

# Plekhb2 Cas9-CKO Strategy

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# **Project Overview**



Project Name Plekhb2

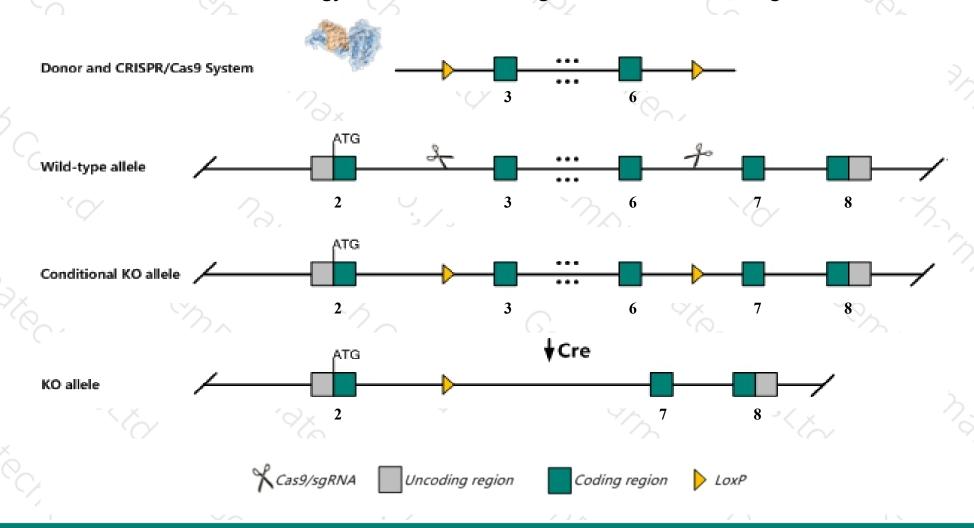
Project type Cas9-CKO

Strain background C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Plekhb2* gene. The schematic diagram is as follows:



#### **Technical routes**



- ➤ The *Plekhb2* gene has 4 transcripts. According to the structure of *Plekhb2* gene, exon3-exon6 of *Plekhb2*-201(ENSMUST00000027297.10) transcript is recommended as the knockout region. The region contains 386bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Plekhb2* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector 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 *Plekhb2* 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)



#### Plekhb2 pleckstrin homology domain containing, family B (evectins) member 2 [Mus musculus (house mouse)]

Gene ID: 226971, updated on 13-Mar-2020

#### Summary



Official Symbol Plekhb2 provided by MGI

Official Full Name pleckstrin homology domain containing, family B (evectins) member 2 provided by MGI

Primary source MGI:MGI:2385825

See related Ensembl:ENSMUSG00000026123

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 2310009M15Rik, AU018239, Evt2, Phdc, evt-2

Expression Broad expression in colon adult (RPKM 50.2), large intestine adult (RPKM 42.4) and 23 other tissuesSee more

Orthologs <u>human</u> all

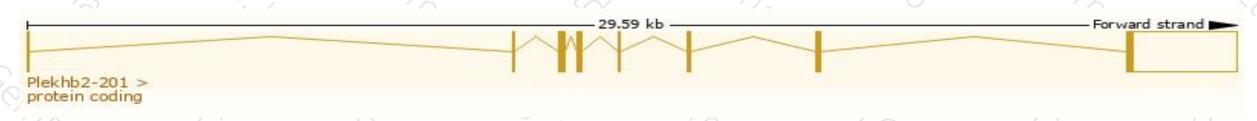
# Transcript information (Ensembl)



