

Flt1 Cas9-CKO Strategy

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Reviewer:

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Design Date:

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Project Overview



Project Name

Flt1

Project type

Cas9-CKO

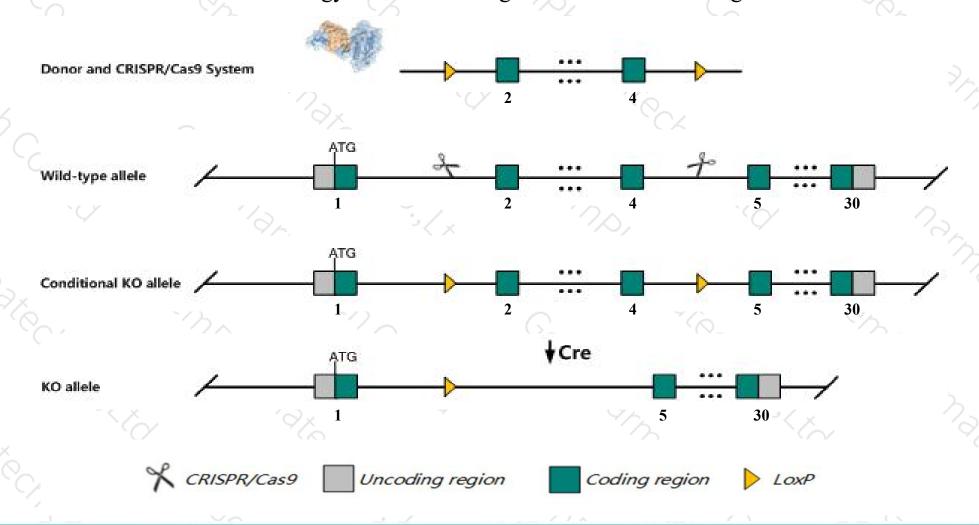
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Flt1 gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Flt1* gene has 6 transcripts. According to the structure of *Flt1* gene, exon2-exon4 of *Flt1-202*(ENSMUST00000031653.11) transcript is recommended as the knockout region. The region contains 452bp coding sequence.

 Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Flt1* 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 sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.
- 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.

Notice



- ➤ According to the existing MGI data, Homozygotes for targeted null mutations exhibit an excess of hemangioblasts resulting in an overgrowth of endothelial cells, abnormalities of vascular channels and blood islands, and lethality at the mid-somite developmental stage.
- > The *Flt1* 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)



FIt1 FMS-like tyrosine kinase 1 [Mus musculus (house mouse)]

Gene ID: 14254, updated on 3-Mar-2019

Summary

↑ ?

Official Symbol Flt1 provided by MGI

Official Full Name FMS-like tyrosine kinase 1 provided by MGI

Primary source MGI:MGI:95558

See related Ensembl: ENSMUSG00000029648

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 Al323757, Fit-1, VEGFR-1, VEGFR1, sFit1

Expression Biased expression in placenta adult (RPKM 35.7), lung adult (RPKM 11.8) and 11 other tissuesSee more

Orthologs human all

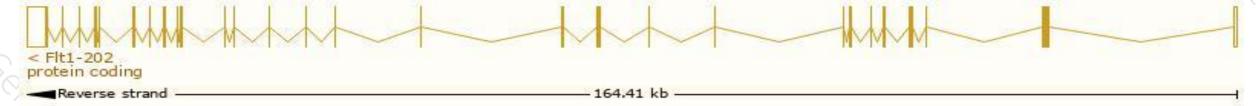
Transcript information (Ensembl)



The gene has 6 transcripts, all transcripts are shown below:

