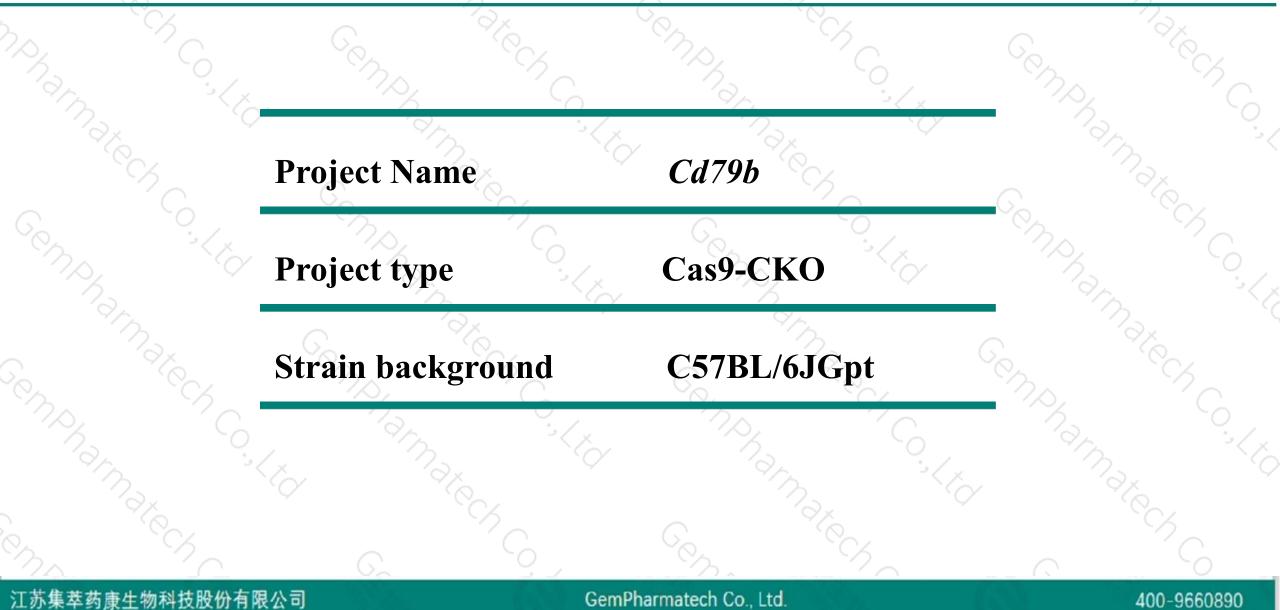


Cd79b Cas9-CKO Strategy

Designer: Reviewer: Design Date: Huimin Su Ruirui Zhang 2019/10/12

Project Overview



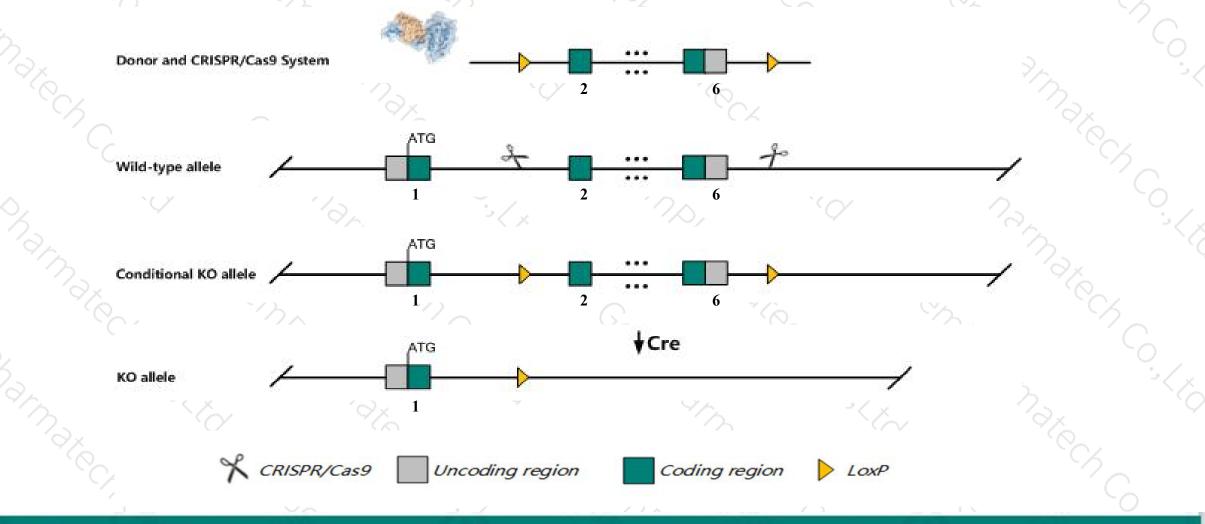


Conditional Knockout strategy



400-9660890

This model will use CRISPR/Cas9 technology to edit the Cd79b gene. The schematic diagram is as follows:



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The Cd79b gene has 2 transcripts. According to the structure of Cd79b gene, exon2-exon6 of Cd79b-202 (ENSMUST00000167143.1) 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 *Cd79b* 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.



- According to the existing MGI data, Homozygotes for targeted null mutations exhibit arrested development of B cells at the pro-B cell stage due to diminished signaling of the B cell receptor.
- > The *Cd79b* gene is located on the Chr11. 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)



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Cd79b CD79B antigen [Mus musculus (house mouse)]

Gene ID: 15985, updated on 11-Sep-2019

Summary

Official Symbol Cd79b provided by MGI Official Full Name CD79B antigen provided by MGI Primary source MGI:MGI:96431 See related Ensembl:ENSMUSG00000040592 Gene type protein coding RefSeg status REVIEWED Organism Mus musculus Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus Also known