

# Lhfpl3 Cas9-CKO Strategy

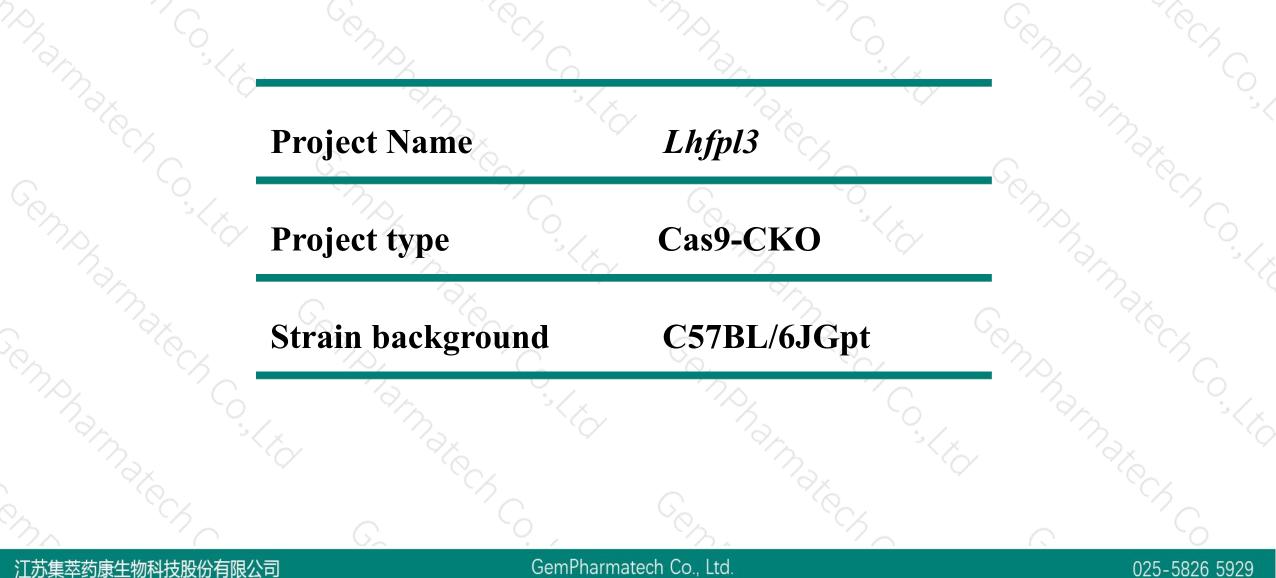
Designer: Miaomiao Cui

**Reviewer: Shilei Zhu** 

Design Date: 2020-12-8

## **Project Overview**





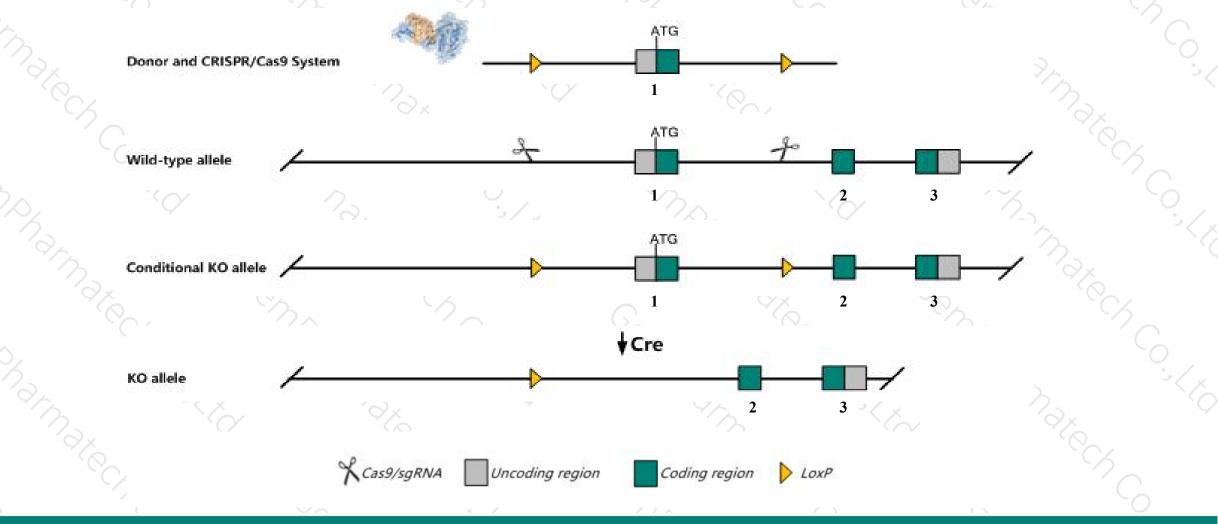
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### **Conditional Knockout strategy**



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This model will use CRISPR/Cas9 technology to edit the *Lhfpl3* gene. The schematic diagram is as follows:



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The Lhfpl3 gene has 2 transcripts. According to the structure of Lhfpl3 gene, exon1 of Lhfpl3-202(ENSMUST00000197992.1) transcript is recommended as the knockout region. The region contains start codon ATG.Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Lhfpl3* 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.



