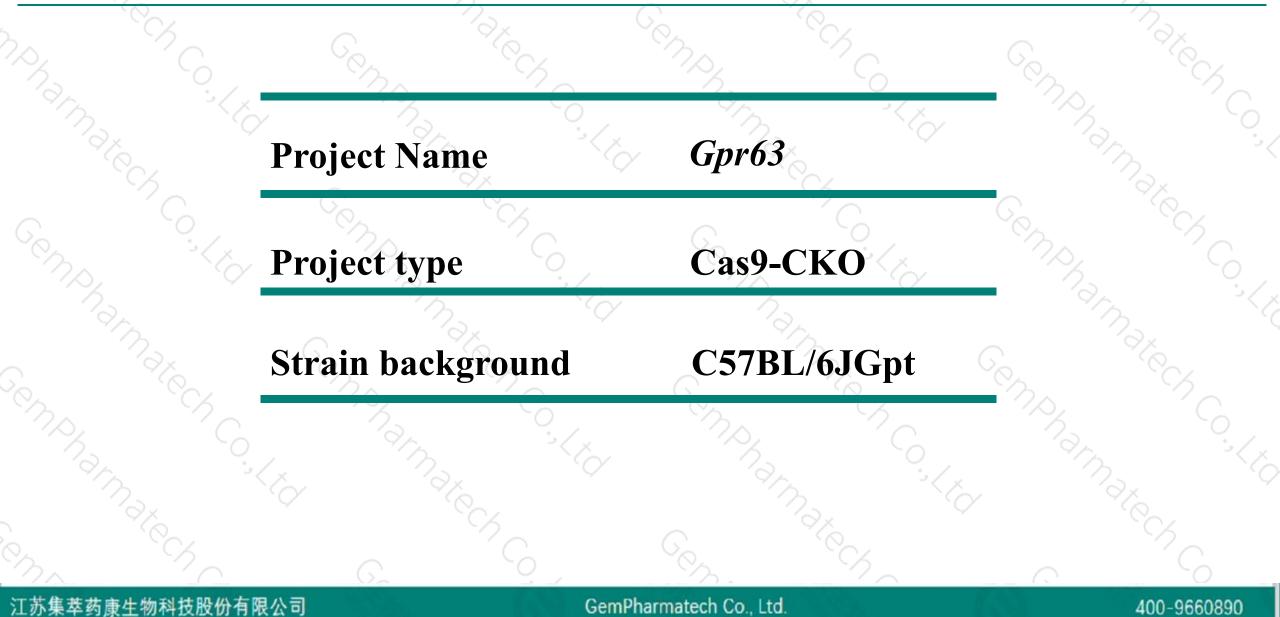
# Gpr63 Cas9-CKO Strategy

Designer: Design Date: Jinling Wang 2019-7-22

# **Project Overview**

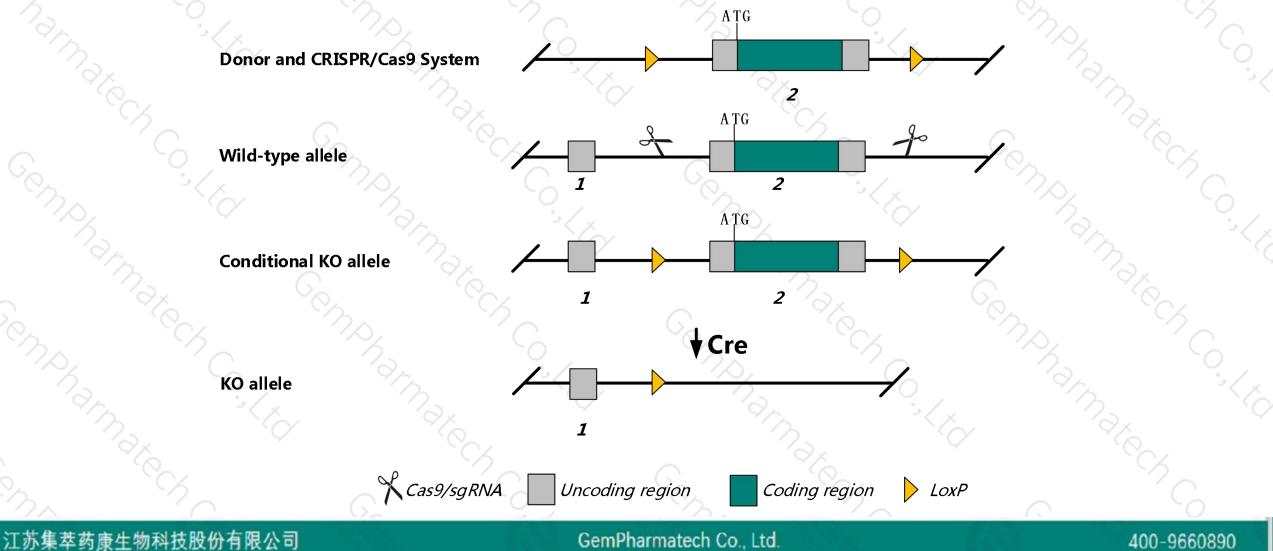




# **Conditional Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Gpr63* gene. The schematic diagram is as follows:





- The *Gpr63* gene has 2 transcript.According to the structure of *Gpr63* gene, exon2 of *Gpr63*-201 (ENSMUST00000038920.1) transcript is recommended as the knockout region.The region contains all coding sequence.Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Gpr63* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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 or cell types.

