

# Ebi3 Cas9-CKO Strategy

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Reviewer: Rui xiong

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# **Project Overview**



**Project Name** 

**Project type** 

Strain background

Cas9-CKO

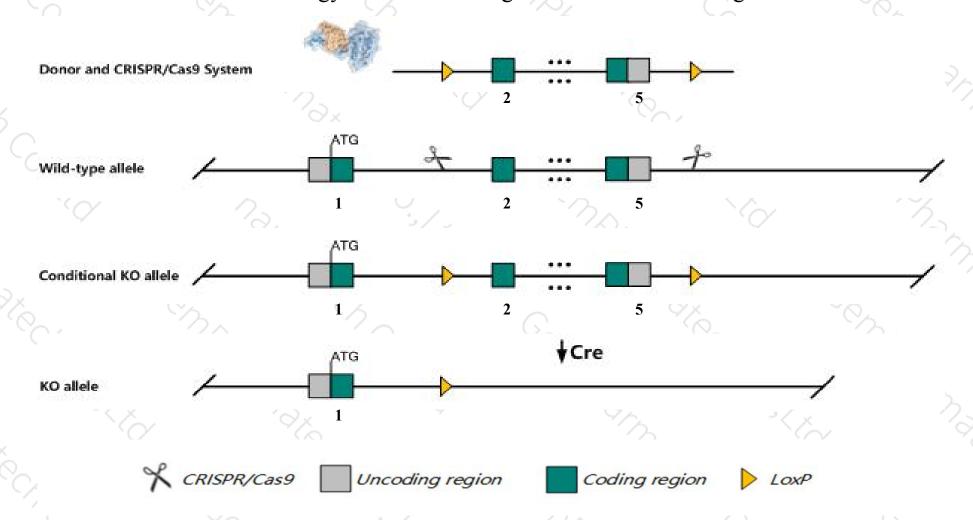
Ebi3

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Ebi3* gene. The schematic diagram is as follows:



### **Technical routes**



- ➤ The *Ebi3* gene has 1 transcript. According to the structure of *Ebi3* gene, exon2-exon5 of *Ebi3*201(ENSMUST0000003274.7) transcript is recommended as the knockout region. The region contains 620bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Ebi3* 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,mice homozygous for a knock-out allele exhibit significantly reduced numbers of invariant natural killer (NK) T cells, show impaired Th2 responses, are resistant to the induction of oxazolone-induced colitis, and display impaired early Th1 immunity against Leishmania parasitic infections.
- The floxed region is near to the N-terminal of Yju2 gene, this strategy may influence the regulatory function of the N-terminal of Yju2 gene.
- > The *Ebi3* gene is located on the Chr17. 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)



#### Ebi3 Epstein-Barr virus induced gene 3 [Mus musculus (house mouse)]

Gene ID: 50498, updated on 13-Mar-2020

#### Summary

☆ ?

Official Symbol Ebi3 provided by MGI

Official Full Name Epstein-Barr virus induced gene 3 provided by MGI

Primary source MGI:MGI:1354171

See related Ensembl:ENSMUSG00000003206

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 EBI-3, IL-27

Expression Broad expression in spleen adult (RPKM 18.1), mammary gland adult (RPKM 10.2) and 21 other tissuesSee more

Orthologs <u>human all</u>

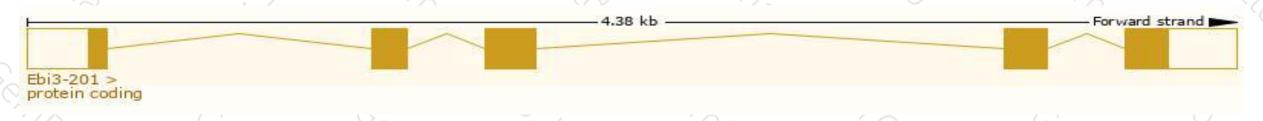
# Transcript information (Ensembl)



