

# Pkd2 Cas9-CKO Strategy

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Reviewer: yun Li

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# **Project Overview**



**Project Name** 

Pkd2

**Project type** 

Cas9-CKO

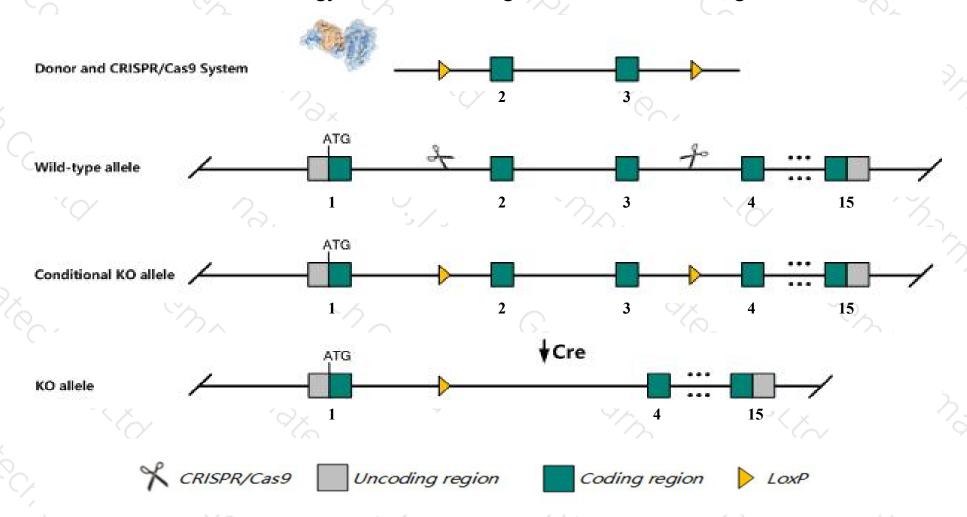
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Pkd2* gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Pkd2* gene has 3 transcripts. According to the structure of *Pkd2* gene, exon2-exon3 of *Pkd2*201(ENSMUST00000086831.3) transcript is recommended as the knockout region. The region contains 248bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Pkd2* 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, homozygotes for targeted null mutations exhibit defects in cardiac septation, kidney and pancreatic cysts, impaired left-right axis determination, and late-gestation lethality. Heterozygotes show kidney and liver lesions and have reduced longevity.
- > The Pkd2 gene is located on the Chr5. 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)



Pkd2 polycystin 2, transient receptor potential cation channel [Mus musculus (house mouse)]

Gene ID: 18764, updated on 13-Mar-2020





Official Symbol Pkd2 provided by MGI

Official Full Name polycystin 2, transient receptor potential cation channel provided by MGI

Primary source MGI:MGI:1099818

See related Ensembl: ENSMUSG00000034462

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 C030034P18Rik, PC2, TRPP2

Expression Ubiquitous expression in limb E14.5 (RPKM 13.0), bladder adult (RPKM 11.6) and 27 other tissuesSee more

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

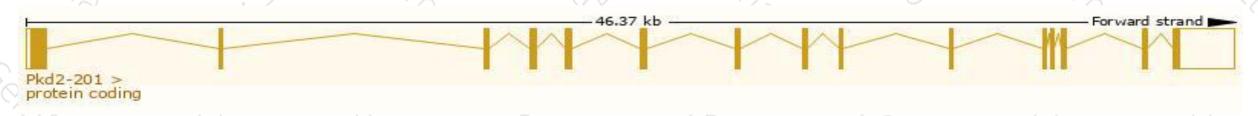
# Transcript information (Ensembl)



The gene has 3 transcripts, all transcripts are shown below:

