

# **Ppih Cas9-CKO Strategy**

**Designer:** 

**Reviewer:** 

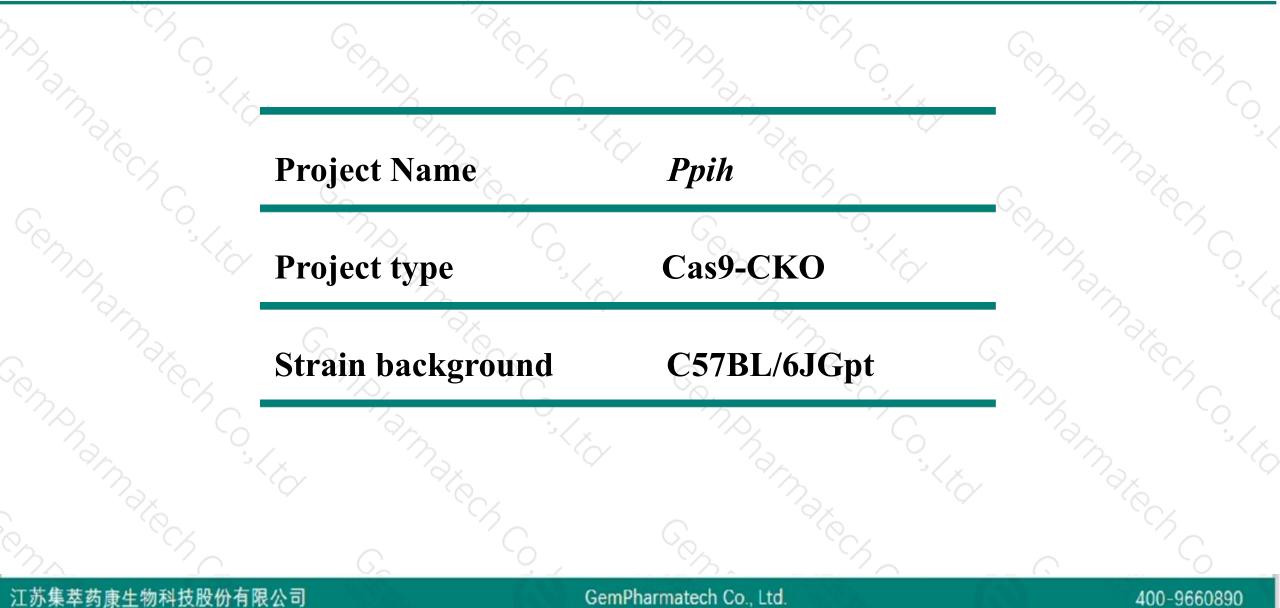
**Design Date:** 

Xueting Zhang Yanhua Shen

2020-4-15

# **Project Overview**

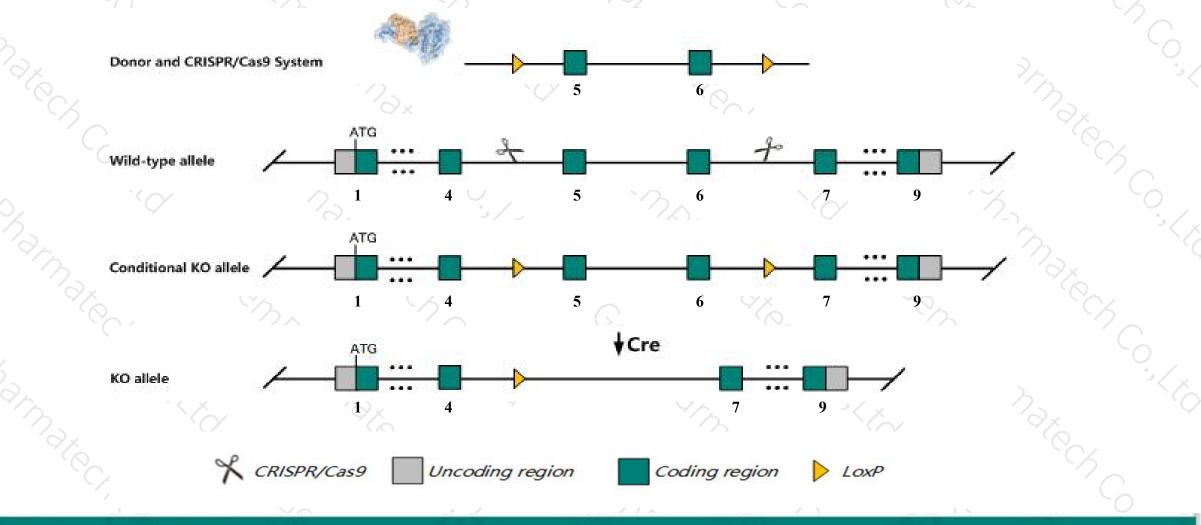




### **Conditional Knockout strategy**



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



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The *Ppih* gene has 6 transcripts. According to the structure of *Ppih* gene, exon5-exon6 of *Ppih-202* (ENSMUST00000106317.1) transcript is recommended as the knockout region. The region contains 136bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Ppih* 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 N-terminal of *Ppih* gene will remain several amino acids ,it may remain the partial function of *Ppih* gene.
- The *Ppih* gene is located on the Chr4. 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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### Ppih peptidyl prolyl isomerase H [Mus musculus (house mouse)]