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

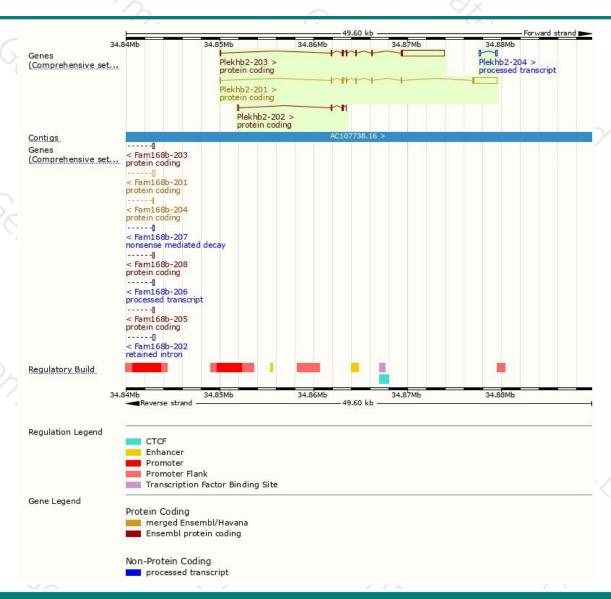
Name Transcript ID bp Protein Biotype CCDS	UniProt	Flags
		. lags
Plekhb2-203         ENSMUST00000156687.7         5176         204aa         Protein coding         CCDS78558	Q3UFC9 Q9QZC7	TSL:1 GENCODE basic
Plekhb2-201         ENSMUST000000027297.10         3279         221aa         Protein coding         CCDS14874	Q9QZC7	TSL:1 GENCODE basic APPRIS P1
Plekhb2-202         ENSMUST00000152654.2         344         65aa         Protein coding         -	D3Z3L2	CDS 3' incomplete TSL:5
Plekhb2-204 ENSMUST00000191722.1 302 No protein Processed transcript -	-	TSL:5

The strategy is based on the design of *Plekhb2-201* transcript, the transcription is shown below:



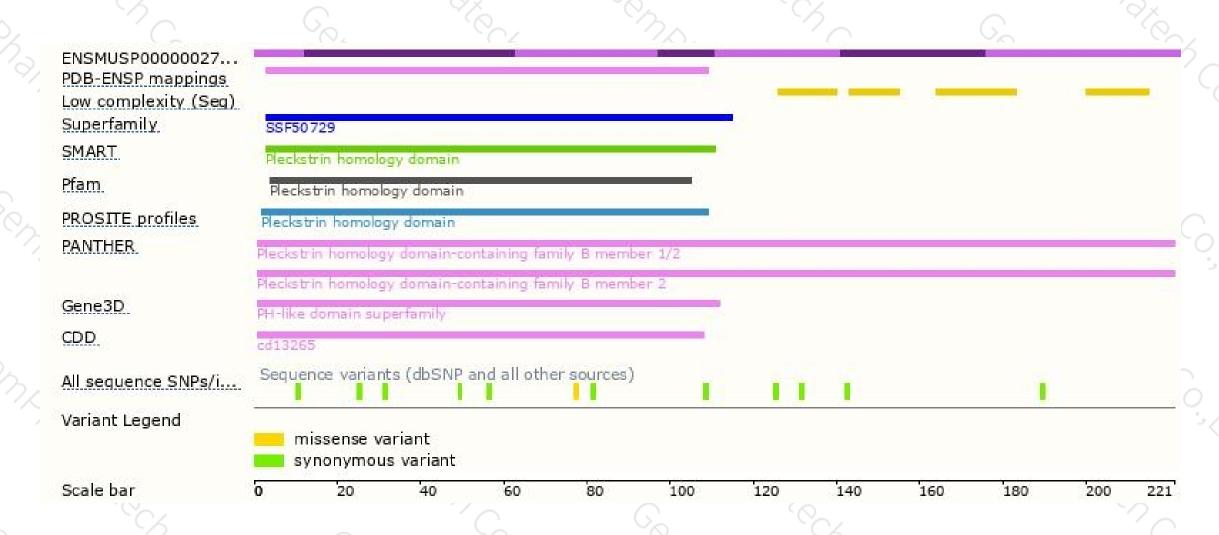
#### Genomic location distribution





#### Protein domain







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

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