

Kl Cas9-CKO Strategy

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Overview

Target Gene Name

• K1

Project Type

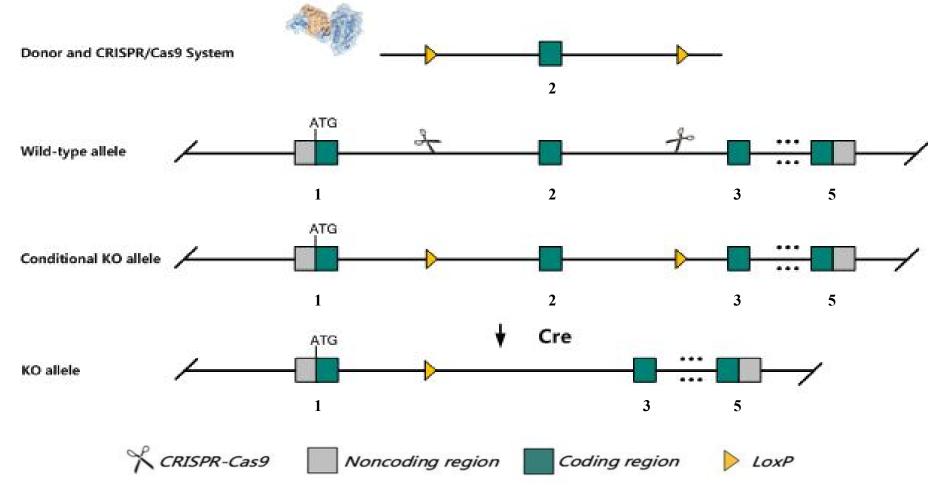
• Cas9-CKO

Genetic Background

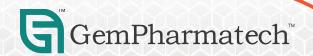
• C57BL/6JGpt



Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the Kl gene.



Technical Information

- The *Kl* gene has 2 transcripts. According to the structure of *Kl* gene, exon2 of *Kl*-201 (ENSMUST00000078856.8) transcript is recommended as the knockout region. The region contains 511bp coding sequence. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Kl* 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.
- 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.



Gene Information

KI klotho [Mus musculus (house mouse)]

Gene ID: 16591, updated on 13-Mar-2020

Summary

Official Symbol KI provided by MGI
Official Full Name klotho provided by MGI
Primary source MGI:MGI:1101771

See related Ensembl: ENSMUSG00000058488

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 alpha-kl

Expression Restricted expression toward kidney adult (RPKM 58.5)See more

Orthologs <u>human all</u>

Source: https://www.ncbi.nlm.nih.gov/

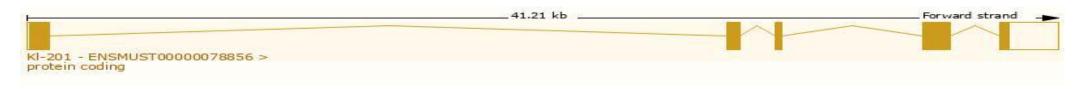


Transcript Information

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



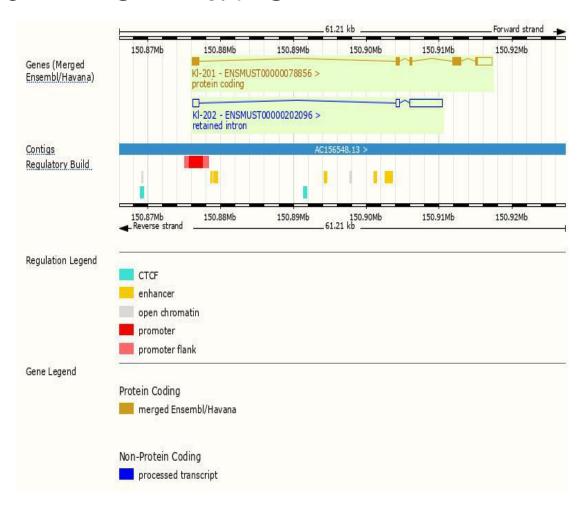
The strategy is based on the design of Kl-201 transcript, the transcription is shown below:



Source: https://www.ensembl.org



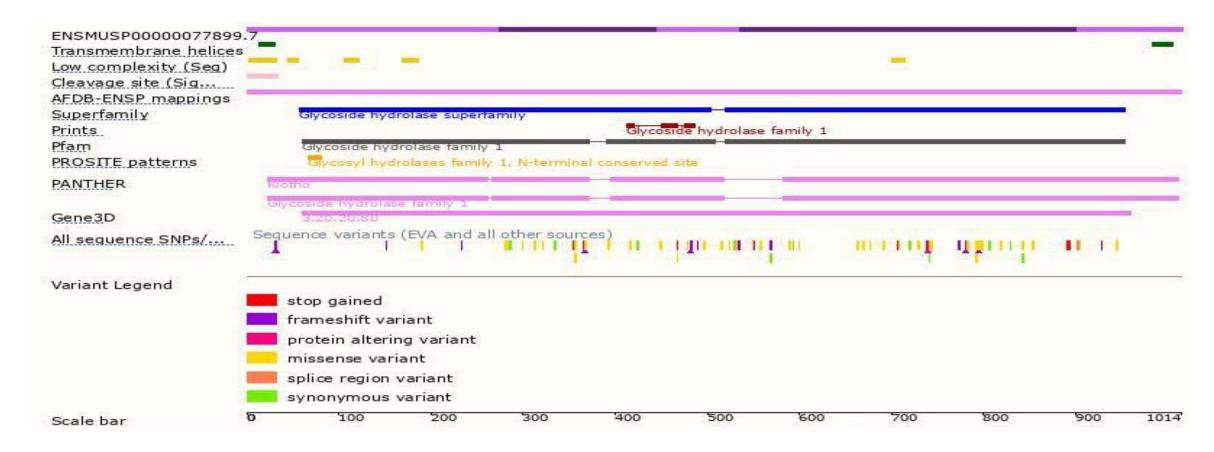
Genomic Information

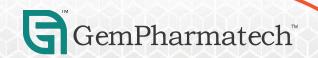




Source: : https://www.ensembl.org

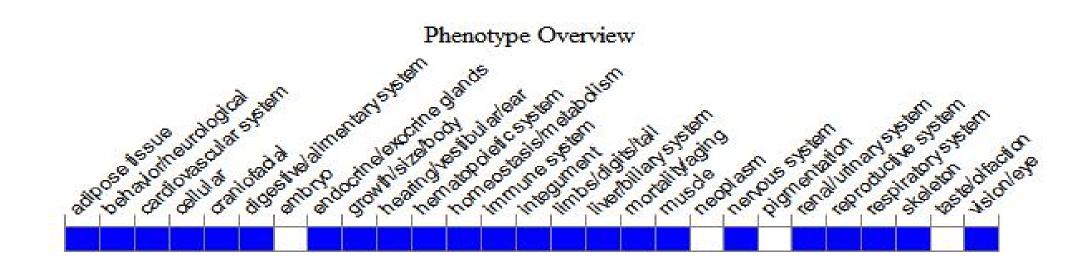
Protein Information





Source: : https://www.ensembl.org

Mouse Phenotype Information (MGI)



• Homozygous mutant mice have a short lifespan and growth retardation with one allele homeostatic imbalances and soft tissue calcification are also seen. With a second allele abnormal cancellous bone and femur morphology are seen.



Source: https://www.informatics.jax.org

Important Information

- *Kl* is located on Chr5. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is 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 the existing technology level.

