

# Illr1 Cas9-CKO Strategy Romphamakech Co. 1/4

QiongZhou

# **Project Overview**



Project Name Il1r1

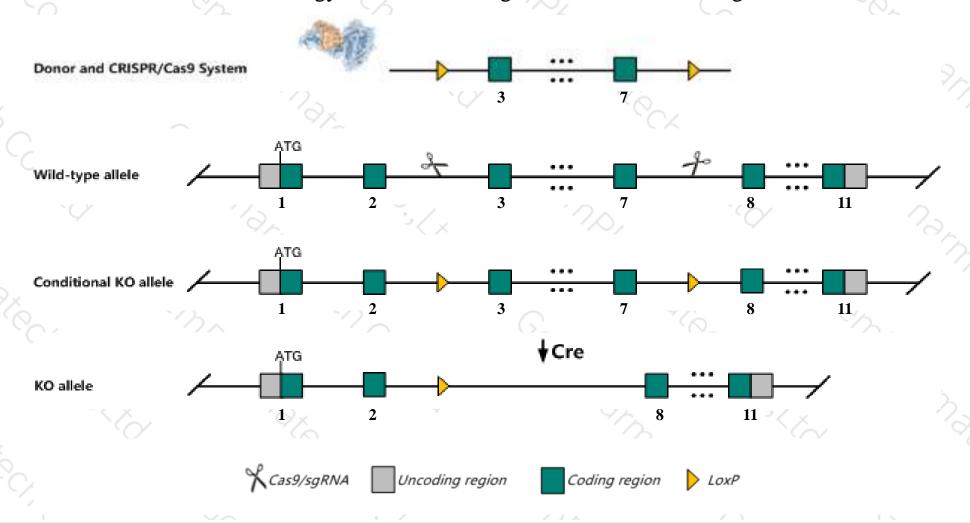
Project type Cas9-CKO

Strain background C57BL/6JGpt

# **Conditional Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Il1r1* gene. The schematic diagram is as follows:



## **Technical routes**



- ➤ The *Il1r1* gene has 3 transcripts. According to the structure of *Il1r1* gene, exon3-exon7 of *Il1r1-201*(ENSMUST00000027241.10) transcript is recommended as the knockout region. The region contains 781bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Il1r1* 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.

## **Notice**



- ➤ According to the existing MGI data, Mice homozygous for a knock-out allele exhibit increased susceptibility to bacterial infection, reduced IL1b responsiveness, delayed tooth eruption, decreased susceptibility to experimental autoimmune uveoritinitis, decreased susceptibility to kidney reperfusion injury, and late onset obesity.
- ➤ The *Il1r1* gene is located on the Chr1. 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)



#### Il1r1 interleukin 1 receptor, type I [Mus musculus (house mouse)]

Gene ID: 16177, updated on 9-Apr-2019

#### Summary

☆ ?

Official Symbol II1r1 provided by MGI

Official Full Name interleukin 1 receptor, type I provided by MGI

Primary source MGI:MGI:96545

See related Ensembl:ENSMUSG00000026072

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 CD121a, CD121b, IL-1R1, IL-iR, II1r-1

Expression Ubiquitous expression in liver E18 (RPKM 5.6), bladder adult (RPKM 4.5) and 27 other tissuesSee more

Orthologs <u>human</u> all

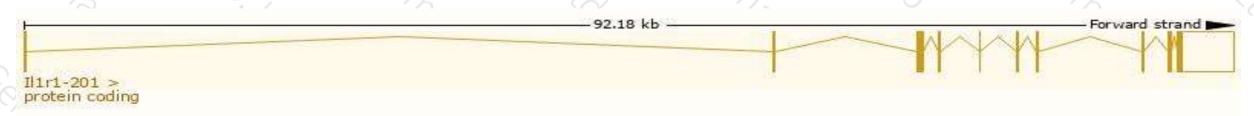
# Transcript information (Ensembl)