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- The Gpr63 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 the loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Notice

# Gene information (NCBI)



### Gpr63 G protein-coupled receptor 63 [ Mus musculus (house mouse) ]

Gene ID: 81006, updated on 8-Dec-2018

#### Summary

Official Symbol Gpr63 provided by MGI Official Full Name G protein-coupled receptor 63 provided by MGI Primary source MGI:MGI:2135884 See related Ensembl:ENSMUSG0000040372 Gene type protein coding **RefSeq status** PROVISIONAL Organism Mus musculus Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Lineage Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus Also known as Psp24-2; PSP24beta

Expression Biased expression in cerebellum adult (RPKM 4.1), CNS E18 (RPKM 1.2) and 14 other tissues See more

Orthologs human all

### \* ?

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### **Transcript information (Ensembl)**



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

Name 🍦	Transcript ID 👙	bp 🖕	Protein 🝦	Biotype 🖕	CCDS 🖕	UniProt 👙	Flags 🍦		
Gpr63-201	ENSMUST0000038920.1	2370	<u>425aa</u>	Protein coding	<u>CCDS18008</u> &	<u>Q9EQQ3</u> &	TSL:1	GENCODE basic	APPRIS P1
Gpr63-202	ENSMUST00000151006.1	389	No protein	IncRNA	-		TSL:3		

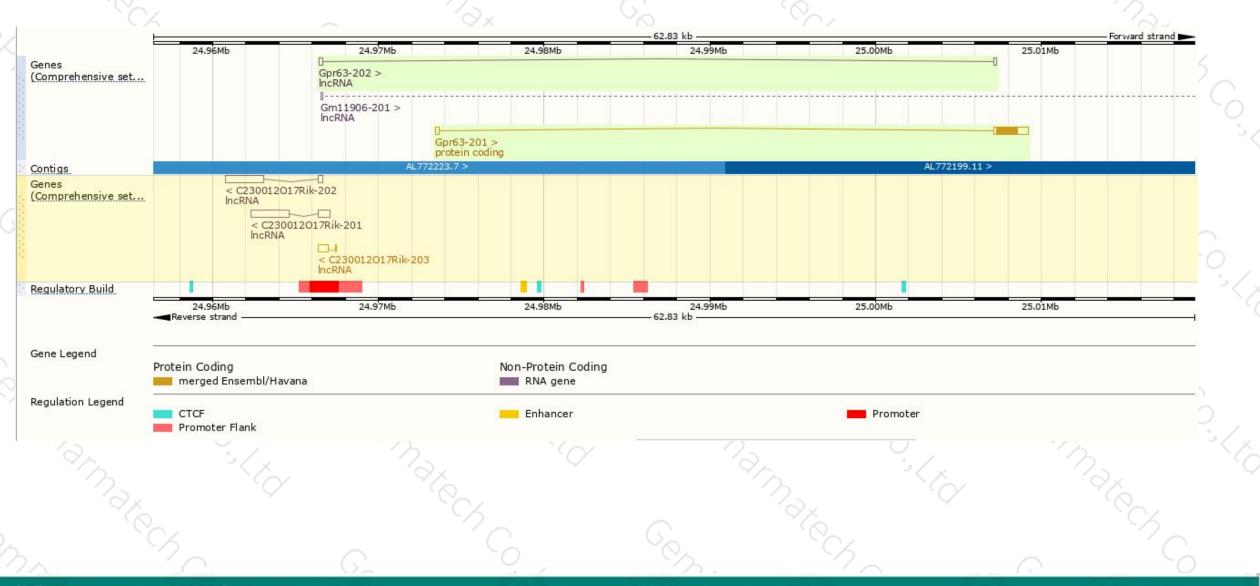
The strategy is based on the design of *Gpr63*-201transcript, The transcription is shown below

		3	5.81 kb			Forward strand
pr63-201 > otein coding	77.5					
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		~? C				

### **Genomic location distribution**



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# Protein domain



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Low complexity (Sec Conserved Domains										
hmmpanther	PTHR242	45								$\sim 0$
	PTHR242	45:SF1							124	
Superfamily domain	5.	SSF8132	1							
SMART domains			G protein-coupled n	eceptor, rhodopsin	like					
Prints domain			G protein-coupled rece	ptor, rhodopsin-like	-	_	_	_		
Pfam domain			G protein-coupled	d receptor, rhodops	in-like					
PROSITE profiles			GPCR, rhodopsin	-like, 7TM						9
Gene3D		1.20.10								
All sequence SNPs/i.	Sequence v	variants (dbSNP and	all other sources)	T	11.01	10.0		1	111	
Variant Legend										
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If you have any questions, you are welcome to inquire. Tel: 400-9660890



