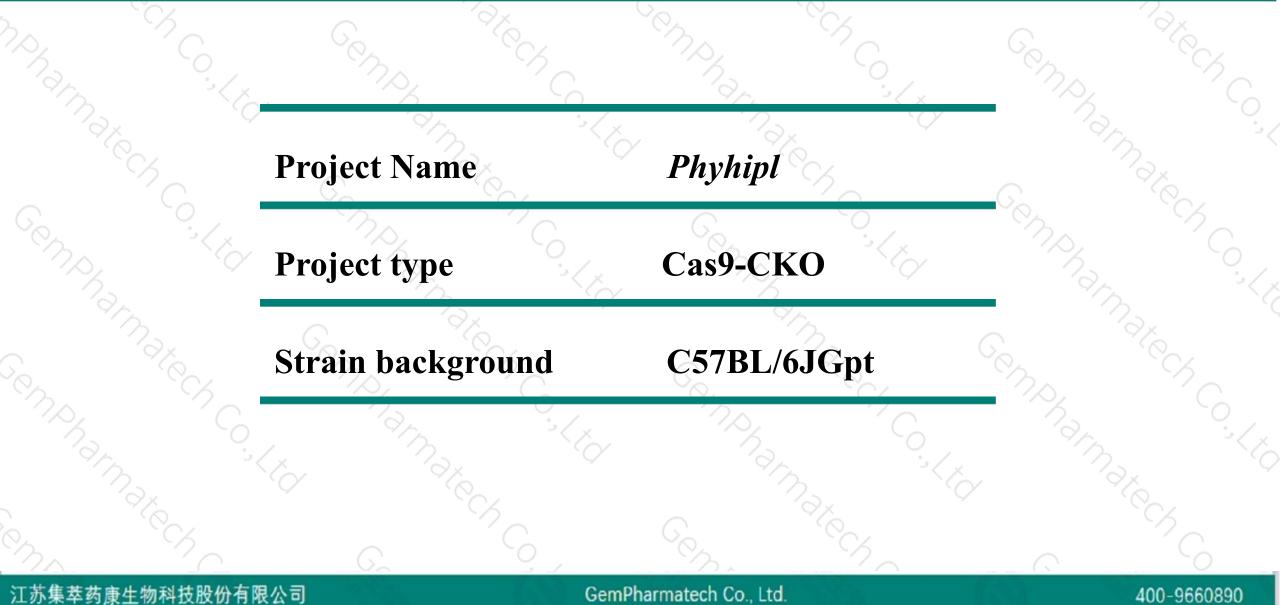


# **Phyhipl Cas9-CKO Strategy**

Designer: Reviewer: Design Date: JiaYu Xiaojing Li 2019-8-28

## **Project Overview**

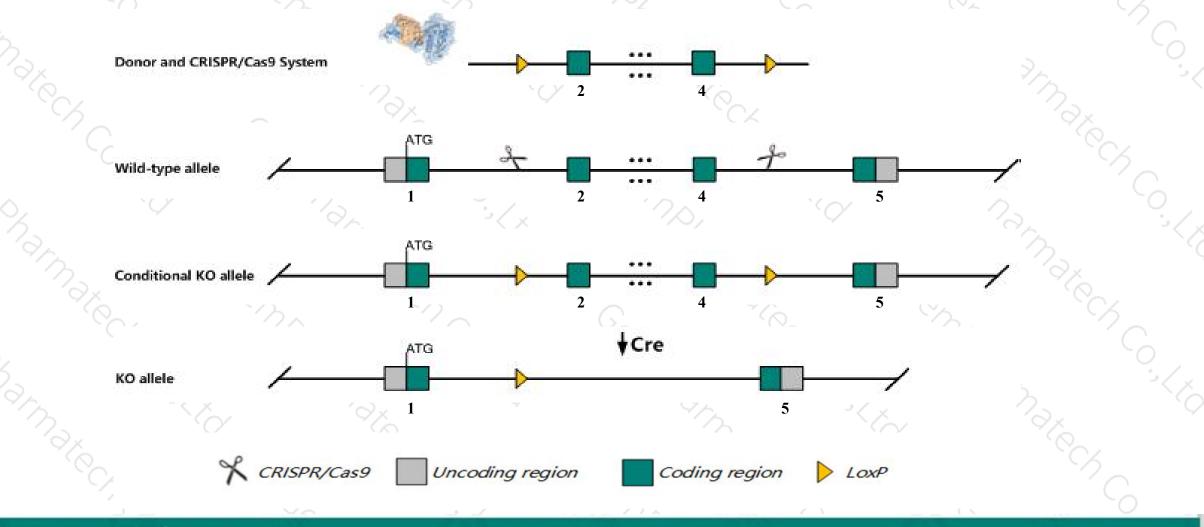




### **Conditional Knockout strategy**



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



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The Phyhipl gene has 10 transcripts. According to the structure of Phyhipl gene, exon2-exon4 of Phyhipl-201 (ENSMUST00000046513.9) transcript is recommended as the knockout region. The region contains 490bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Phyhipl* 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.



- The *Phyhipl* gene is located on the Chr10. 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)**



\$ ?