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

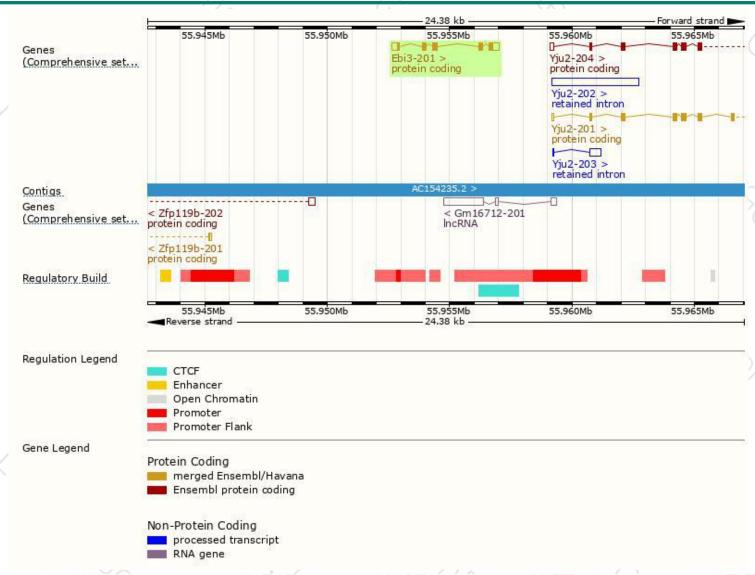
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags	
Ebi3-201	ENSMUST00000003274.7	1154	228aa	Protein coding	CCDS28888	O35228 Q3U1K3	TSL:1 GENCODE basic APPRIS P1	

The strategy is based on the design of *Ebi3-201* transcript, the transcription is shown below:



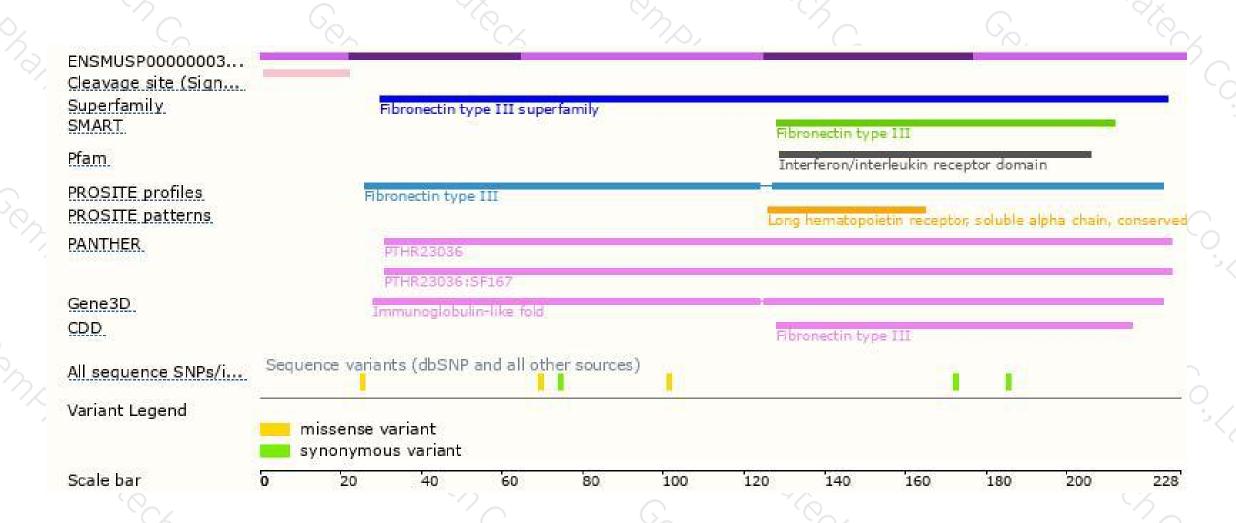
### Genomic location distribution





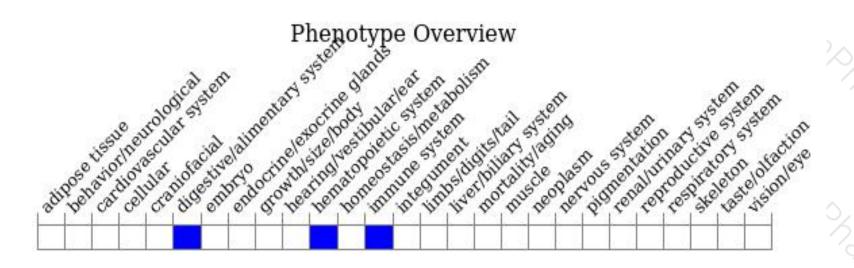
### Protein domain





# 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 significantly reduced numbers of invariant natural killer (NK) T cells, show impaired Th2 responses, are resistant to the induction of oxazolone-induced colitis, and display impaired early Th1 immunity against Leishmania parasitic infections.



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





