

# Cryba4 Cas9-CKO Strategy

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# **Project Overview**



**Project Name** 

Cryba4

**Project type** 

Cas9-CKO

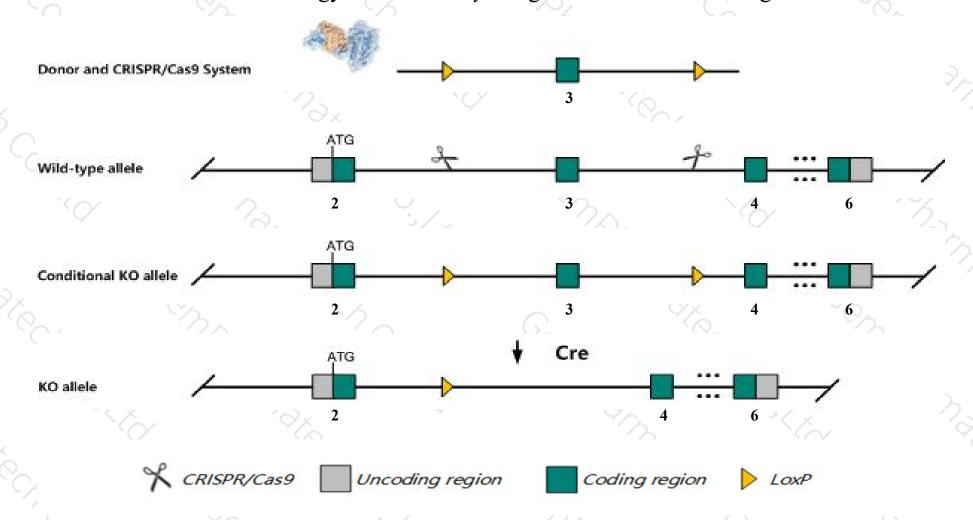
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Cryba4 gene. The schematic diagram is as follows:



### Technical routes



- The *Cryba4* gene has 3 transcripts. According to the structure of *Cryba4* gene, exon3 of *Cryba4-203* (ENSMUST00000112385.7) transcript is recommended as the knockout region. The region contains 119bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Cryba4* 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.

### **Notice**



- > The *Cryba4* gene is located on the Chr5. 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)



#### Cryba4 crystallin, beta A4 [Mus musculus (house mouse)]

Gene ID: 12959, updated on 5-Mar-2019

#### Summary

↑ ?

Official Symbol Cryba4 provided by MGI

Official Full Name crystallin, beta A4 provided by MGI

Primary source MGI:MGI:102716

See related Ensembl:ENSMUSG00000066975

Gene type protein coding
RefSeq status REVIEWED
Organism Mus musculus

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;

Muroidea; Muridae; Murinae; Mus; Mus

Summary This gene encodes a member of the crystallin family of proteins that contribute to the transparency and refractive properties of the ocular lens.

Certain mutations in the human ortholog of this gene are associated with cataract and bilateral microphthalmia. This gene is located adjacent to a related crystallin gene on chromosome 5. Alternative splicing results in multiple transcript variants encoding different isoforms. [provided

by RefSeq, Aug 2015]

Expression Biased expression in genital fat pad adult (RPKM 82.6), adrenal adult (RPKM 22.3) and 2 other tissuesSee more

Orthologs <u>human all</u>

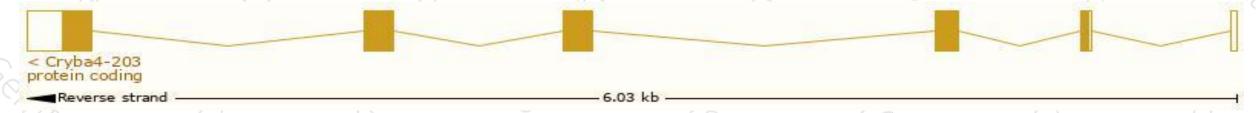
# Transcript information (Ensembl)



