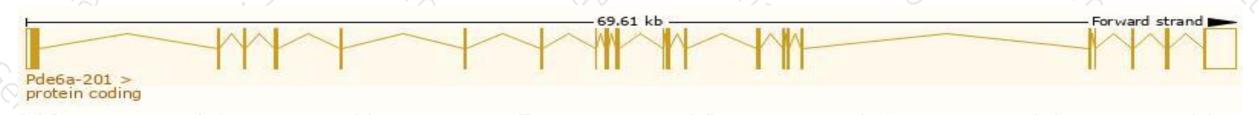
Transcript information (Ensembl)



The gene has 2 transcripts, all transcripts are shown below:

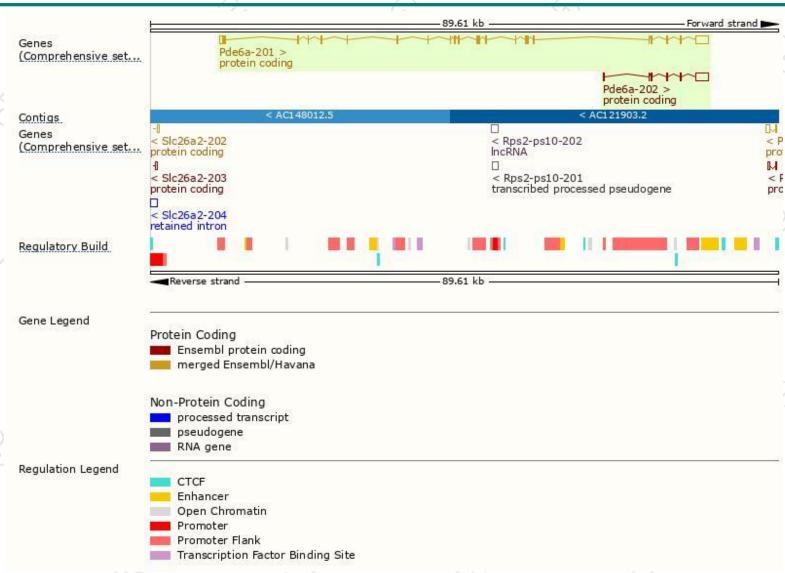
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pde6a-201	ENSMUST00000025468.14	4588	860aa	Protein coding	CCDS29283	Q8K0A8	TSL:1 GENCODE basic APPRIS P1
Pde6a-202	ENSMUST00000135688.1	2263	<u>182aa</u>	Protein coding	- 8	F6UDG9	CDS 5' incomplete TSL:1

The strategy is based on the design of *Pde6a-201* transcript, The transcription is shown below



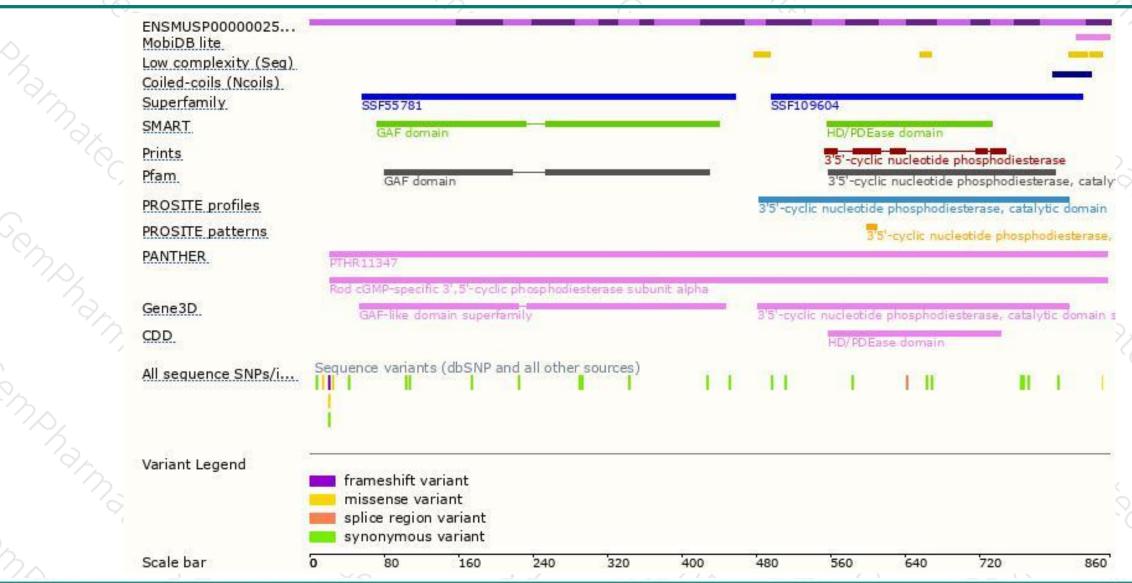
Genomic location distribution





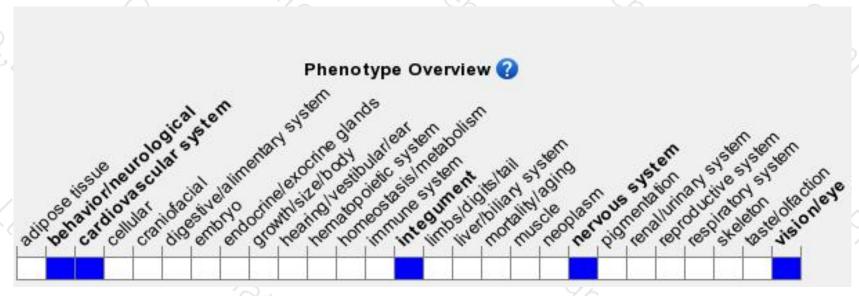
Protein domain





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 mutant mice have retinal degeneration.



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