as B29; Igb; Igbeta; Ig-beta Summary The B lymphocyte antigen receptor is a multimeric complex that includes the antigen-specific component, surface immunoglobulin (Ig). Surface Ig non-covalently associates with two other proteins, Ig-alpha and Ig-beta, which are necessary for expression and function of the B-cell antigen receptor. This gene encodes the Ig-beta protein of the B-cell antigen component. Alternatively spliced transcript variants encoding different isoforms have been described. [provided by RefSeq, Sep 2015] Biased expression in spleen adult (RPKM 246.8), mammary gland adult (RPKM 54.7) and 2 other tissues See more Expression Orthologs human all

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Transcript information (Ensembl)



The gene has 2 transcripts, all transcripts are shown below:

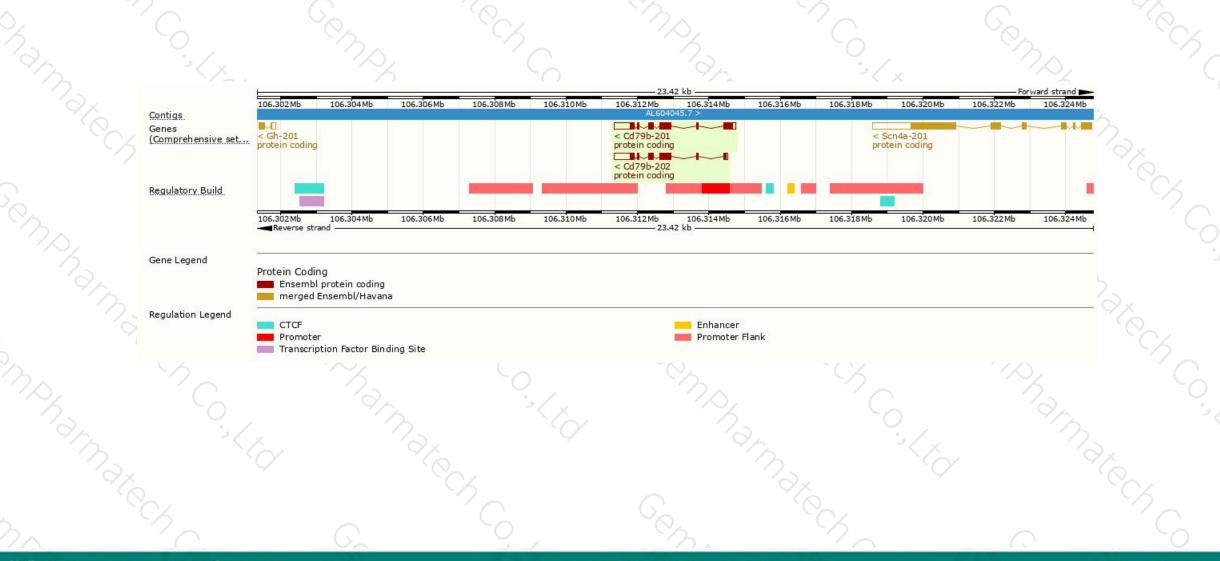
| Name 🖕 | Transcript ID 💧 | bp 🖕 | Protein 🖕 | Biotype 💧 | CCDS 🖕 | UniProt | Flags | | |
|-----------|----------------------|------|--------------|----------------|--------------------|-----------------|-------|---------------|-------------|
| Cd79b-202 | ENSMUST00000167143.1 | 1195 | <u>228aa</u> | Protein coding | <u>CCDS48960</u> & | <u>P15530</u> @ | TSL:1 | GENCODE basic | APPRIS P2 |
| Cd79b-201 | ENSMUST0000044228.10 | 1431 | <u>288aa</u> | Protein coding | 17 A | B1ARJ9@ | TSL:1 | GENCODE basic | APPRIS ALT2 |