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

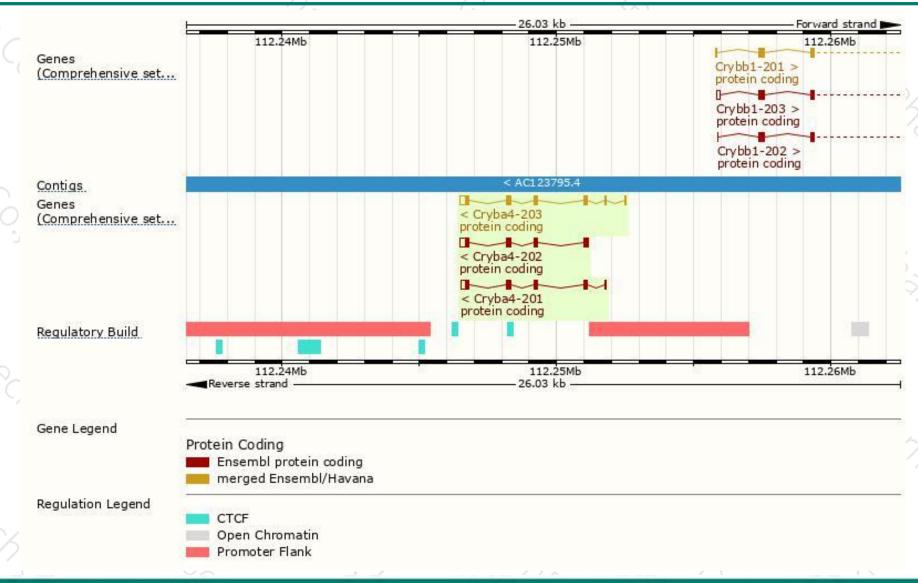
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cryba4-203	ENSMUST00000112385.7	808	<u>196aa</u>	Protein coding	CCDS19536	Q3V1A0 Q9JJV0	TSL:1 GENCODE basic APPRIS P1
Cryba4-202	ENSMUST00000112383.7	754	<u>183aa</u>	Protein coding	CCDS84931	E9QAS6	TSL:1 GENCODE basic
Cryba4-201	ENSMUST00000086629.5	746	<u>196aa</u>	Protein coding	CCDS19536	Q3V1A0 Q9JJV0	TSL:1 GENCODE basic APPRIS P1

The strategy is based on the design of Cryba4-203 transcript, The transcription is shown below



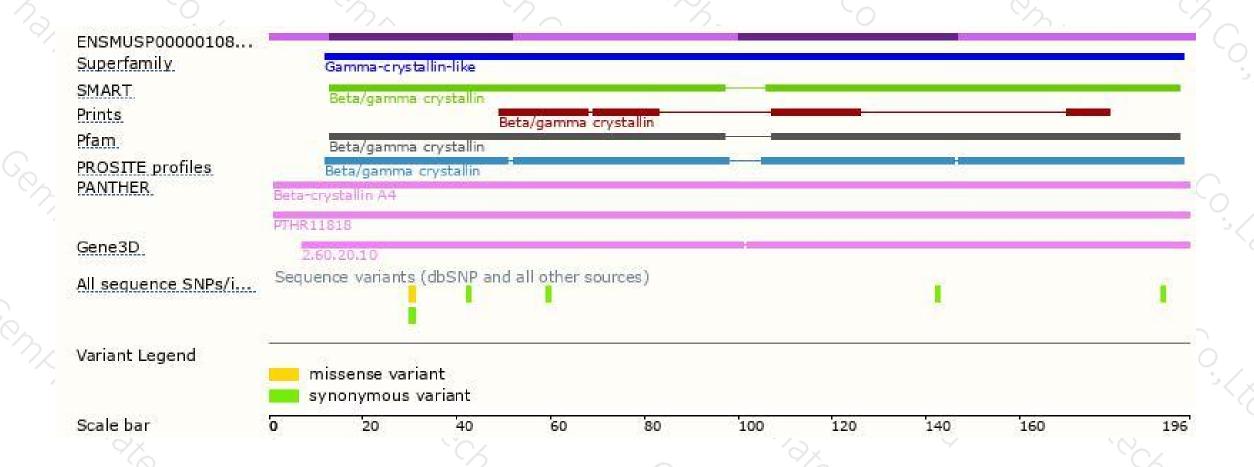
### Genomic location distribution





### Protein domain







If you have any questions, you are welcome to inquire. Tel: 400-9660890





