# Gpr18 Cas9-CKO Strategy

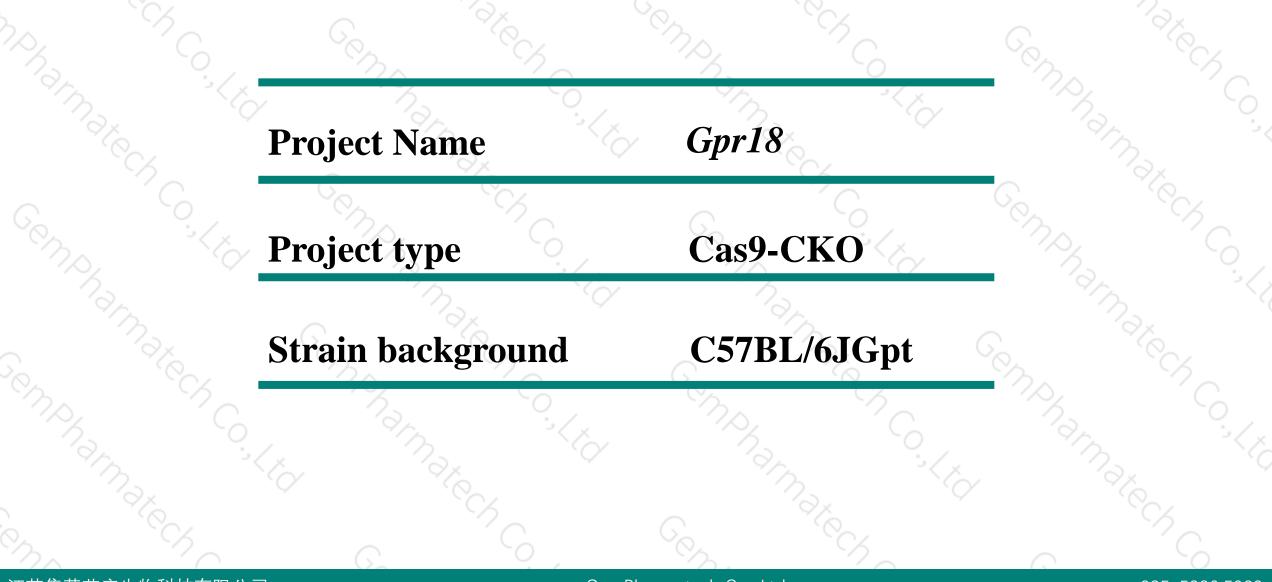
Designer: Reviewer :

**Design Date:** 

Daohua Xu Huimin Su 2019-11-26

### **Project Overview**



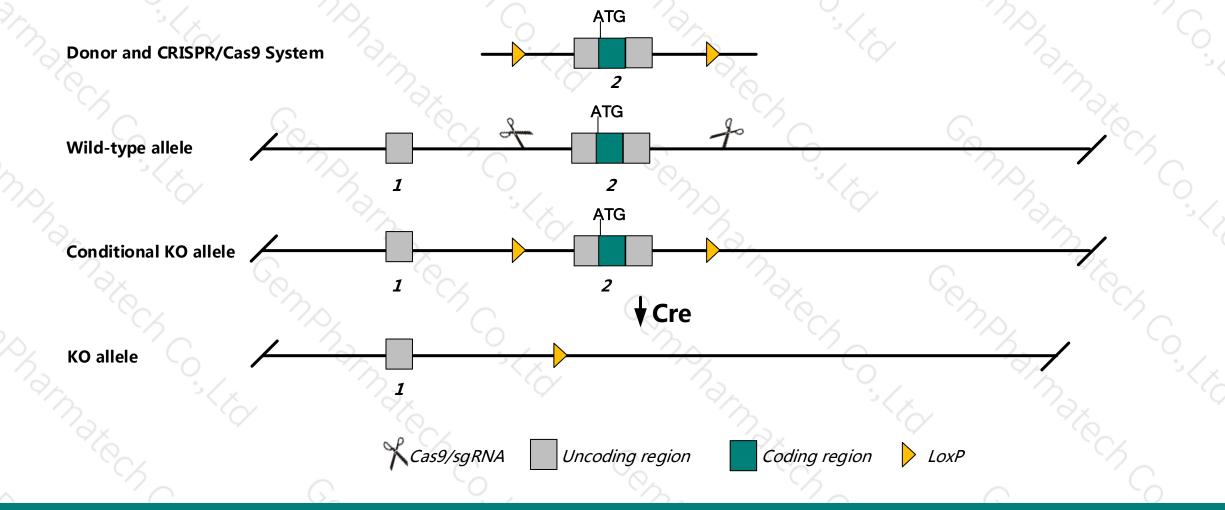


### **Conditional Knockout strategy**



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This model will use CRISPR/Cas9 technology to edit the Gpr18 gene. The schematic diagram is as follows:



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- The *Gpr18* gene has 1 transcript.According to the structure of *Gpr18* gene, exon2 of *Gpr18*-201 (ENSMUST00000055475.8) transcript is recommended as the knockout region.The region contains all of the coding sequence.Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Gpr18* 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 or cell types.

