

Fga Cas9-CKO Strategy

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Overview

Target Gene Name

• Fga

Project Type

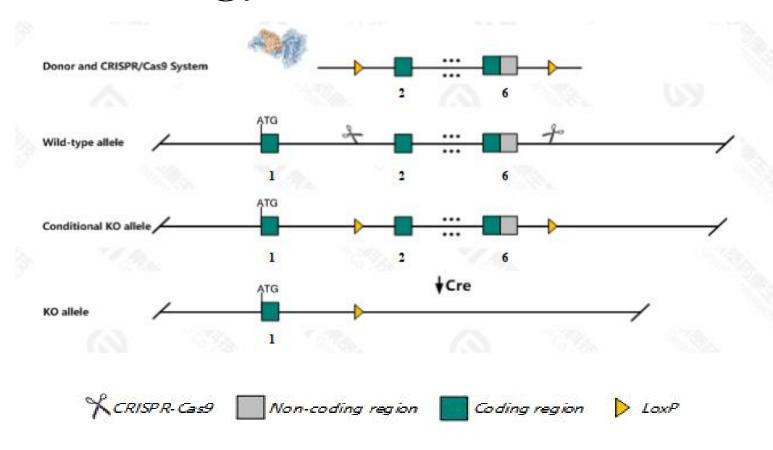
• Cas9-CKO

Genetic Background

• C57BL/6JGpt



Strain Strategy



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



Technical Information

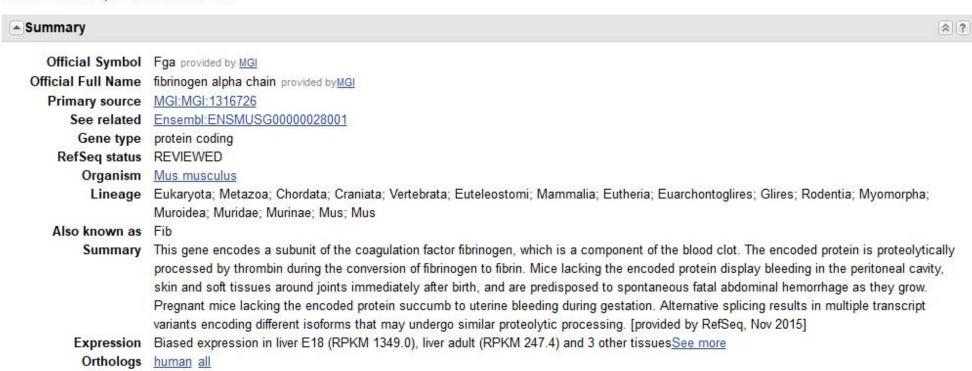
- The Fga gene has 2 transcripts. According to the structure of Fga gene, exon2-exon6 of Fga-202(ENSMUST00000166581.4) transcript is recommended as the knockout region. The region contains most of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Fga* 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.
- 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

Fga fibrinogen alpha chain [Mus musculus (house mouse)]

Gene ID: 14161, updated on 13-Mar-2020



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

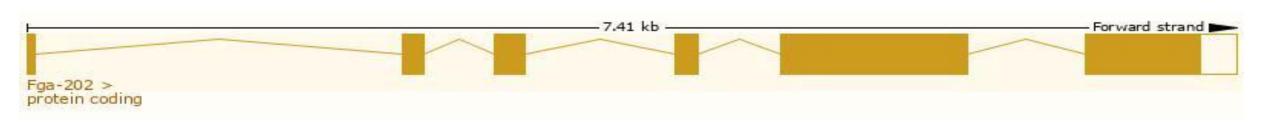


Transcript Information

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

Show/hide columns (1 hidden)							Filter
Transcript ID 👙	Name 🌲	bp 🌲	Protein .	Biotype A	CCDS 🍦	UniProt Match	Flags
ENSMUST00000166581.4	Fga-202	2587	789aa	Protein coding	CCDS50939@	E9PV24-1 ₺	Ensembl Canonical GENCODE basic APPRIS P1 TSL:5
ENSMUST00000029630.15	Fga-201	2083	<u>557aa</u>	Protein coding	CCDS17431 ₽	E9PV24-2 €	GENCODE basic TSL:1

