Pkhd1 Cas9-CKO Strategy

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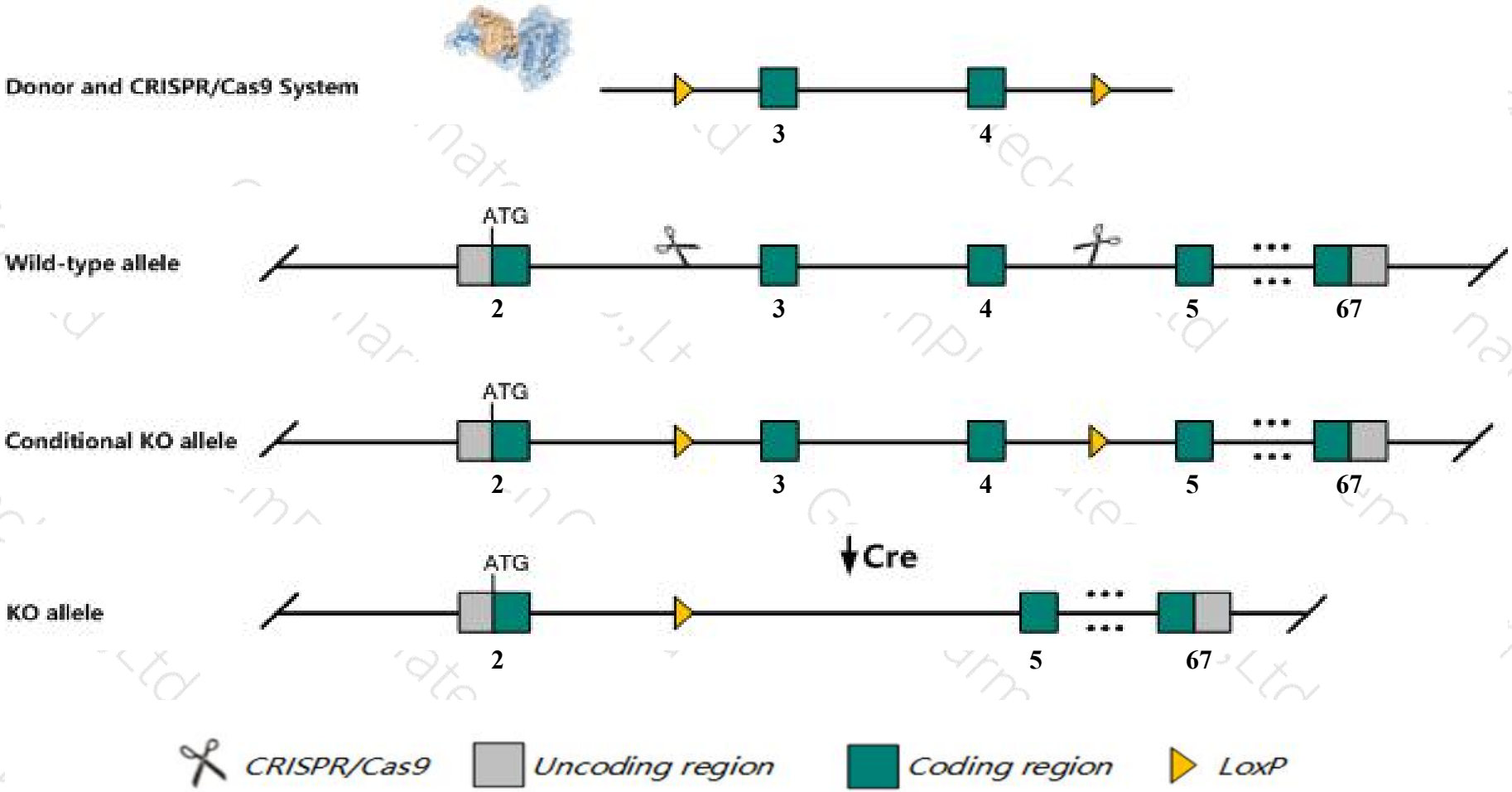
Project Overview

Project Name	***Pkhd1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Pkhd1* gene. The schematic diagram is as follows:



Technical routes

- The *Pkhd1* gene has 4 transcripts. According to the structure of *Pkhd1* gene, exon3-exon4 of *Pkhd1-201* (ENSMUST00000088448.11) transcript is recommended as the knockout region.The region contains 220bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Pkhd1* 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,Mice homozygous for a mutation in this gene display variable progressive liver cysts and fibrosis, but do not display kidney cysts and are fertile. Mice homozygous for a hypomorphic and null allele display renal, pancreatic, billiary and liver cysts.