### Notice



- According to the existing MGI data, Mice homozygous for a knock-out allele exhibit a reduction in CD8-positive, alphabeta T cell number and CD8-positive, gamma-delta intraepithelial T cell number. Mice homozygous for a different knockout allele exhibit increased susceptibility to E. coli infection and diminished response to RvD2.
- > The KO region contains partial intron of the *Ubac2* gene.
- The Gpr18 gene is located on the Chr14. 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.

### Gene information (NCBI)

### Gpr18 G protein-coupled receptor 18 [ Mus musculus (house mouse) ]

Gene ID: 110168, updated on 16-Nov-2018

#### 🔺 Summary

Official SymbolGpr18 provided by MGIOfficial Full NameG protein-coupled receptor 18 provided by MGIPrimary sourceMGI:MGI:107859See relatedEnsembl:ENSMUSG0000050350Gene typeprotein codingRefSeq statusPROVISIONALOrganismMus musculusLineageEukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;<br/>Muroidea; Muridae; Murinae; Mus; MusExpressionBiased expression in spleen adult (RPKM 55.1), mammary gland adult (RPKM 20.1) and 4 other tissues See more<br/>human\_all

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## **Transcript information (Ensembl)**



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The gene has 1 transcript, and the transcript is shown below:

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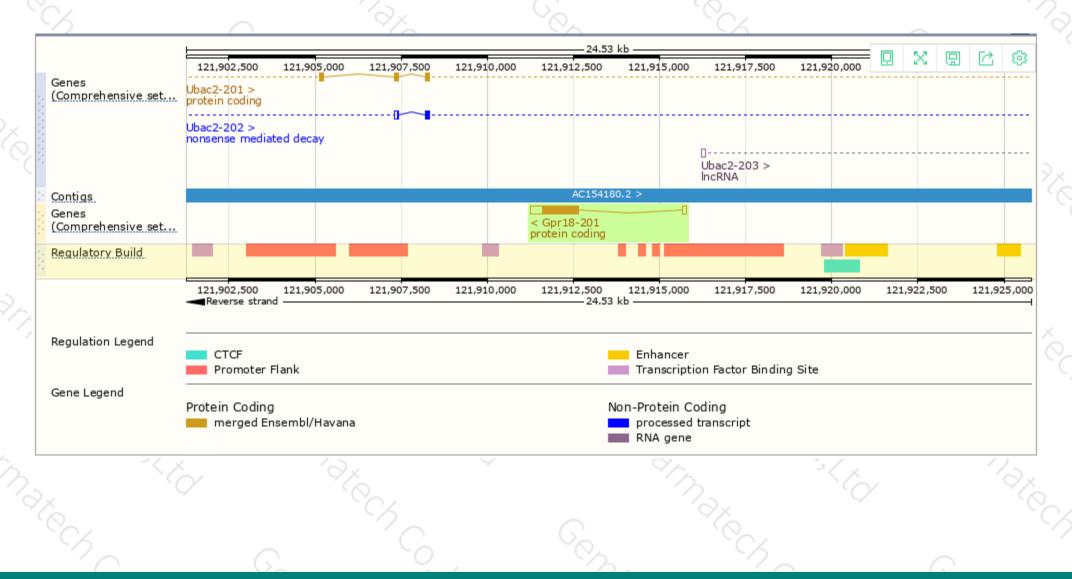
The strategy is based on the design of Gpr18-201 transcript, The transcription is shown below

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Reverse strand	4.53 k	b			
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### **Genomic location distribution**





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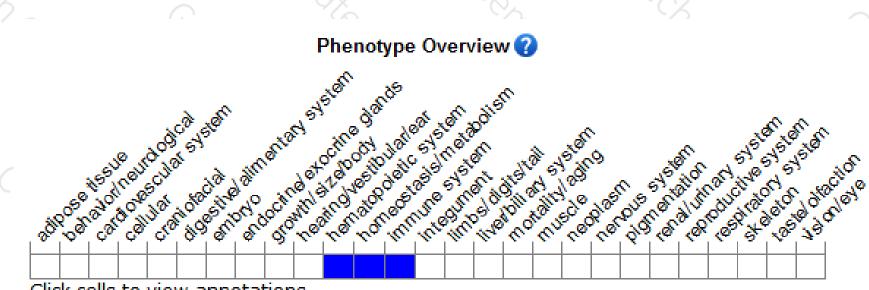
### **Protein domain**



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### Mouse phenotype description(MGI)





Click cells to view annotations.

Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(http://www.informatics.jax.org/).

According to the existing MGI data, Mice homozygous for a knock-out allele exhibit a reduction in CD8-positive, alpha-beta T cell number and CD8-positive, gamma-delta intraepithelial T cell number. Mice homozygous for a different knock-out allele exhibit increased susceptibility to E. coli infection and diminished response to RvD2.

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If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



