

# Cblc Cas9-CKO Strategy

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## **Project Overview**



Project Name Cblc

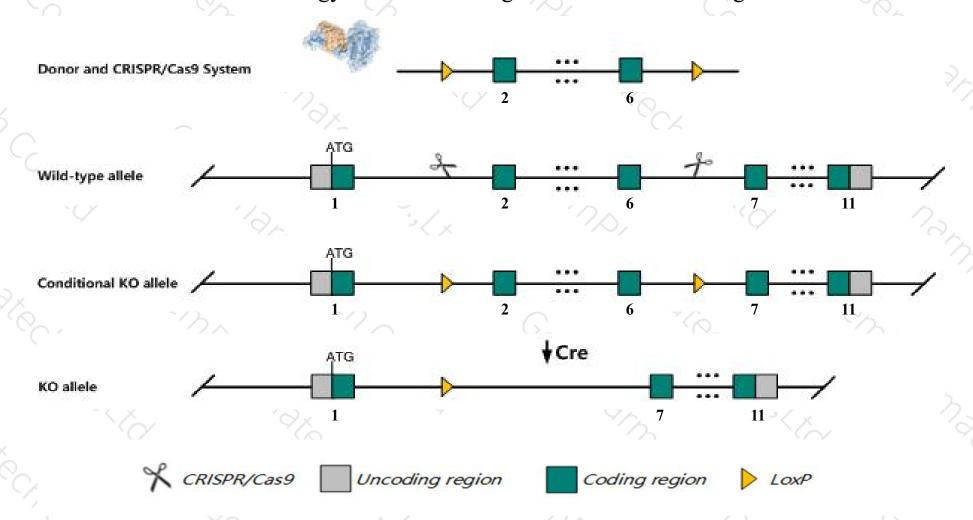
Project type Cas9-CKO

Strain background C57BL/6JGpt

## Conditional Knockout strategy



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



## Technical routes



- The *Cblc* gene has 7 transcripts. According to the structure of *Cblc* gene, exon2-exon6 of *Cblc-201*(ENSMUST00000043822.7) transcript is recommended as the knockout region. The region contains 652bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Cblc* 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, Homozygous null mice are viable, fertile, and show no abnormalities of the epithelium or other tissues.
- The *Cblc* gene is located on the Chr7. 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)



△ ?

#### Cblc Casitas B-lineage lymphoma c [ Mus musculus (house mouse) ]

Gene ID: 80794, updated on 31-Dec-2019

Summary

Official Full Name Casitas B-lineage lymphoma c provided by MGI

Primary source MGI:MGI:1931457

Official Symbol Cblc provided by MGI

See related Ensembl: ENSMUSG00000040525

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 Cbl3; 2310076l21Rik; 2310079L19Rik

Expression Biased expression in colon adult (RPKM 57.9), duodenum adult (RPKM 51.4) and 10 other tissues See more

Orthologs human all

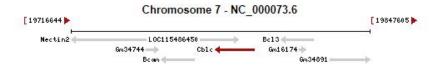
#### Genomic context

Location: 7:7 A3

See Cblc in Genome Data Viewe

Exon count: 12

| Annotation release | Status            | Assembly  | Chr Location   |  |  |
|--------------------|-------------------|---|--|--|--|
| 108                | current           | GRCm38.p6 (GCF_000001635.26) 7 NC_000073.6 (1977971819796809, compl |  | NC_000073.6 (1977971819796809, complement) |  |
| Build 37.2         | previous assembly | MGSCv37 (GCF_000001635.18)  | 0001635.18) 7 NC_000073.5 (2036506720382158, complement) |  |  |



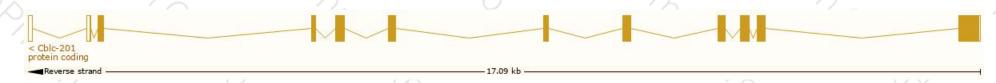
## Transcript information (Ensembl)



The gene has 7 transcripts, all transcripts are shown below:

