

Extl3 Cas9-CKO Strategy

Designer: Yanhua Shen

Reviewer: Xueting Zhang

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Overview

Target Gene Name

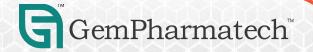
• Extl3

Project Type

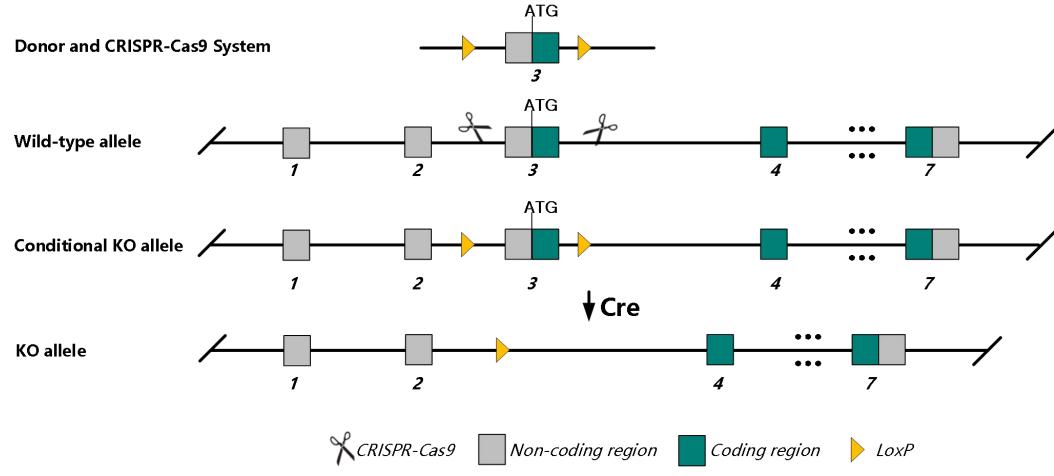
• Cas9-CKO

Genetic Background

• C57BL/6JGpt



Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the Extl3 gene.



Technical Information

- The Extl3 gene has 3 transcripts. According to the structure of Extl3 gene, exon3 of Extl3-203 (ENSMUST00000225633.2) transcript is recommended as the knockout region. The region contains ATG and most of coding sequences. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Extl3* 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



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

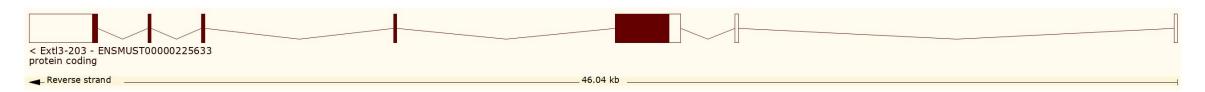


Transcript Information

The gene has 3 transcripts, all transcripts are shown below:

Transcript ID	Name 🍦	bp 🛊	Protein	Biotype	CCDS 🍦	UniProt Match	Flags
ENSMUST00000225633.2	Extl3-203	6013	<u>919aa</u>	Protein coding	CCDS27211 ₺	<u>Q6P1H4</u> ₽	Ensembl Canonical GENCODE basic APPRIS P1
ENSMUST00000022550.8	Extl3-201	5977	<u>919aa</u>	Protein coding	CCDS27211 ₺	Q6P1H4₽	GENCODE basic APPRIS P1 TSL:5
ENSMUST00000223989.2	Extl3-202	582	No protein	Protein coding CDS not defined			10