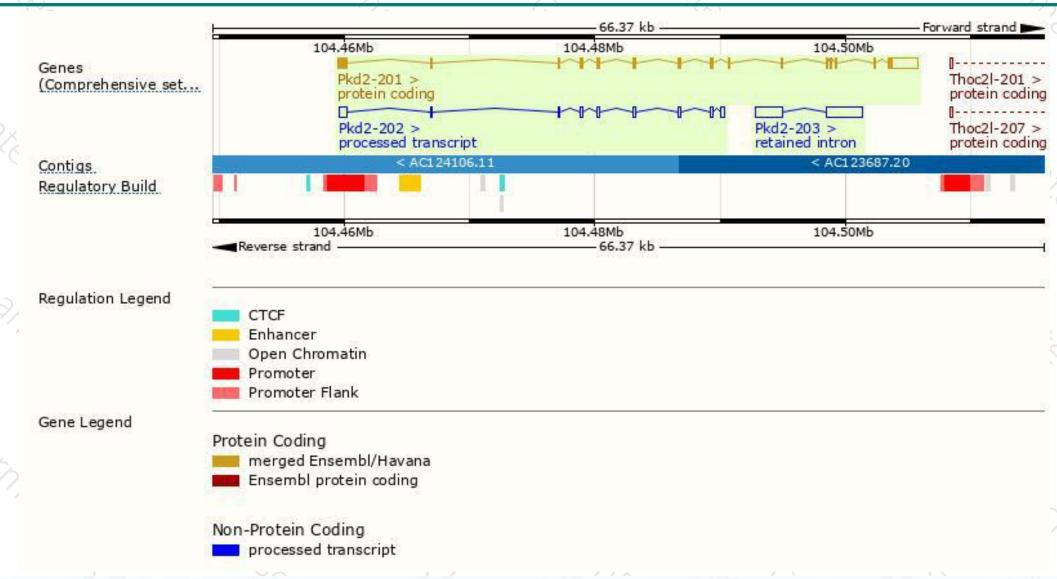
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pkd2-201	ENSMUST00000086831.3	5219	966aa	Protein coding	CCDS19487	035245	TSL:1 GENCODE basic APPRIS P1
Pkd2-202	ENSMUST00000130931.1	2282	No protein	Processed transcript	5-E	-	TSL:1
Pkd2-203	ENSMUST00000133540.1	5130	No protein	Retained intron	0.20	- 1	TSL:2

The strategy is based on the design of *Pkd2-201* transcript,the transcription is shown below:



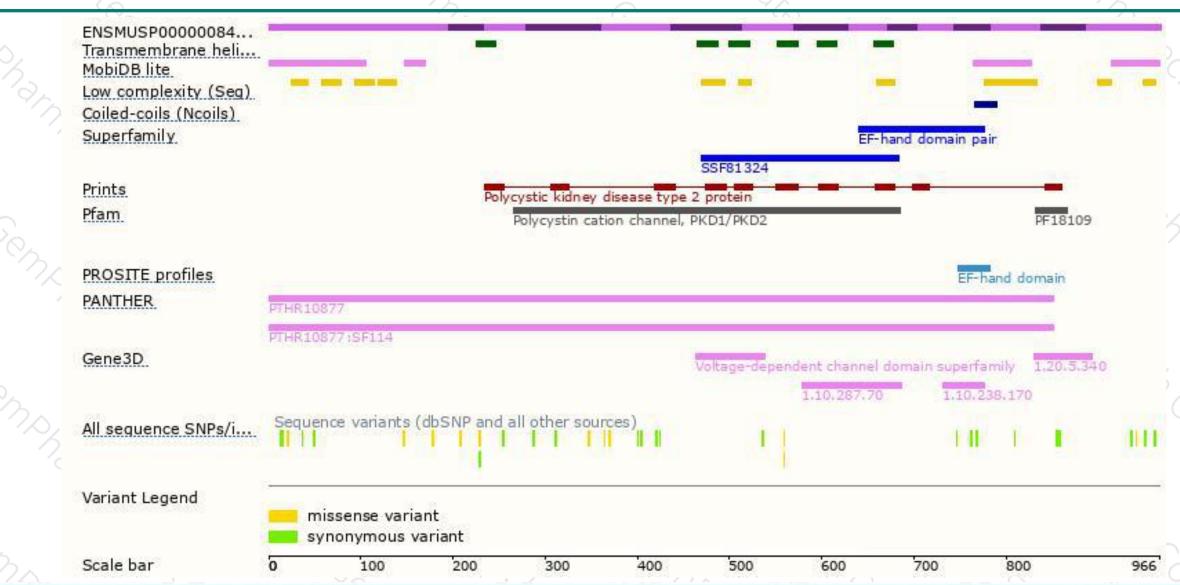
#### Genomic location distribution





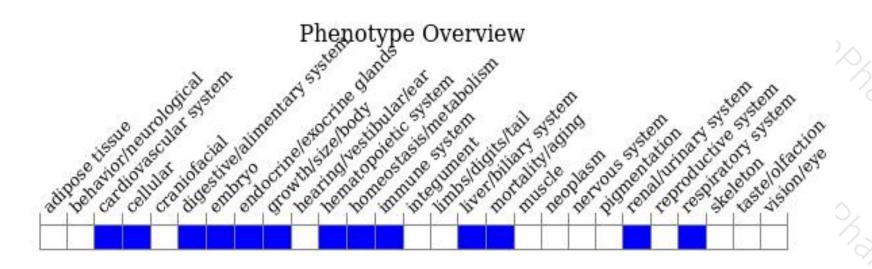
#### Protein domain





## 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, homozygotes for targeted null mutations exhibit defects in cardiac septation, kidney and pancreatic cysts, impaired left-right axis determination, and late-gestation lethality. Heterozygotes show kidney and liver lesions and have reduced longevity.



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