➢ The *Pkhd1* 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information （NCBI）

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Pkhd1 polycystic kidney and hepatic disease 1 [Mus musculus (house mouse)]

Gene ID: 241035, updated on 19-Mar-2019

Summary

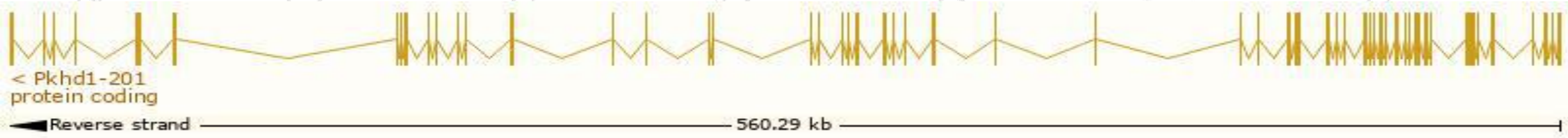
Official Symbol	Pkhd1 provided by MGI
Official Full Name	polycystic kidney and hepatic disease 1 provided by MGI
Primary source	MGI:MGI:2155808
See related	Ensembl:ENSMUSG00000043760
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI118496, AI182499, FPC, Tigm1
Expression	Biased expression in kidney adult (RPKM 6.2), genital fat pad adult (RPKM 1.3) and 3 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

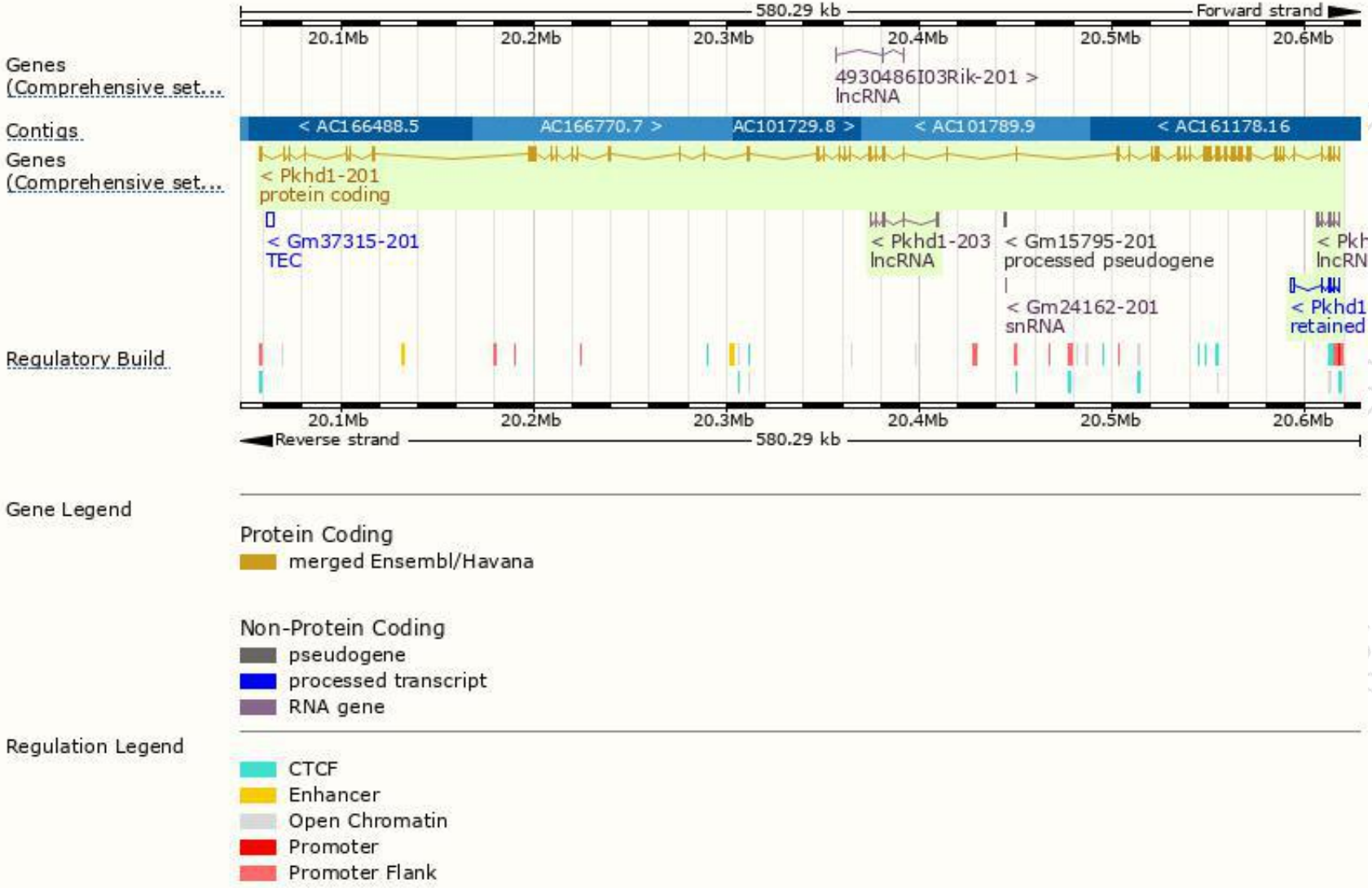
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pkhd1-201	ENSMUST00000088448.11	12935	4059aa	Protein coding	CCDS14841	E9PZ36	TSL:1 GENCODE basic APPRIS P1
Pkhd1-204	ENSMUST00000147480.7	1636	No protein	Retained intron	-	-	TSL:1
Pkhd1-202	ENSMUST00000128051.1	921	No protein	lncRNA	-	-	TSL:1
Pkhd1-203	ENSMUST00000132647.1	615	No protein	lncRNA	-	-	TSL:3