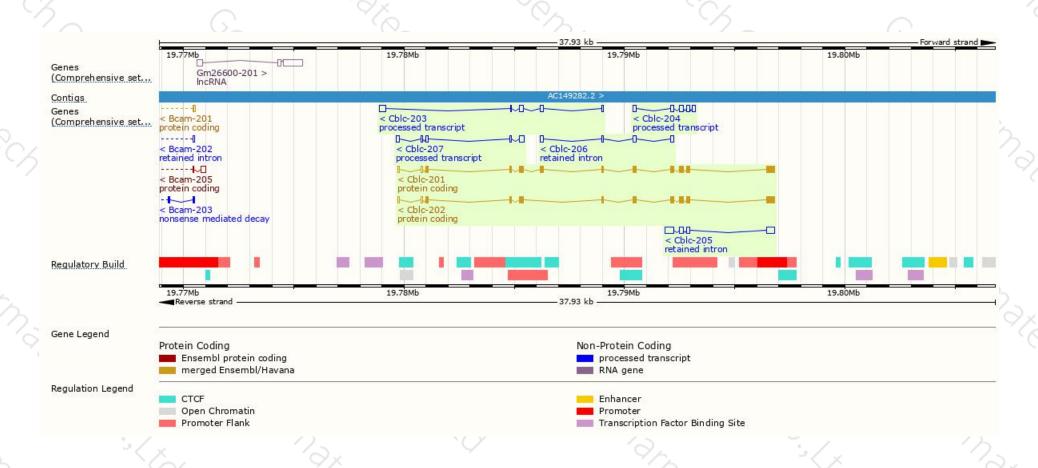
| Name 🍦   | Transcript ID #      | bp 🌲 | Protein 4    | Biotype              | CCDS 🍦     | UniProt         | Flags                         |  |
|----------|----------------------|------|--------------|----------------------|------------|-----------------|-------------------------------|--|
| Cblc-201 | ENSMUST00000043822.7 | 1662 | <u>496aa</u> | Protein coding       | CCDS39804₽ | Q80XL1@         | TSL:1 GENCODE basic APPRIS P1 |  |
| Cblc-202 | ENSMUST00000108449.8 | 1514 | 452aa        | Protein coding       | CCDS52064₽ | <u>G3X9U0</u> ₽ | TSL:1 GENCODE basic           |  |
| Cblc-204 | ENSMUST00000131276.7 | 746  | No protein   | Processed transcript | 17         | (2)             | TSL:5                         |  |
| Cblc-203 | ENSMUST00000125050.1 | 739  | No protein   | Processed transcript | 10         | (5)             | TSL:3                         |  |
| Cblc-207 | ENSMUST00000148416.1 | 635  | No protein   | Processed transcript | 17         | (2)             | TSL:2                         |  |
| Cblc-205 | ENSMUST00000131602.1 | 1052 | No protein   | Retained intron      | 17         | (2)             | TSL:1                         |  |
| Cblc-206 | ENSMUST00000145755.1 | 497  | No protein   | Retained intron      | 10         | 8 <u>5</u> 8    | TSL:5                         |  |

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



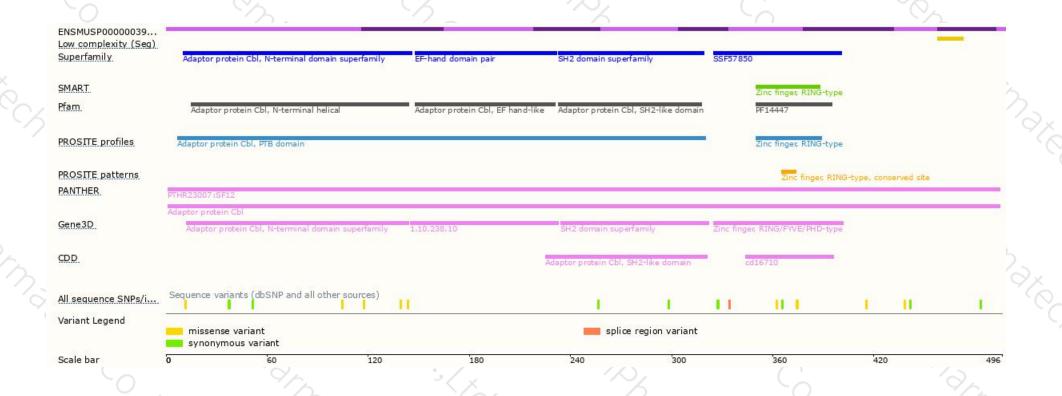
## Genomic location distribution





### Protein domain







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





