

Klhl3 Cas9-CKO Strategy

Designer: Xueting Zhang

Reviewer: Yanhua Shen

Date:2020-03-03

Project Overview



Project Name

Klhl3

Project type

Cas9-CKO

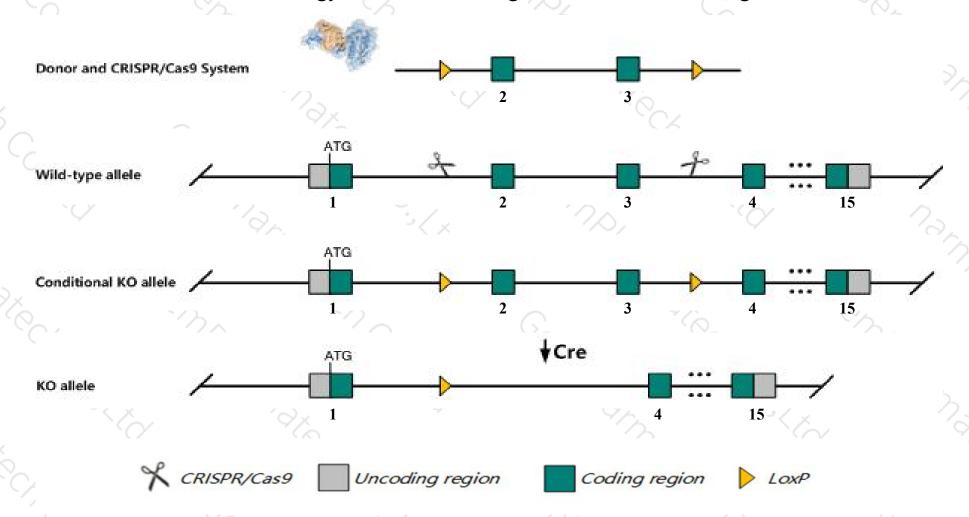
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Klhl3 gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Klhl3* gene has 2 transcripts. According to the structure of *Klhl3* gene, exon2-exon3 of *Klhl3-202*(ENSMUST00000160860.8) transcript is recommended as the knockout region. The region contains 227bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Klhl3* 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 carrying a point mutation display salt-sensitive hypertension, hyperkalemia and metabolic acidosis.
- The *Klhl3* gene is located on the Chr13. 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)



KIhl3 kelch-like 3 [Mus musculus (house mouse)]

Gene ID: 100503085, updated on 24-Oct-2019

Summary

☆ ?

Official Symbol Klhl3 provided by MGI

Official Full Name kelch-like 3 provided by MGI

Primary source MGI:MGI:2445185

See related Ensembl: ENSMUSG00000014164

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 Al430941; EG627648; 7530408C15Rik

Expression Broad expression in kidney adult (RPKM 6.1), frontal lobe adult (RPKM 4.8) and 21 other tissues See more

Orthologs human all

Genomic context



Location: 13; 13 B1

See Klhl3 in Genome Data Viewer

Exon count: 17

Annotation release	Status	Assembly	Chr	Location			
108	current	GRCm38.p6 (GCF_000001635.26)	13	NC_000079.6 (5800022658123477, complement)			
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	13	NC_000079.5 (5810221158203789, complement)			

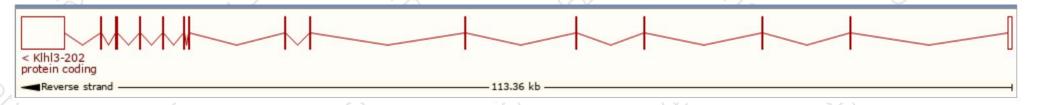
Transcript information (Ensembl)



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

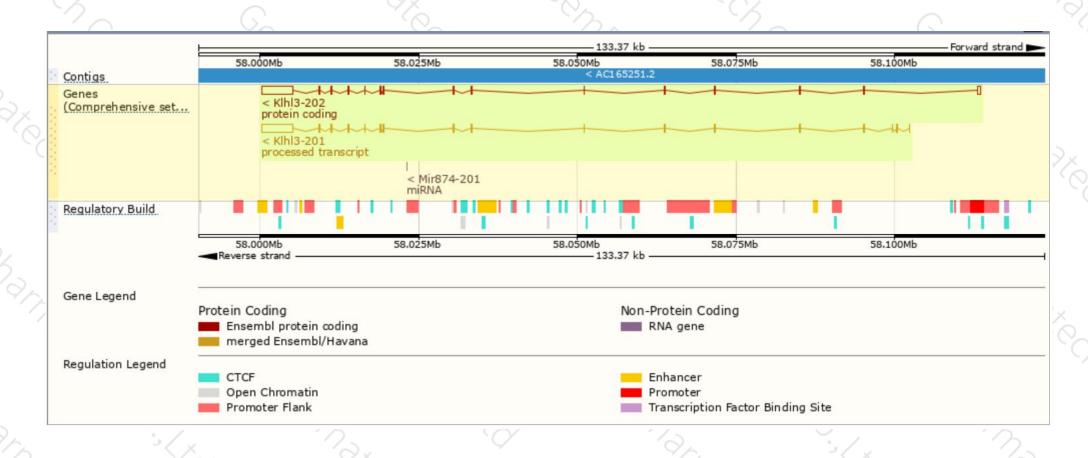
Name 🍦	Transcript ID 👙	bp 🍦	Protein	Biotype	CCDS	UniProt	Flags		
KIhl3-202	ENSMUST00000160860.8	7149	<u>587aa</u>	Protein coding	-	E0CZ16母	TSL:5	GENCODE basic	APPRIS P1
Klhl3-201	ENSMUST00000091583.5	6871	No protein	Processed transcript	-	-	TSL:5		

The strategy is based on the design of Klhl3-202 transcript, The transcription is shown below



Genomic location distribution





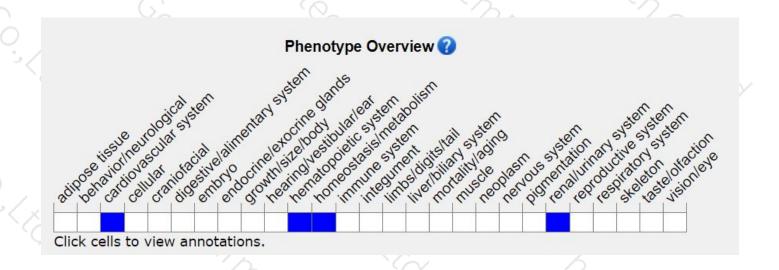
Protein domain





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 carrying a point mutation display salt-sensitive hypertension, hyperkalemia and metabolic acidosis.



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