The strategy is based on the design of *Cd79b-202* transcript, The transcription is shown below



Genomic location distribution





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Protein domain



| | ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~ | | Solo. | ax Ch | Const. | °°Y | S. | | Nate Ch |
|------------------------|--|-------------------------------|-------------------------------|--|-------------|--------------|---------|-----------------------------------|---------------|
| 22 | ENSMUSP00000129 Transmembrane heli PDB-ENSP mappings Low complexity (Seg) Cleavage site (Sign Superfamily SMART. | | | ulin-like domain superfamily globulin subtype | | | _ | Phosphorylated immunorec | aptor signalX |
| | <u>Pfam</u> | | Im | munoglobulin I-set | | | | Phosphorylated immunorec | eptor signa |
| | PROSITE profiles | | | Immunoglobulin-like | e domain | _ | | Phosphorylated immunoreceptor sig | nalling ITA |
| 2 | PANTHER | B-cell antigen receptor compl | ex-associated protein alpha/b | eta chain | | | | | = |
| $\langle \phi \rangle$ | Gene3D | PTHR14334:SF2 | Immunoglobulir | 1-like fold | | | | | |
| | CDD | | cd1609 | | | | | | |
| | All sequence SNPs/i | Sequence variants (dbSNP | and all other sources) | 1 I.) | | (a) | | | |
| | Variant Legend | missense variant | | synonyme | ous variant | | | | |
| | Scale bar | | 40 | 60 80 | 100 120 | 140 | 160 180 | 200 | 228 |
| | C. S. | | <u>6</u> | ~^ ~ | Gen. | | 3 | | |

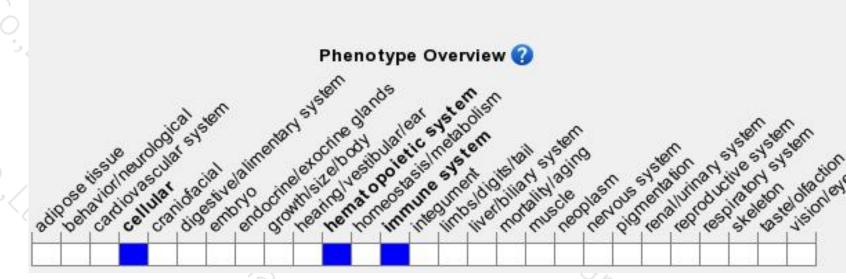
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Mouse phenotype description(MGI)





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 arrested development of B cells at the pro-B cell stage due to diminished signaling of the B cell receptor.



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



