

# Alkbh2 Cas9-KO Strategy

Designer: Xueting Zhang

Reviewer: Yanhua Shen

Date:2020-03-04

## **Project Overview**



**Project Name** 

Alkbh2

**Project type** 

Cas9-KO

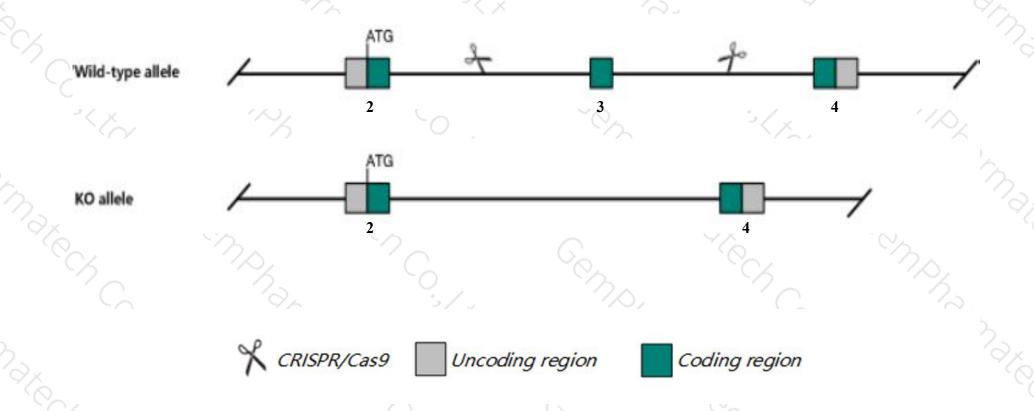
Strain background

C57BL/6JGpt

## **Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the Alkbh2 gene. The schematic diagram is as follows:



### **Technical routes**



- ➤ The Alkbh2 gene has 3 transcripts. According to the structure of Alkbh2 gene, exon3 of Alkbh2-201

  (ENSMUST00000053657.12) transcript is recommended as the knockout region. The region contains 199bp coding sequence Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Alkbh2* gene. The brief process is as follows: CRISPR/Cas9 system

### **Notice**



- According to the existing MGI data, Homozygous null mice are viable and overtly normal but show progressive accumulation of 1-methyladenine (1meA) in their genomic DNA due to impaired DNA repair. Mutant MEFs fail to remove methyl methane sulfate (MMS)-induced 1meA from genomic DNA and showincreased cytotoxicity after MMS exposure.
- The effect on transcript *Alkbh2*-203 is unknown.
- The knockout region is near to the C-terminal of *Usp30* gene and N-terminal of *Ung* gene, this strategy may influence the regulatory function of the C-terminal of *Usp30* gene and N-terminal of *Ung* gene.
- > The *Alkbh2* gene is located on the Chr5. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

## Gene information (NCBI)



#### Alkbh2 alkB homolog 2, alpha-ketoglutarate-dependent dioxygenase [ Mus musculus (house mouse) ]

Gene ID: 231642, updated on 27-Feb-2020

#### Summary

☆ ?

Official Symbol Alkbh2 provided by MGI

Official Full Name alkB homolog 2, alpha-ketoglutarate-dependent dioxygenase provided by MGI

Primary source MGI:MGI:2141032

See related Ensembl: ENSMUSG00000044339

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 Abh2; mABH2; AU016977; 9530023G02

Expression Ubiquitous expression in ovary adult (RPKM 4.9), CNS E11.5 (RPKM 4.1) and 28 other tissues See more

Orthologs <u>human</u> all

#### Genomic context



Location: 5; 5 F

See Alkbh2 in Genome Data Viewer

Exon count: 6

Annotation release	Status	Assembly	Chr	Location	
108	current	GRCm38.p6 (GCF_000001635.26)	5	NC_000071.6 (114123926114131233, complement)	
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	5	NC_000071.5 (114573943114578185, complement)	

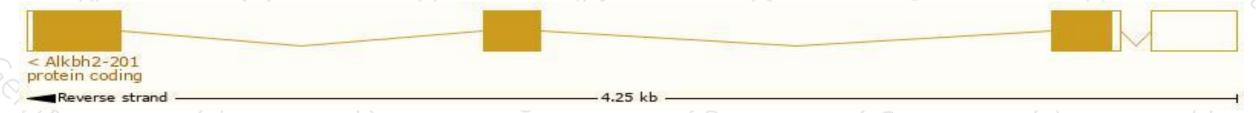
## Transcript information (Ensembl)



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

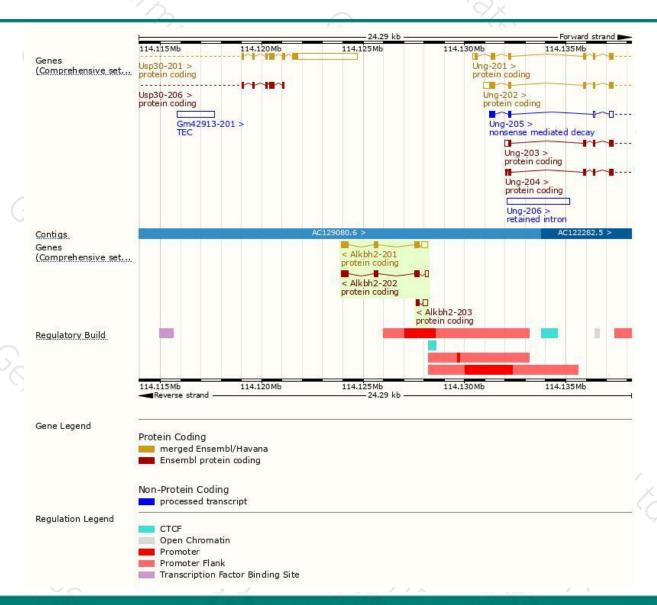
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Alkbh2-201	ENSMUST00000053657.12	1073	239aa	Protein coding	CCDS19559	Q6P6J4	TSL:1 GENCODE basic APPRIS P1
Alkbh2-202	ENSMUST00000112279.1	899	239aa	Protein coding	CCDS19559	Q6P6J4	TSL:1 GENCODE basic APPRIS P1
Alkbh2-203	ENSMUST00000149418.1	385	<u>44aa</u>	Protein coding	-	A0A0G2JER8	CDS 3' incomplete TSL:2

The strategy is based on the design of Alkbh2-201 transcript, The transcription is shown below



### Genomic location distribution





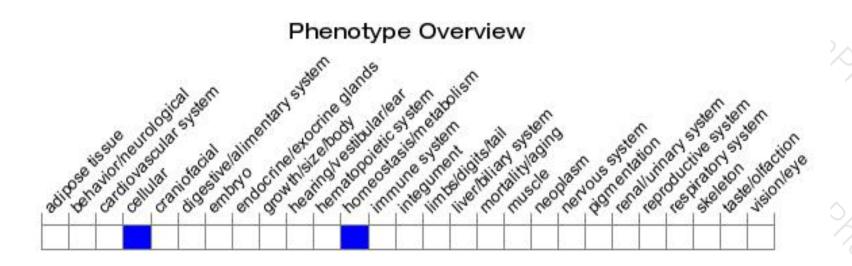
### Protein domain





## 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, Homozygous null mice are viable and overtly normal but show progressive accumulation of 1-methyladenine (1meA) in their genomic DNA due to impaired DNA repair. Mutant MEFs fail to remove methyl methan sulfate (MMS)-induced 1meA from genomic DNA and showincreased cytotoxicity after MMS exposure.



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





