

# Rab11fip2 Cas9-CKO Strategy

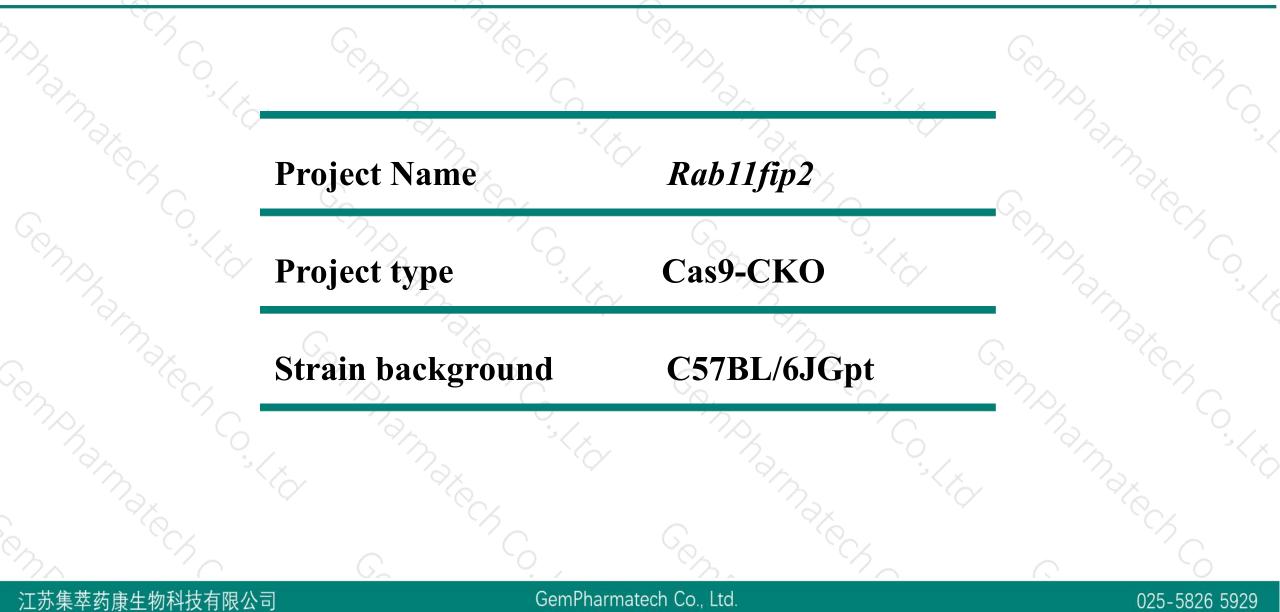
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**Reviewer: Daohua Xu** 

**Design Date: 2020-7-28** 

# **Project Overview**



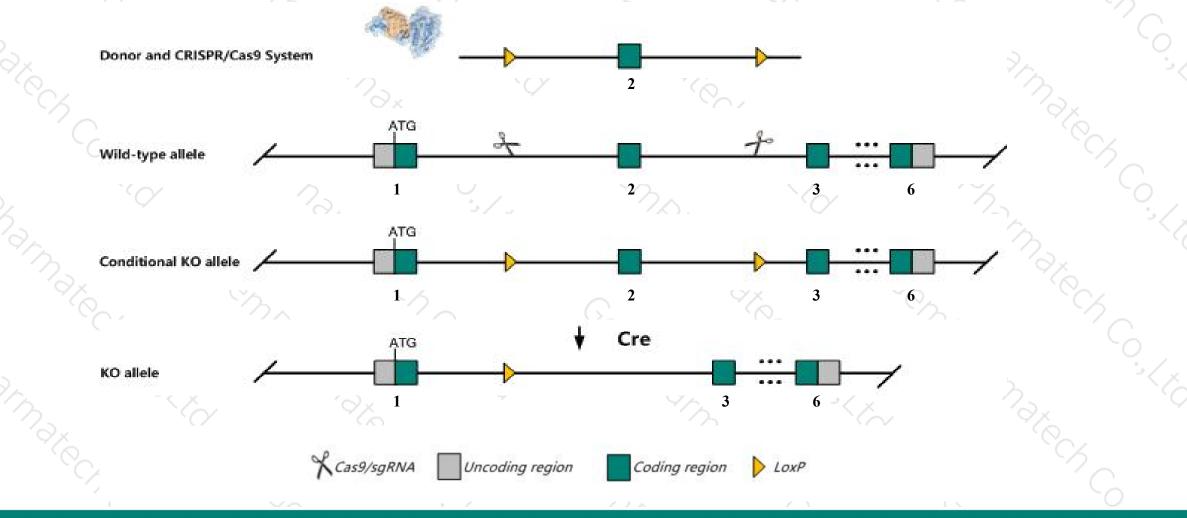


## **Conditional Knockout strategy**



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This model will use CRISPR/Cas9 technology to edit the *Rab11fip2* gene. The schematic diagram is as follows:



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The Rab11fip2 gene has 3 transcripts. According to the structure of Rab11fip2 gene, exon2 of Rab11fip2-201(ENSMUST00000051996.12) transcript is recommended as the knockout region. The region contains 443bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Rab11fip2* 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.