Gene ID: 66101, updated on 13-Mar-2020

#### Summary

Official SymbolPpih provided by MGIOfficial Full Namepeptidyl prolyl isomerase H provided by MGIPrimary sourceMGI:MGI:106499See relatedEnsembl:ENSMUSG0000060288Gene typeprotein codingRefSeq statusVALIDATEDOrganismMus musculusLineageEukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;<br/>Muroidea; Murinae; Mus; MusAlso knownas110001J08Rik, 2010111B15Rik, 4833408F11Rik, Al464484, CYP-20, CYPH, D4Wsu43eOrthologhuman all

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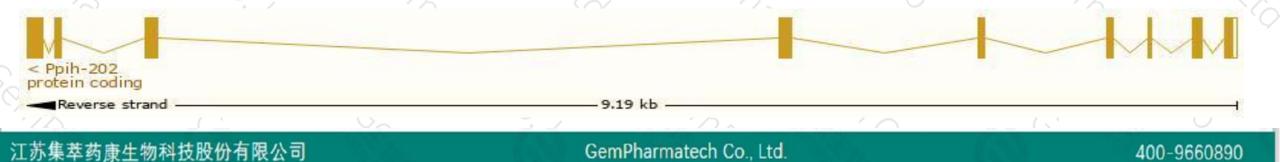
# **Transcript information (Ensembl)**



### The gene has 6 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ppih-205	ENSMUST00000106321.8	1288	<u>177aa</u>	Protein coding	CCDS18580	Q4G0C5 Q9D868	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Ppih-203	ENSMUST00000106318.7	800	<u>177aa</u>	Protein coding	CCDS18580	Q4G0C5 Q9D868	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Ppih-201	ENSMUST00000056458.13	757	<u>177aa</u>	Protein coding	CCDS18580	Q4G0C5 Q9D868	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Ppih-202	ENSMUST00000106317.1	613	<u>188aa</u>	Protein coding	CCDS51288	<u>Q9D868</u>	TSL:1 GENCODE basic
Ppih-206	ENSMUST00000147077.7	725	<u>158aa</u>	Protein coding	151	A2BGI8	CDS 5' incomplete TSL:5
Ppih-204	ENSMUST00000106319.7	622	<u>134aa</u>	Protein coding	143	A2BGI9	TSL:3 GENCODE basic

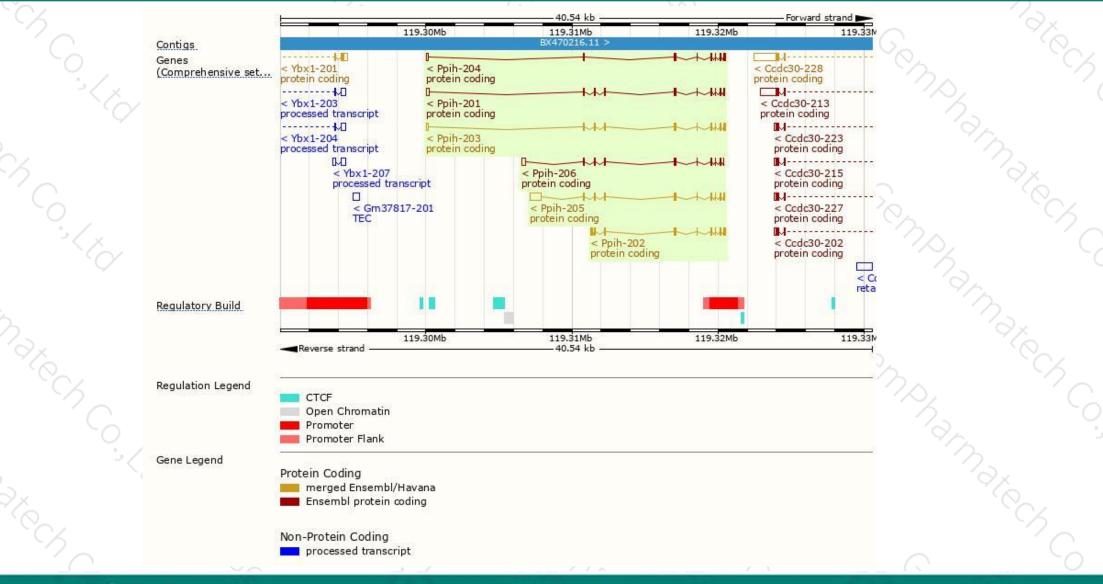
The strategy is based on the design of *Ppih-202* transcript, the transcription is shown below



### **Genomic location distribution**



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### **Protein domain**



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	<u>Pfam</u>	Cyclop	hilin-type pepti	dyl-prolyl cis-	trans isomerase	domain					
	PROSITE profiles	Cyclophilin	-type peptidyl-p	rolyl cis-trans	isomerase doma	iin.					
	PROSITE patterns			Cycloph	ilin-type peptidyl	-prolyl cis-tra	ns isomerase,	conserved sit			
	PANTHER	PTHR11071									
		PTHR11071:SF443									9. /
	Gene3D	Oyclophilin-like dom	ain superfamily								
	CDD	cd01926									
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If you have any questions, you are welcome to inquire. Tel: 400-9660890