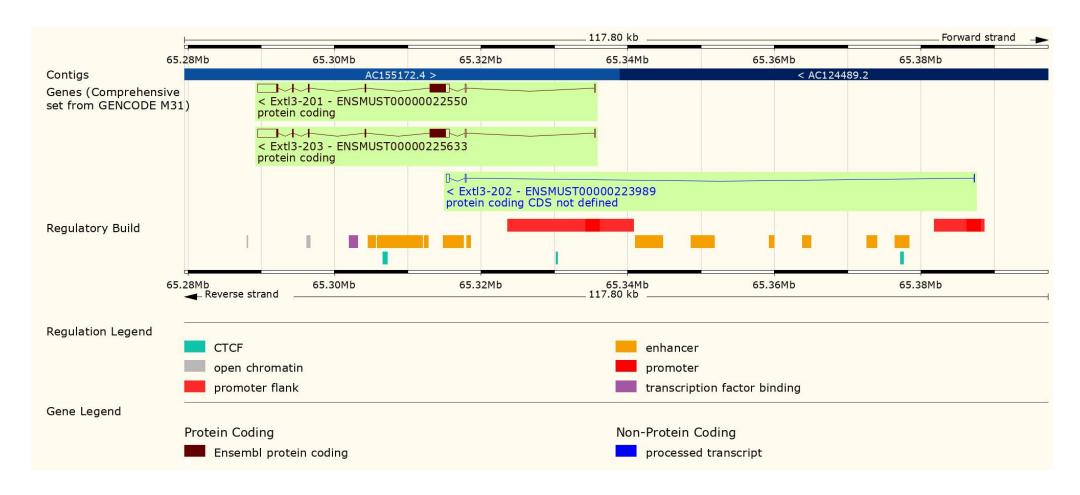
The strategy is based on the design of Extl3-203 transcript, the transcription is shown below:



Source: https://www.ensembl.org



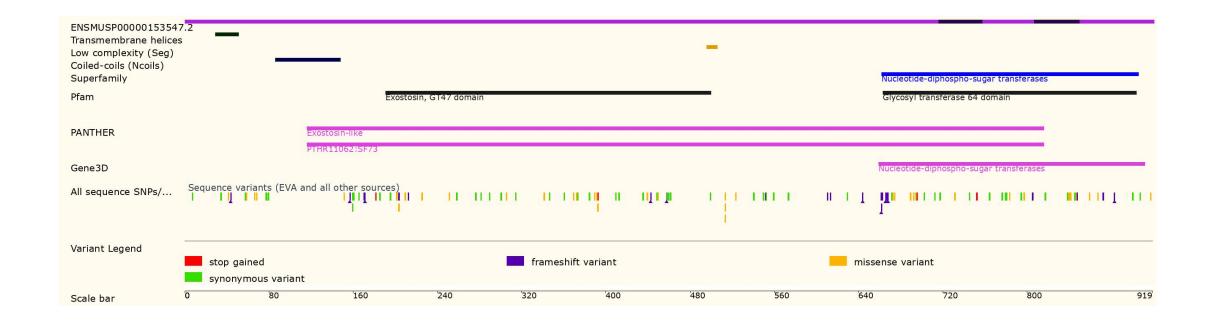
Genomic Information





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

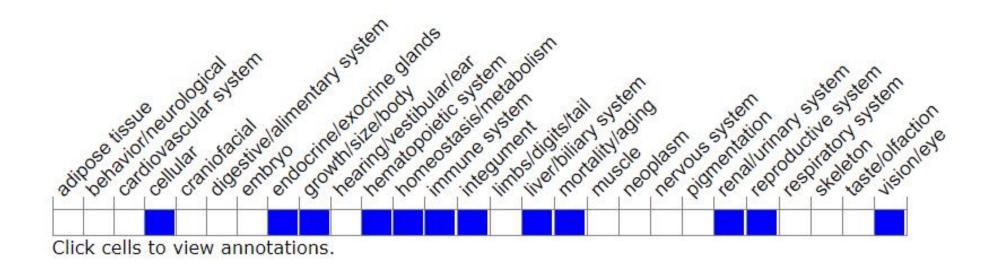
Protein Information





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

Mouse Phenotype Information (MGI)



• Mice homozygous for a null mutation display embryonic lethality during organogenesis and lack heparan sulfate derived disaccharides.



Important Information

- Extl3 is located on Chr14. 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.

