Efs Cas9-KO Strategy

Designer: Ruirui Zhang

Reviewer: Huimin Su

Design Date: 2020-2-18

Project Overview



Project Name

Project type Cas9-KO

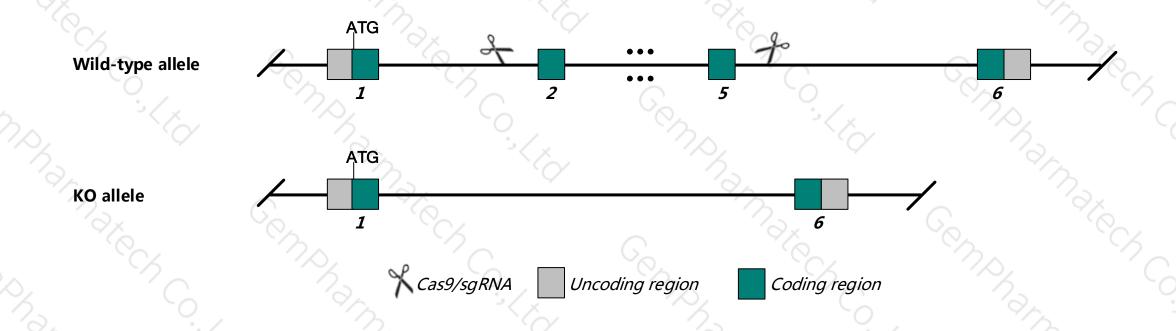
Strain background C57BL/6JGpt

Efs

Knockout strategy



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



Technical routes



- The *Efs* gene has 3 transcripts. According to the structure of *Efs* gene, exon2~exon5 of *Efs*-201 (ENSMUST00000022813.7) transcript is recommended as the knockout region. The region contains most of coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Efs* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.

Notice



According to the existing MGI data, mice homozygous for a disruption in this gene display an increased inflammatory response characterized by excessive T cell responses, enhanced cytokine secretion and antibody production, and intestinal, kidney, liver, and lung inflammation.

- > The KO region deletes most of the coding sequence, but does not result in frameshift.
- The *Efs* gene is located on the Chr14. 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)



Efs embryonal Fyn-associated substrate [Mus musculus (house mouse)]

Gene ID: 13644, updated on 12-Aug-2019

Summary



Official Symbol Efs provided by MGI

Official Full Name embryonal Fyn-associated substrate provided by MGI

Primary source MGI:MGI:105311

See related Ensembl:ENSMUSG00000022203

Gene type protein coding
RefSeq status VALIDATED
Organism <u>Mus musculus</u>

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

Murinae; Mus; Mus

Expression Broad expression in limb E14.5 (RPKM 32.5), ovary adult (RPKM 23.9) and 18 other tissues See more

Orthologs <u>human</u> <u>all</u>

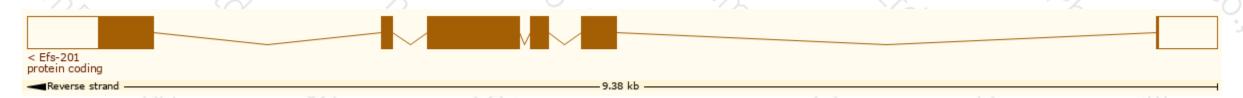
Transcript information (Ensembl)



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

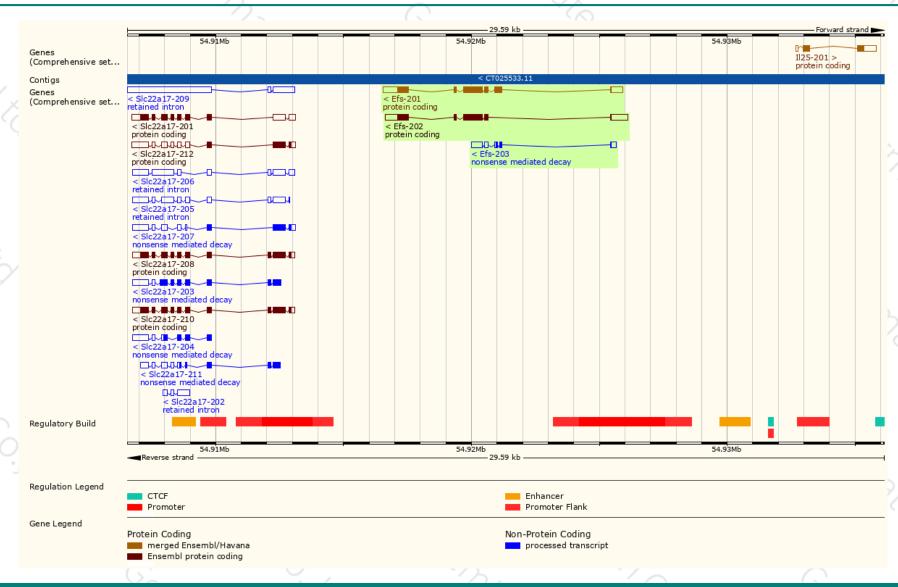
Name 🍦	Transcript ID	bp 🏺	Protein 🍦	Biotype	CCDS 🍦	UniProt 🝦	Flags
Efs-201	ENSMUST00000022813.7	2705	<u>560aa</u>	Protein coding	CCDS36925 ₺	<u>Q64355</u> ₽	TSL:1 GENCODE basic APPRIS P1
Efs-202	ENSMUST00000227037.1	2540	<u>467aa</u>	Protein coding	-	A0A2I3BRP5 ₺	GENCODE basic
Efs-203	ENSMUST00000227587.1	961	<u>45aa</u>	Nonsense mediated decay	-	A0A2I3BQ52₺	-

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



Genomic location (Ensembl)





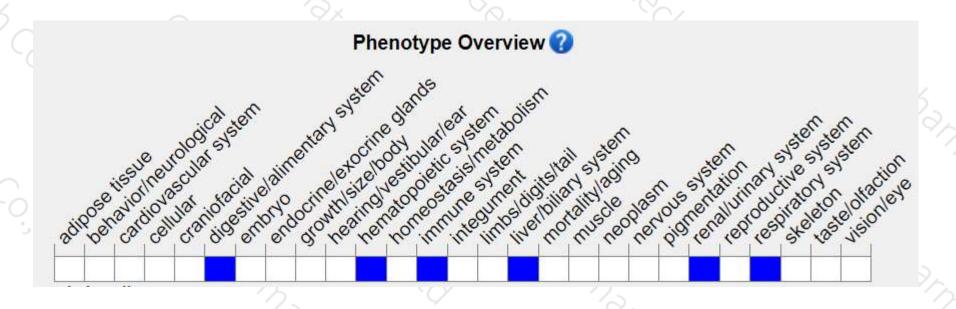
Protein domain (Ensembl)





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 homozygous for a disruption in this gene display an increased inflammatory response characterized by excessive T cell responses, enhanced cytokine secretion and antibody production, and intestinal, kidney, liver, and lung inflammation.

If you have any questions, you are welcome to inquire. Tel: 025-5864 1534





