

# Eef2 Cas9-CKO Strategy

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#### Overview

#### Target Gene Name

• Eef2

#### Project Type

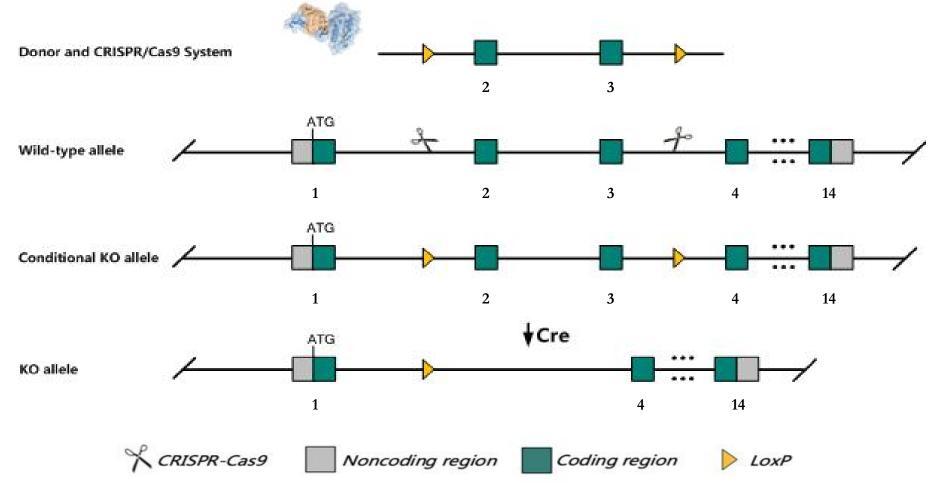
• Cas9-CKO

#### Genetic Background

• C57BL/6JGpt



## Strain Strategy

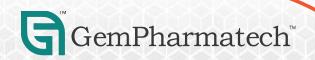


Schematic representation of CRISPR-Cas9 engineering used to edit the *Eef2* gene.



#### Technical Information

- The *Eef2* gene has 3 transcripts. According to the structure of *Eef2* gene, exon2-exon3 of *Eef2*-201 (ENSMUST00000047864.11) transcript is recommended as the knockout region. The region contains 397bp coding sequence. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Eef2* 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.



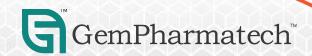
#### Gene Information

#### Eef2 eukaryotic translation elongation factor 2 [Mus musculus (house mouse)]

Gene ID: 13629, updated on 31-May-2023



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

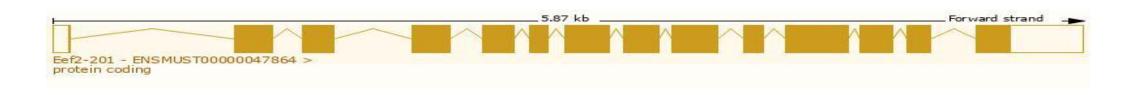


## Transcript Information

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

Show/hide columns (1 hidden)							Filter			
Transcript ID	Name 🌲	bp 🌲	Protein 🌲	Biotype	CCDS	UniProt Match	Flags			9
ENSMUST00000047864.11	Eef2-201	3089	<u>858aa</u>	Protein coding	CCDS35993 ₽	P58252 ₺	Ensembl Canonical	GENCODE basic	APPRIS P1	TSL:1
ENSMUST00000219497.2	Eef2-202	651	No protein	Retained intron		-	TSL:3			
ENSMUST00000219943.2	Eef2-203	519	No protein	Retained intron		886		TSL:1		

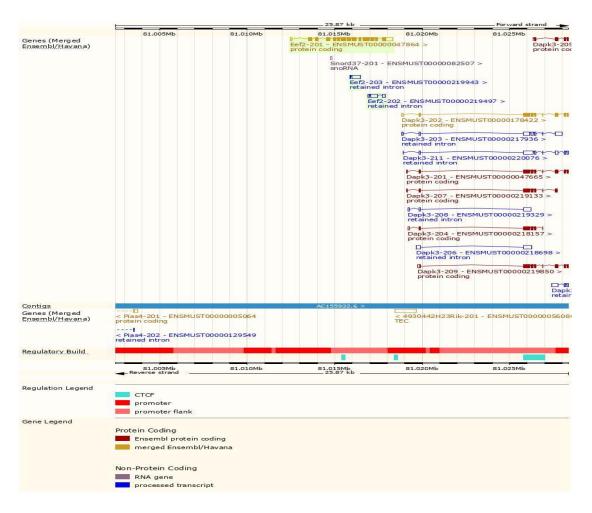
The strategy is based on the design of Eef2-201 transcript, the transcription is shown below:



Source: https://www.ensembl.org



### Genomic Information





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

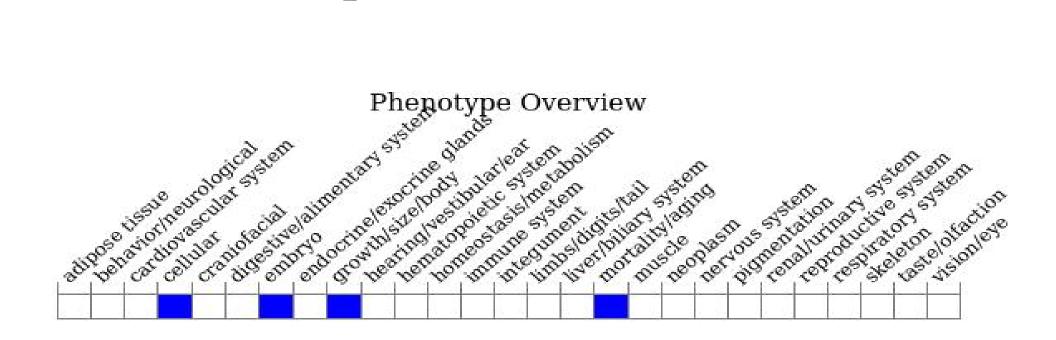
#### Protein Information





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

## Mouse Phenotype Information (MGI)



• Mice homozygous for a mutation removing the diphthamide modification display partial neonatal lethality, fetal growth retardation and abnormal cell physiology.



## Important Information

- The effect of Eef2-202, Eef2-203 is unknown.
- This Strategy may effect the N-terminal regulation of *Snord37* and *Dapk3*.
- The loxp which insert in intron 3-4 may affects the splicing function.
- *Eef2* is located on Chr10. 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.