- > The *Rab11fip2* gene is located on the Chr19. 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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#### Rab11fip2 RAB11 family interacting protein 2 (class I) [Mus musculus (house mouse)]

Gene ID: 74998, updated on 13-Mar-2020

#### Summary

Official Symbol	Rab11fip2 provided by MGI
Official Full Name	RAB11 family interacting protein 2 (class I) provided by MGI
<b>Primary source</b>	MGI:MGI:1922248
See related	Ensembl:ENSMUSG0000040022
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	4930470G04Rik, A830046J09Rik, AW558126, Nrip11
Expression	Broad expression in cortex adult (RPKM 5.3), CNS E18 (RPKM 5.1) and 25 other tissuesSee more
Orthologs	human all

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# **Transcript information (Ensembl)**

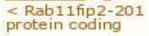


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The gene has 3 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rab11fip2-203	ENSMUST00000171986.7	5363	<u>370aa</u>	Protein coding	CCD550482	<u>G3UW57</u>	TSL:1 GENCODE basic
Rab11fip2-201	ENSMUST00000051996.12	4482	<u>529aa</u>	Protein coding	CCDS38033	<u>G3XA57</u>	TSL:1 GENCODE basic
Rab11fip2-202	ENSMUST00000170819.1	1539	<u>512aa</u>	Protein coding	2	<u>G3XA57</u>	TSL:5 GENCODE basic APPRIS P1

The strategy is based on the design of *Rab11fip2-201* transcript, the transcription is shown below:



Reverse strand -

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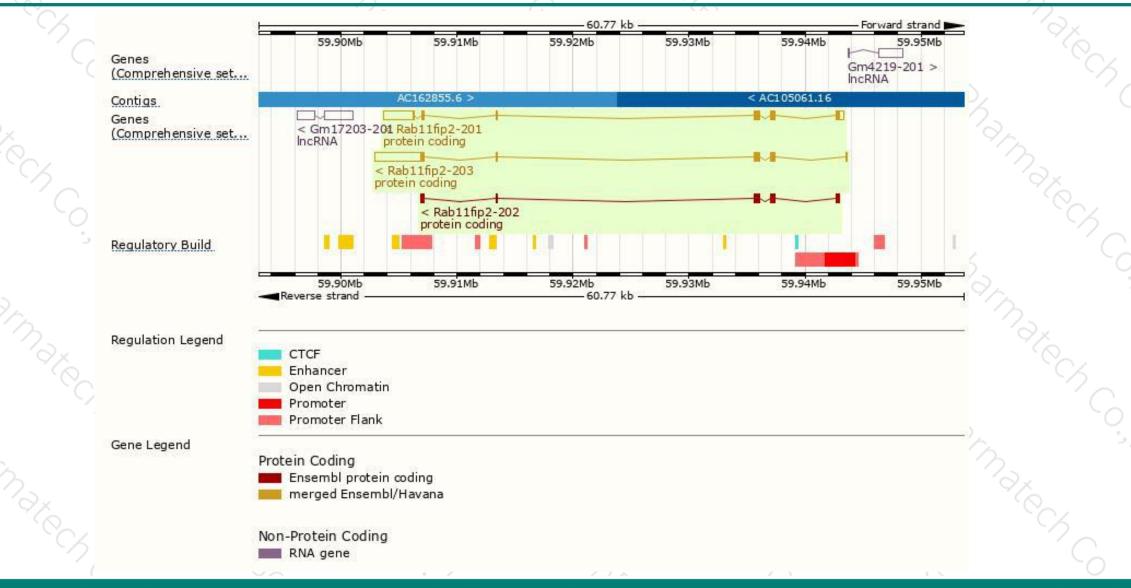
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39.67 kb

### **Genomic location distribution**



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## **Protein domain**



D B J B J B J B J B J B J B J B J B J B	ENSMUSP00000059 MobiDB lite Low complexity (Seg) Coiled-coils (Ncoils) Superfamily	SSF49562	FIP-R	IBD, C-term
	SMART Pfam	C2 domain C2 domain	Rab-bindi	ing domain
~°n	PROSITE profiles	C2 domain	Rab-binding o	domain FIP
	PANTHER	ab11-family interacting protein class I		
	Gene3D CDD	C2 domain superfamily cd08682		
	All sequence SNPs/i	Sequence variants (dbSNP and all other sources)		111 76
ò.	Variant Legend	missense variant synonymous variant		
~??\	Scale bar	60 120 180 240		529
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If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