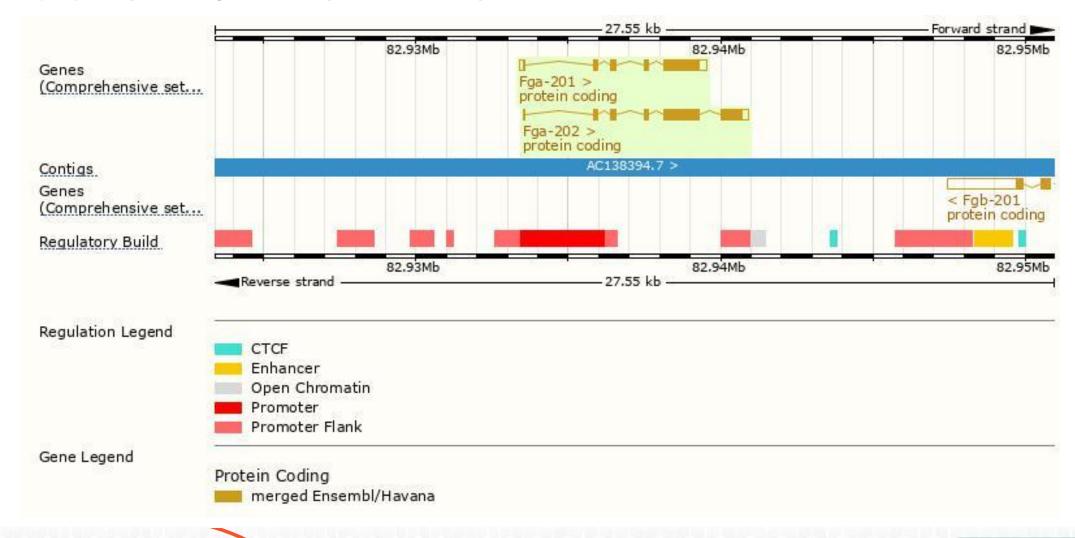
The strategy is based on the design of Fga-202 transcript, the transcription is shown below:



Source: https://www.ensembl.org



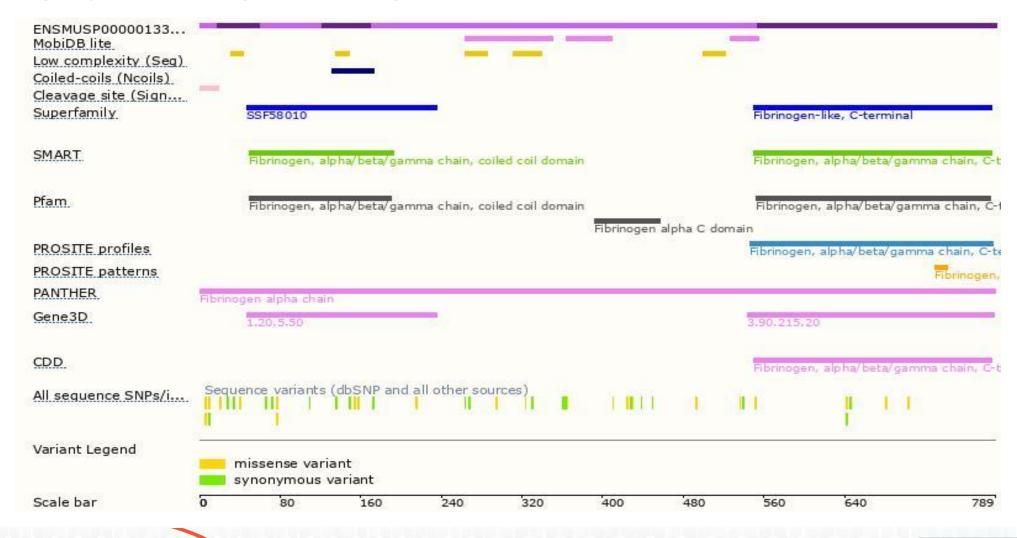
Genomic Information





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

Protein Information

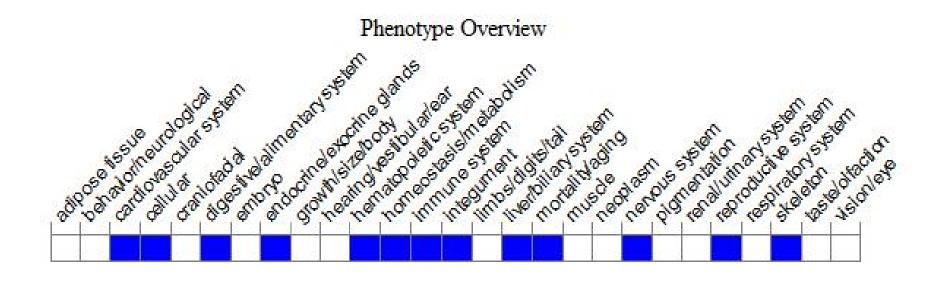




Source: : https://www.ensembl.org

Mouse Phenotype Information (MGI)

• Mice homozygous for disruptions of this gene have blood that is unable to clot. On some genetic backgrounds this can lead to fatal hemorrhaging.





Important Information

- According to the existing MGI data, mice homozygous for disruptions of this gene have blood that is unable to clot. On some genetic backgrounds this can lead to fatal hemorrhaging.
- The Fga gene is located on the Chr 3. 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.

