

# Phldb3 Cas9-CKO Strategy

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# **Project Overview**

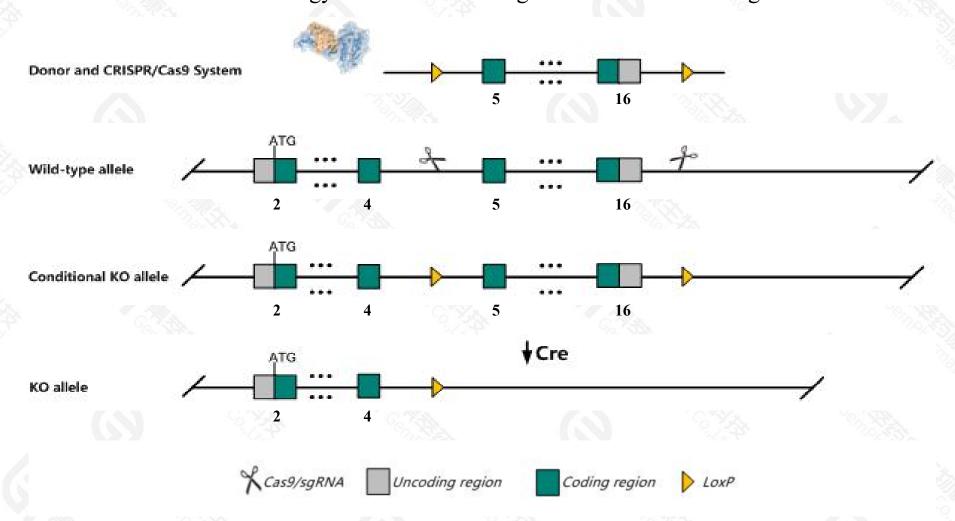


Project Name	Phldb3			
Project type	Cas9-CKO			
Strain background	C57BL/6JGpt			

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Phldb3* gene. The schematic diagram is as follows:



### **Technical routes**



- ➤ The *Phldb3* gene has 4 transcripts. According to the structure of *Phldb3* gene, exon5-exon16 of *Phldb3*-204(ENSMUST00000206422.2) transcript is recommended as the knockout region. The region contains 1396bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Phldb3* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor was constructed.Cas9, sgRNA 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 was 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**



- > The *Phldb3* gene is located on the Chr7. 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)



#### Phidb3 pleckstrin homology like domain, family B, member 3 [Mus musculus (house mouse)]

Gene ID: 232970, updated on 17-Dec-2020

#### Summary



Official Symbol Phldb3 provided by MGI

Official Full Name pleckstrin homology like domain, family B, member 3 provided by MGI

Primary source MGI:MGI:3642959

See related Ensembl:ENSMUSG00000074277

Gene type protein coding
RefSeq status PROVISIONAL
Organism Mus musculus

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

Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as EG232970, Gm10102

Expression Broad expression in placenta adult (RPKM 3.5), stomach adult (RPKM 3.2) and 20 other tissuesSee more

Orthologs <u>human all</u>

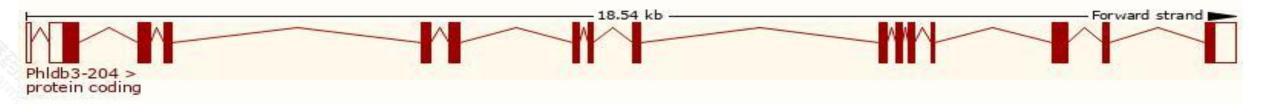
## Transcript information (Ensembl)