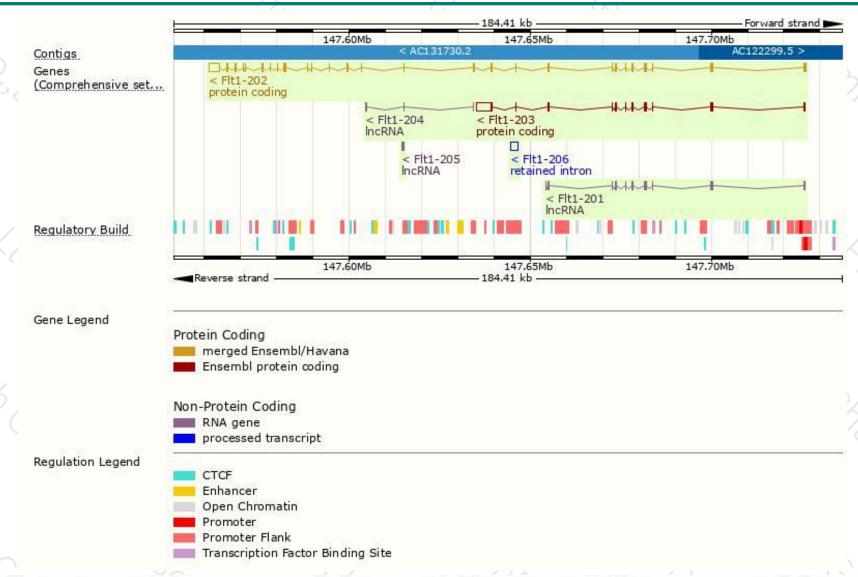
Name	Transcript ID	bp	Protein	Biotype	ccps	UniProt	Flags
Flt1-202	ENSMUST00000031653.11	6895	1333aa	Protein coding	CCDS19879	A0A0R4J0A4	TSL:1 GENCODE basic APPRIS P1
FIt1-203	ENSMUST00000110529.5	6292	688aa	Protein coding	-	<u>055095</u>	TSL:1 GENCODE basic
FIt1-206	ENSMUST00000200895.1	2030	No protein	Retained intron	ů.	127	TSL:NA
FIt1-201	ENSMUST00000031652.4	2049	No protein	IncRNA	2	3528	TSL:1
FIt1-204	ENSMUST00000125272.1	395	No protein	IncRNA		(25)	TSL:5
FIt1-205	ENSMUST00000142477.1	274	No protein	IncRNA		393	TSL:2

The strategy is based on the design of Flt1-202 transcript, The transcription is shown below



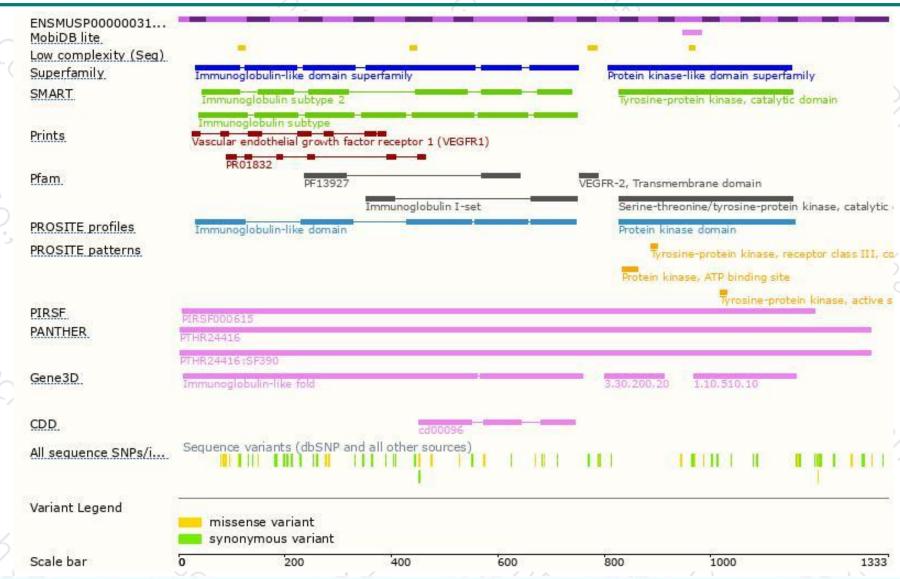
Genomic location distribution





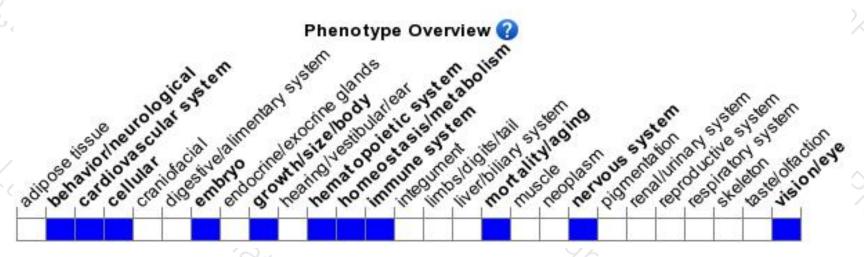
Protein domain





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, Homozygotes for targeted null mutations exhibit an excess of hemangioblasts resulting in an overgrowth of endothelial cells, abnormalities of vascular channels and blood islands, and lethality at the mid-somite developmental stage.



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





