

# H2-Ke6 Cas9-KO Strategy

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### Overview

#### Target Gene Name

• H2-Ke6

#### Project Type

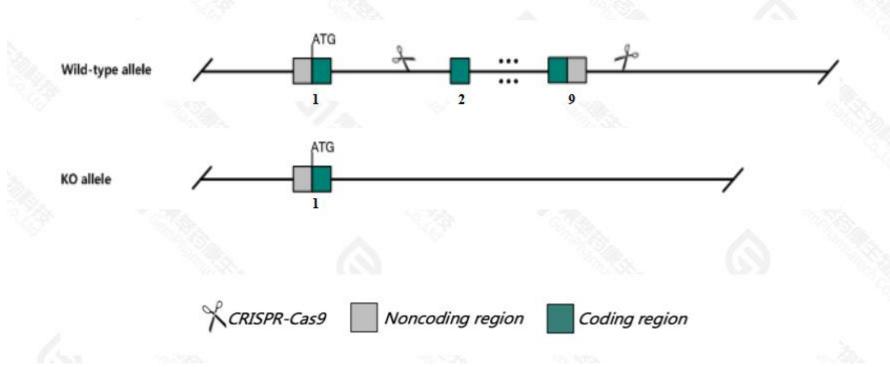
• Cas9-KO

#### Genetic Background

• C57BL/6JGpt



# Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the *H2-Ke6* gene.



#### Technical Information

- The *H2-Ke6* gene has 12 transcripts. According to the structure of *H2-Ke6* gene, exon2-exon9 of *H2-Ke6-201*(ENSMUST00000045467.14) transcript is recommended as the knockout region. The region contains 734bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *H2-Ke6* gene. The brief process is as follows: gRNAs were transcribed in vitro. Cas9 and gRNAs 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.



#### Gene Information

#### H2-Ke6 H2-K region expressed gene 6 [Mus musculus (house mouse)]

Gene ID: 14979, updated on 24-Apr-2022



Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;

Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as D17H6S112E, H-2Ke6, Hsd17b8, Ke-6, Ke6, Ring2

Expression Ubiquitous expression in adrenal adult (RPKM 155.4), ovary adult (RPKM 72.4) and 28 other tissuesSee more

Orthologs human all

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

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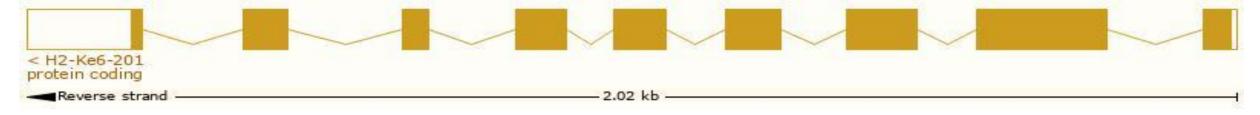


# Transcript Information

The gene has 12 transcripts, all transcripts are shown below:

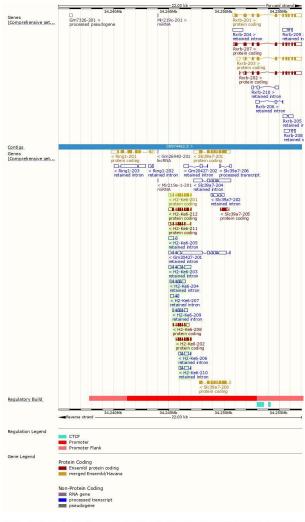
| Name       | Transcript ID         | bp   | Protein      | Biotype         | CCDS      | UniProt | Flags                               |
|------------|-----------------------|------|--------------|-----------------|-----------|---------|-------------------------------------|
| H2-Ke6-201 | ENSMUST00000045467.14 | 965  | 259aa        | Protein coding  | CCDS50071 |         | TSL:1 , GENCODE basic , APPRIS P1 , |
| H2-Ke6-212 | ENSMUST00000237759.2  | 1133 | 274aa        | Protein coding  | 14        |         | GENCODE basic ,                     |
| H2-Ke6-211 | ENSMUST00000236627.2  | 733  | <u>147aa</u> | Protein coding  | -         |         | GENCODE basic ,                     |
| H2-Ke6-208 | ENSMUST00000174399.8  | 728  | 138aa        | Protein coding  | -         |         | CDS 3' incomplete , TSL:5 ,         |
| H2-Ke6-202 | ENSMUST00000114303.4  | 716  | 234aa        | Protein coding  |           |         | CDS 3' incomplete , TSL:5 ,         |
| H2-Ke6-203 | ENSMUST00000170644.8  | 1293 | No protein   | Retained intron | 35        |         | TSL:5,                              |
| H2-Ke6-206 | ENSMUST00000173894.2  | 753  | No protein   | Retained intron | -         |         | TSL:3,                              |
| H2-Ke6-209 | ENSMUST00000235579.2  | 745  | No protein   | Retained intron | 32        |         |                                     |
| H2-Ke6-210 | ENSMUST00000236164.2  | 731  | No protein   | Retained intron | - 25      |         |                                     |
| H2-Ke6-204 | ENSMUST00000173425.9  | 709  | No protein   | Retained intron | =         |         | TSL:3,                              |
| H2-Ke6-205 | ENSMUST00000173616.8  | 534  | No protein   | Retained intron | -         |         | TSL:3,                              |
| H2-Ke6-207 | ENSMUST00000174029.8  | 459  | No protein   | Retained intron | -         |         | TSL:2,                              |

The strategy is based on the design of H2-Ke6-201 transcript, the transcription is shown below:





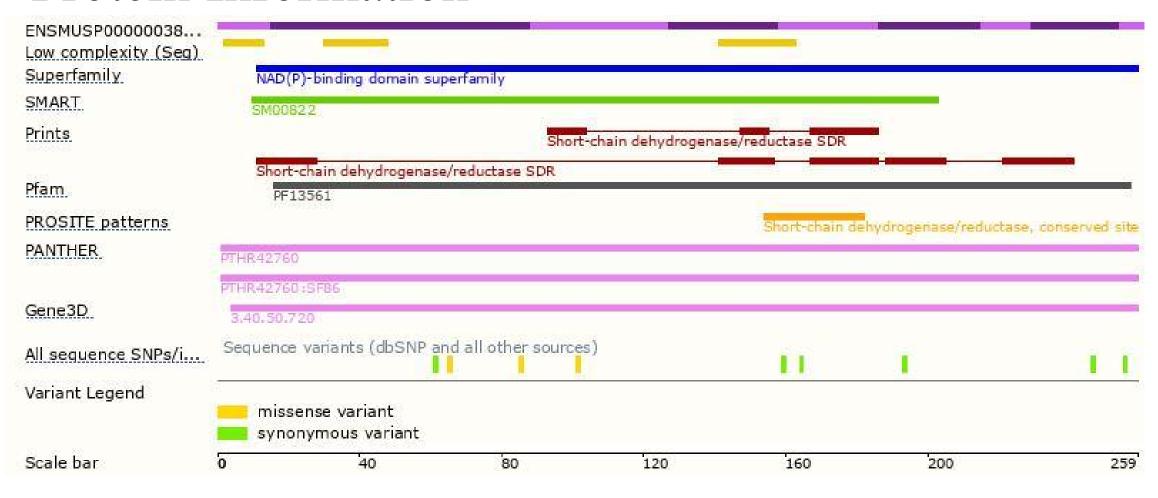
## Genomic Information





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

#### Protein Information





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

# Important Information

- The *H2-Ke6* gene is located on the Chr17. 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 risks of the mutation on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