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

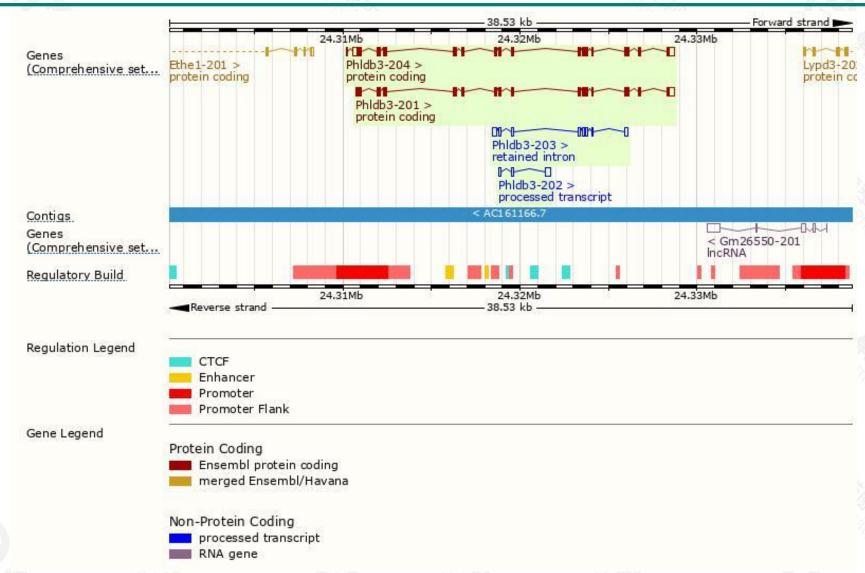
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Phldb3-204	ENSMUST00000206422.2	2564	<u>648aa</u>	Protein coding	CCDS52139		TSL:5 , GENCODE basic , APPRIS P1 ,
Phldb3-201	ENSMUST00000073325.6	2273	<u>648aa</u>	Protein coding	CCDS52139		TSL:5, GENCODE basic, APPRIS P1,
Phldb3-202	ENSMUST00000205330.2	474	No protein	Processed transcript	-		TSL:5,
Phldb3-203	ENSMUST00000205857.2	963	No protein	Retained intron			TSL:1,

The strategy is based on the design of *Phldb3-204* transcript, the transcription is shown below:



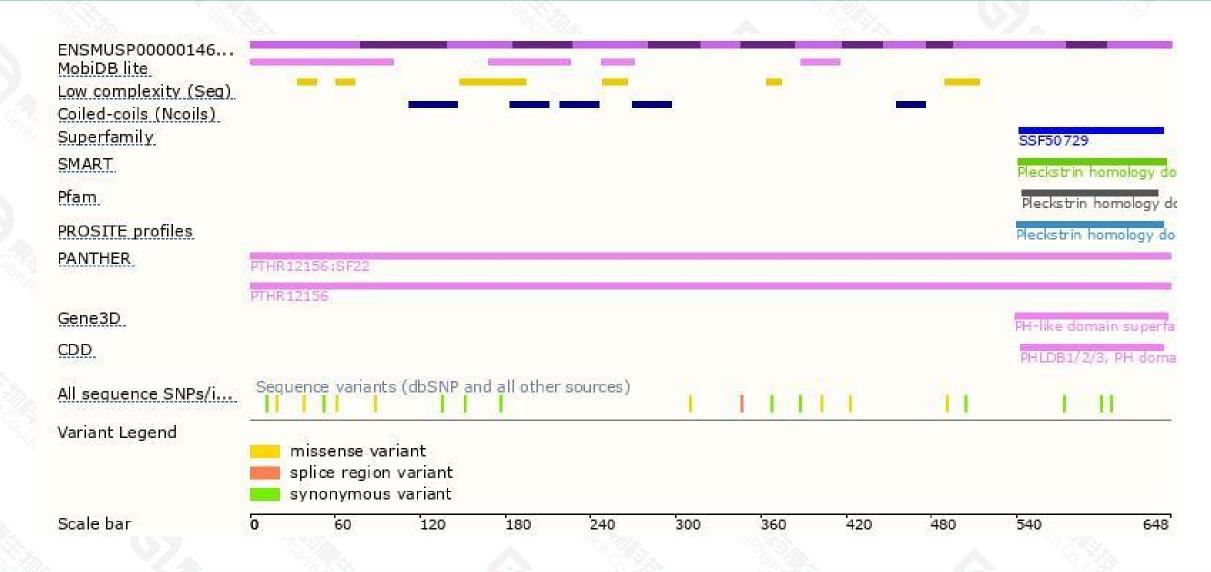
### Genomic location distribution





### Protein domain







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

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