The strategy is based on the design of *Pkhd1-201* transcript,The transcription is shown below

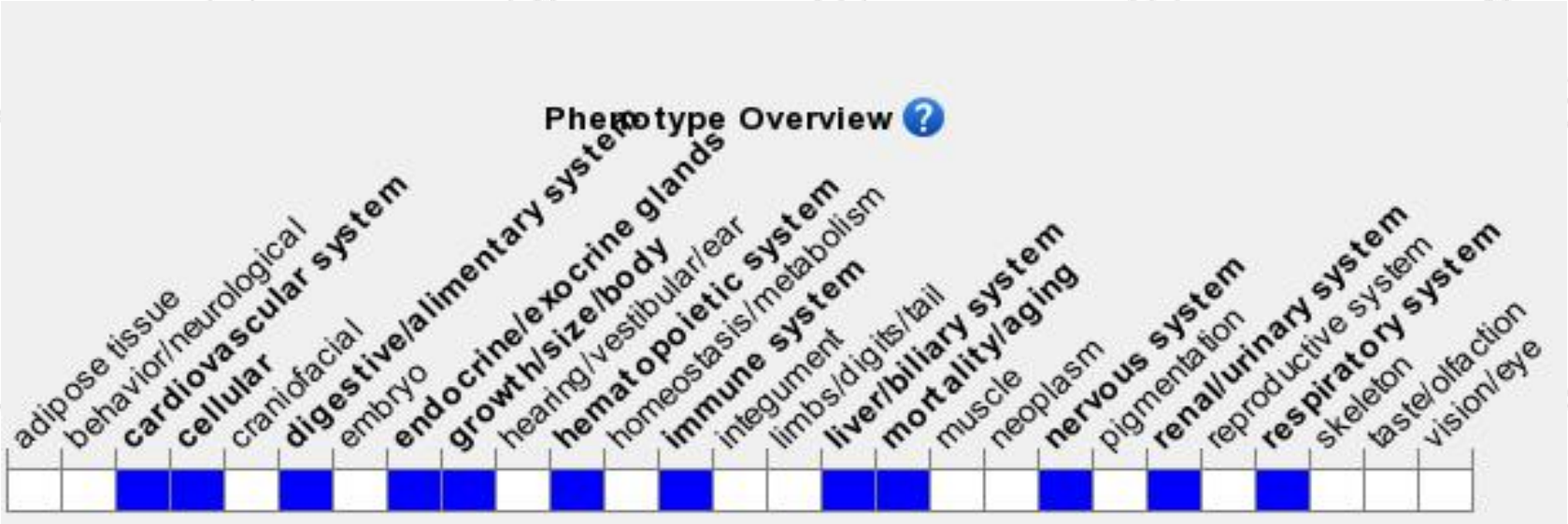


Genomic location distribution



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/).

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If you have any questions, you are welcome to inquire.

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