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- > The *Lhfpl3* 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.
- ≻Transcript *Lhfpl3*-201 will not be affected.
- ➤The KO region contains functional region of the A930003O13Rik gene.Knockout the region may affect the function of A930003O13Rik gene.
- The flox region is in the intron of the 6030443J06Rik gene, which may affect the regulation of this gene.
  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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#### Lhfpl3 lipoma HMGIC fusion partner-like 3 [Mus musculus (house mouse)]

Gene ID: 269629, updated on 13-Mar-2020

#### Summary

Official Symbol	Lhfpl3 provided by MGI
<b>Official Full Name</b>	lipoma HMGIC fusion partner-like 3 provided by MGI
<b>Primary source</b>	MGI:MGI:1925076
See related	Ensembl:ENSMUSG00000106379
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	A930031L14Rik
Expression	Biased expression in frontal lobe adult (RPKM 9.6), cerebellum adult (RPKM 7.7) and 5 other tissuesSee more
Orthologs	human all



# **Transcript information (Ensembl)**



The gene has 2 transcripts, all transcripts are shown below:

Name Transcript ID		bp	Protein	Biotype	CCDS	UniProt	Flags		
Lhfpl3-202	ENSMUST00000197992.1	3115	<u>228aa</u>	Protein coding	CCD580218	A0A0G2JG13	TSL:1 GENCODE basic APPRIS P1		
Lhfpl3-201	ENSMUST00000196406.1	2589	<u>80aa</u>	Protein coding	-	A0A0G2JEN1	TSL:1 GENCODE basic		

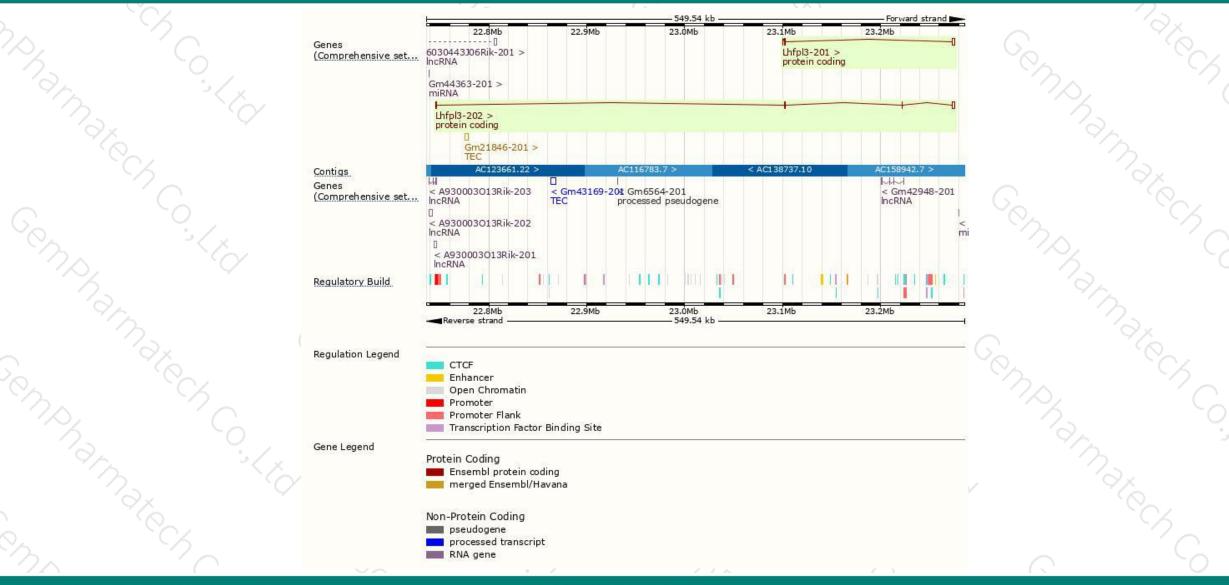
The strategy is based on the design of *Lhfpl3-202* transcript, the transcription is shown below:

Lhfpl3-202			529.	54 kb		For	ward strand
protein co	ding	<u> </u>	· /	12D	10	62	0
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### **Genomic location distribution**



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## **Protein domain**



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	ENSMUSP00000143 Transmembrane heli Low complexity (Seg) Pfam		-	Lipoma HMG	IC fusion r	artner-like	• protein							
20	PANTHER.			IC fusion pa									-	
	Gene3D			0,140,150									_	
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If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



