

# Cr2 Cas9-CKO Strategy

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## **Project Overview**



Project Name Cr2

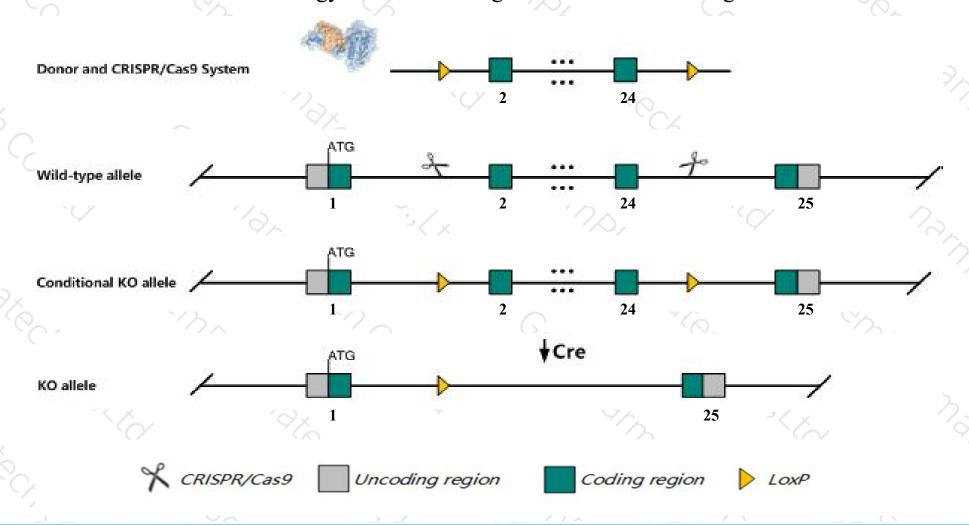
Project type Cas9-CKO

Strain background C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Cr2 gene. The schematic diagram is as follows:



### Technical routes



- The Cr2 gene has 12 transcripts. According to the structure of Cr2 gene, exon2-exon24 of Cr2-212 (ENSMUST00000210219.1) transcript is recommended as the knockout region. The region contains most of coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Cr2* 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**



- ➤ According to the existing MGI data, Homozygotes for targeted null mutations exhibit impaired humoral immune responses to T cell-dependent antigens, with limited affinity maturation, and reduced memory B cell and germinal center formation.
- > The Cr2 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)



#### Cr2 complement receptor 2 [ Mus musculus (house mouse) ]

Gene ID: 12902, updated on 21-Aug-2019

#### Summary

△ ?

Official Symbol Cr2 provided by MGI

Official Full Name complement receptor 2 provided by MGI

Primary source MGI:MGI:88489

See related Ensembl: ENSMUSG00000026616

Gene type protein coding
RefSeq status VALIDATED
Organism <u>Mus musculus</u>

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

Muroidea; Muridae; Murinae; Mus; Mus

Also known as Cr1; C3DR; CD21; CD35; Cr-1; Cr-2

Expression Biased expression in spleen adult (RPKM 7.1), mammary gland adult (RPKM 1.4) and 1 other tissue See more

Orthologs <u>human</u> all

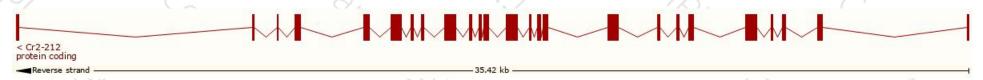
## Transcript information (Ensembl)



The gene has 12 transcripts, all transcripts are shown below:

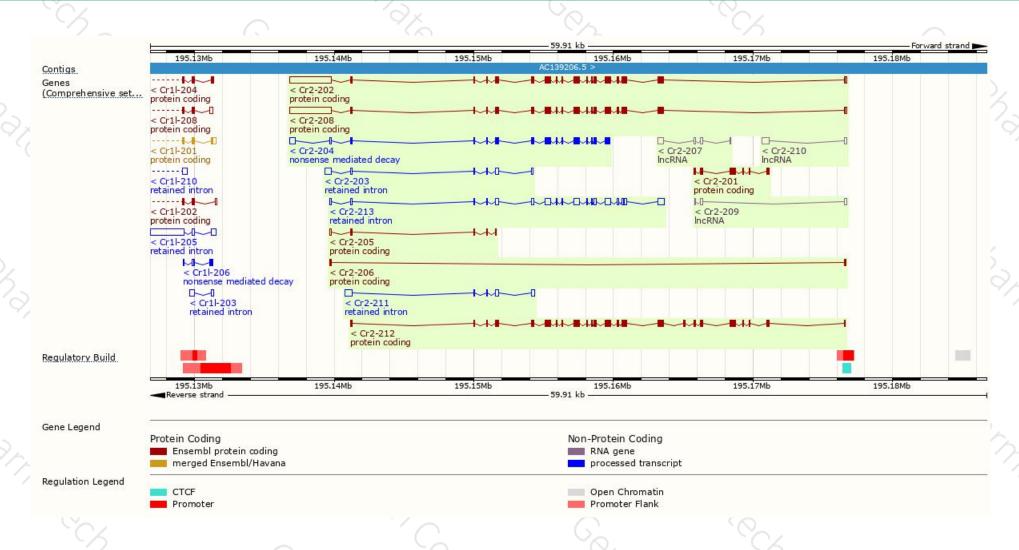
Name 4	Transcript ID	bp 🎂	Protein 4	Biotype	CCDS .	UniProt 🌢	Flags
Cr2-202	ENSMUST00000082321.8	6207	<u>1032aa</u>	Protein coding	CCDS15642 ₽	Q9DC83₽	TSL:1 GENCODE basic
Cr2-208	ENSMUST00000195120.5	6212	1026aa	Protein coding	120	A0A0A6YWG3₺	TSL:5 GENCODE basic
Cr2-212	ENSMUST00000210219.1	4227	<u>1408aa</u>	Protein coding	(2)	A0A1B0GS59₽	TSL:5 GENCODE basic APPRIS P1
Cr2-201	ENSMUST00000043104.8	1044	347aa	Protein coding	123	E9Q3S5 @	TSL:5 GENCODE basic
Cr2-205	ENSMUST00000193436.1	355	<u>62aa</u>	Protein coding	(2)	A0A0A6YXU7₽	CDS 5' incomplete TSL:5
Cr2-206	ENSMUST00000193801.1	206	<u>50aa</u>	Protein coding	120	Q32M14₽	TSL:1 GENCODE basic
Cr2-204	ENSMUST00000193356.1	2736	<u>735aa</u>	Nonsense mediated decay	127	A0A0A6YWU7₽	CDS 5' incomplete TSL:1
Cr2-213	ENSMUST00000238929.1	3220	No protein	Retained intron	129	2	450
Cr2-211	ENSMUST00000195737.1	1000	No protein	Retained intron	20	2	TSL:3
Cr2-203	ENSMUST00000192604.5	974	No protein	Retained intron	323	2	TSL:3
Cr2-207	ENSMUST00000194149.5	751	No protein	IncRNA ■	120	2	TSL:3
Cr2-210	ENSMUST00000195722.1	689	No protein	IncRNA	25	2	TSL:2
Cr2-209	ENSMUST00000195347.5	357	No protein	IncRNA	120	9	TSL:3

The strategy is based on the design of Cr2-212 transcript, The transcription is shown below



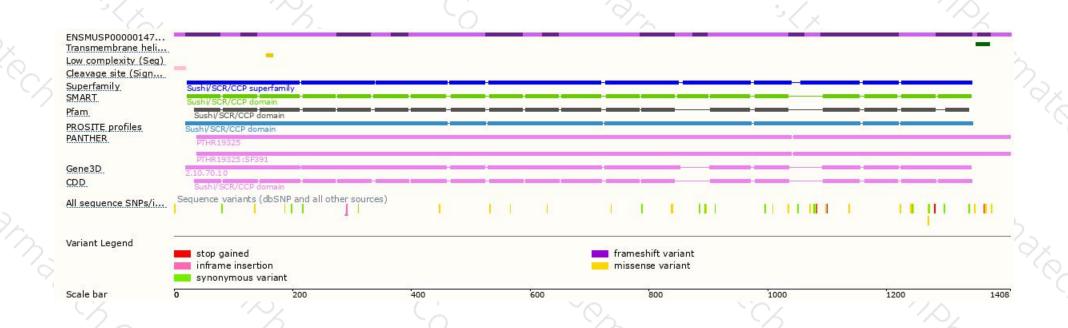
### Genomic location distribution





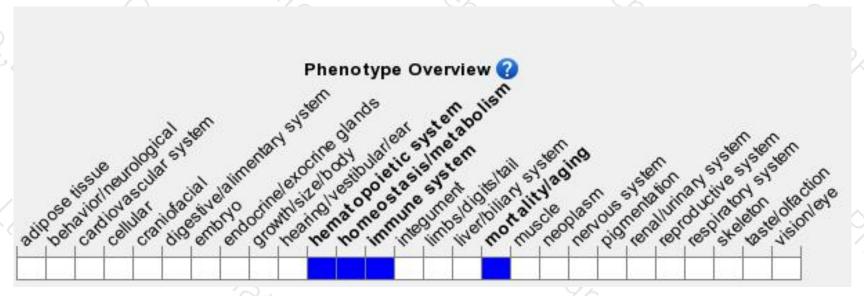
### Protein domain





## 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, Homozygotes for targeted null mutations exhibit impaired humoral immune responses to T cell-dependent antigens, with limited affinity maturation, and reduced memory B cell and germinal center formation.



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