#### Phyhipl phytanoyl-CoA hydroxylase interacting protein-like [Mus musculus (house mouse)]

Gene ID: 70911, updated on 7-Apr-2019

#### Summary

| Official Symbol       | Phyhipl provided by MGI  |
|-----------------------|--|
| Official Full Name    | phytanoyI-CoA hydroxylase interacting protein-like provided byMGI  |
| <b>Primary source</b> | MGI:MGI:1918161  |
| See related           | Ensembl:ENSMUSG00000037747   |
| Gene type             | protein coding   |
| <b>RefSeq status</b>  | 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         | 4921522K17Rik, Al267048  |
| Expression            | Biased expression in testis adult (RPKM 75.5), cerebellum adult (RPKM 31.7) and 6 other tissues See more                             |
| Orthologs             | human all  |

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## **Transcript information (Ensembl)**



#### The gene has 10 transcripts, all transcripts are shown below:

| 1 10        | N.C.                 |      |              |                      |           | and and a |   |
|-------------|----------------------|------|--------------|----------------------|-----------|-----------|---|
| Name        | Transcript ID        | bp   | Protein      | Biotype              | CCDS      | UniProt   | Flags   |
| Phyhipl-201 | ENSMUST0000046513.9  | 2877 | <u>375aa</u> | Protein coding       | CCDS23914 | Q8BGT8    | TSL:1 GENCODE basic APPRIS P3   |
| Phyhipl-206 | ENSMUST00000162251.7 | 1540 | <u>330aa</u> | Protein coding       | CCDS48593 | F7D3N3    | TSL:3 GENCODE basic APPRIS ALT1   |
| Phyhipl-205 | ENSMUST00000162144.1 | 635  | <u>212aa</u> | Protein coding       | -         | F6U6Z2    | 5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3 |
| Phyhipl-209 | ENSMUST00000162793.7 | 598  | <u>41aa</u>  | Protein coding       | -         | E0CXW9    | CDS 3' incomplete TSL:3   |
| Phyhipl-208 | ENSMUST00000162571.7 | 2011 | No protein   | Processed transcript | -         | 1211      | TSL:1   |
| Phyhipl-204 | ENSMUST00000161687.7 | 1665 | No protein   | Processed transcript | -         |           | TSL:1   |
| Phyhipl-203 | ENSMUST00000160127.1 | 359  | No protein   | Processed transcript | 2         | 640       | TSL:5   |
| Phyhipl-202 | ENSMUST00000159025.1 | 326  | No protein   | Processed transcript | -         | 328       | TSL:3   |
| Phyhipl-210 | ENSMUST00000163054.1 | 3485 | No protein   | Retained intron      | -         | 1271      | TSL:1   |
| Phyhipl-207 | ENSMUST00000162470.1 | 1877 | No protein   | Retained intron      | -         | 1943      | TSL:1   |

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

#### < Phyhipl-201 protein coding

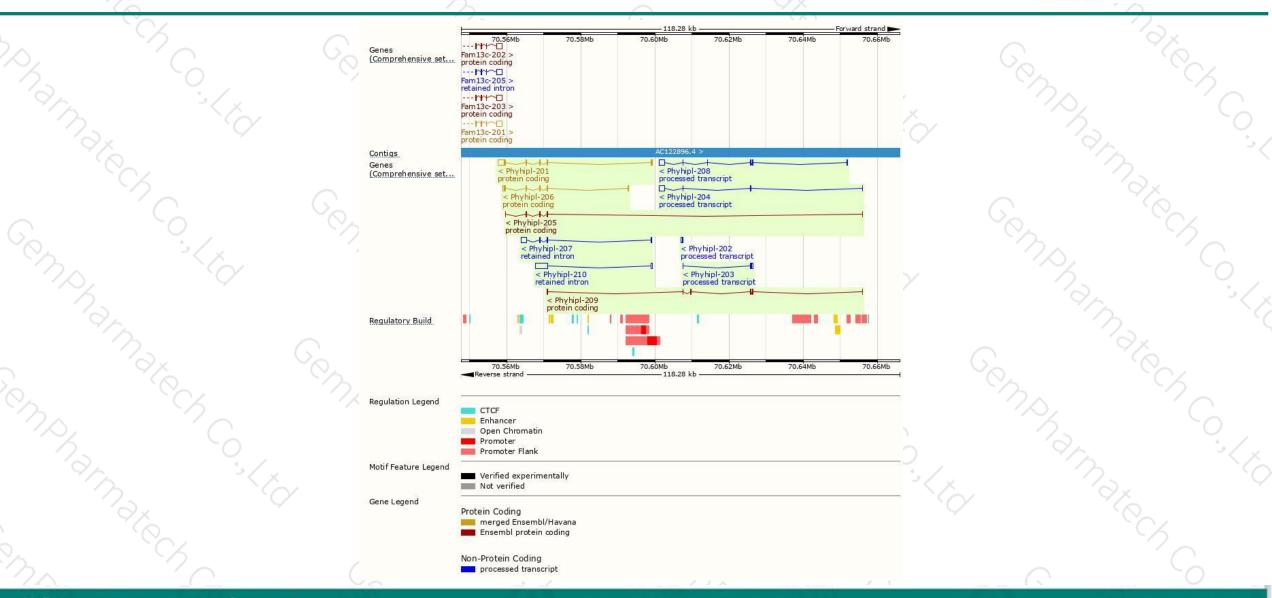
Reverse strand

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41.61 kb

### **Genomic location distribution**



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400-9660890

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## $\frac{\sqrt{2}}{\sqrt{2}}$

**Protein domain** 



ENSMUSP00000045... Low complexity (Seq) Conserved Domains hmmpanther PTHR15698 PTHR15698;SF8 Superfamily domains Fibronectin type III superfamily Pfam domain Fibronectin type III PROSITE profiles Fibronectin type III Gene3D Immunoglobulin-like fold Sequence variants (dbSNP and all other sources) All sequence SNPs/i... Variant Legend synonymous variant Scale bar 40 80 120 160 200 240 280 320 375

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