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

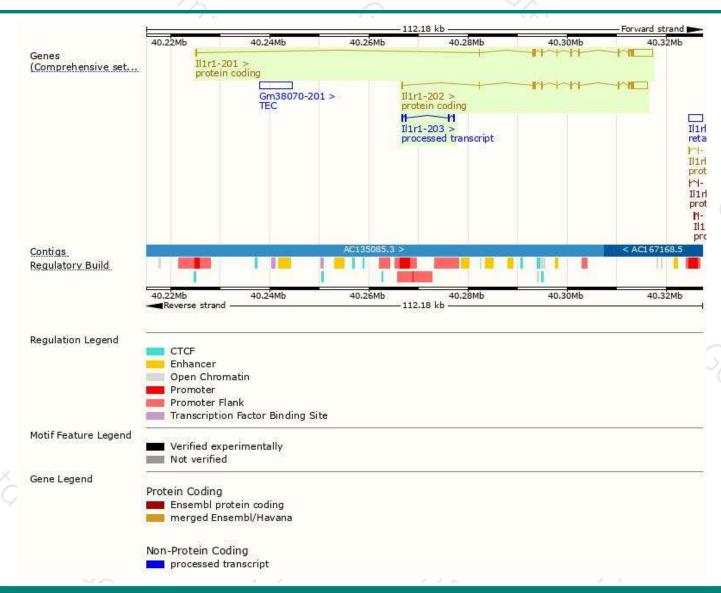
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
II1r1-201	ENSMUST00000027241.10	5819	<u>576aa</u>	Protein coding	CCDS35547	P13504 Q32MH0	TSL:1 GENCODE basic APPRIS P3
II1r1-202	ENSMUST00000114795.2	4832	<u>573aa</u>	Protein coding	CCDS48246	Q8C833	TSL:1 GENCODE basic APPRIS ALT2
II1r1-203	ENSMUST00000195402.1	473	No protein	Processed transcript	-	-	TSL:3

The strategy is based on the design of *Il1r1-201* transcript, The transcription is shown below



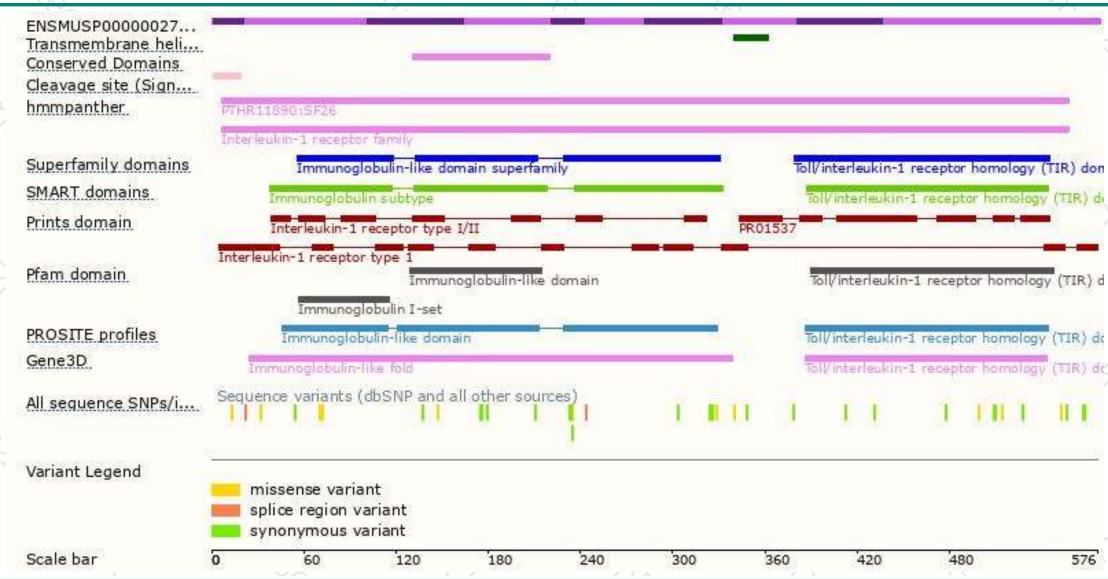
## Genomic location distribution





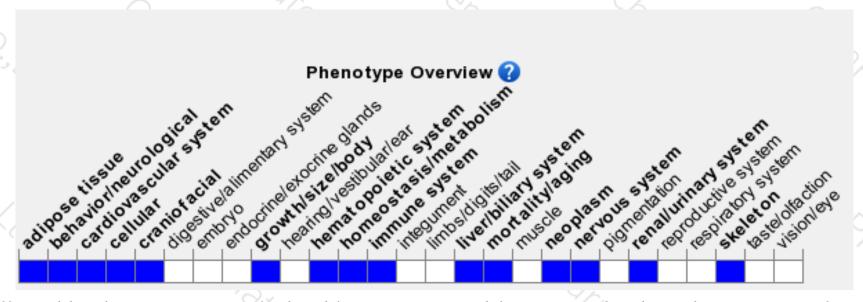
### Protein domain





# Mouse phenotype description(MGI)





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 increased susceptibility to bacterial infection, reduced IL1b responsiveness, delayed tooth eruption, decreased susceptibility to experimental autoimmune uveoritinitis, decreased susceptibility to kidney reperfusion injury, and late onset obesity.



If you have any questions, you are welcome to inquire.

Tel: 025-5864 1534





