

Fzd5-iCre TG Strategy

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Reviewer

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Design Date:

2019-8-22

Project Overview

Project Name

Fzd5-iCre

Project type

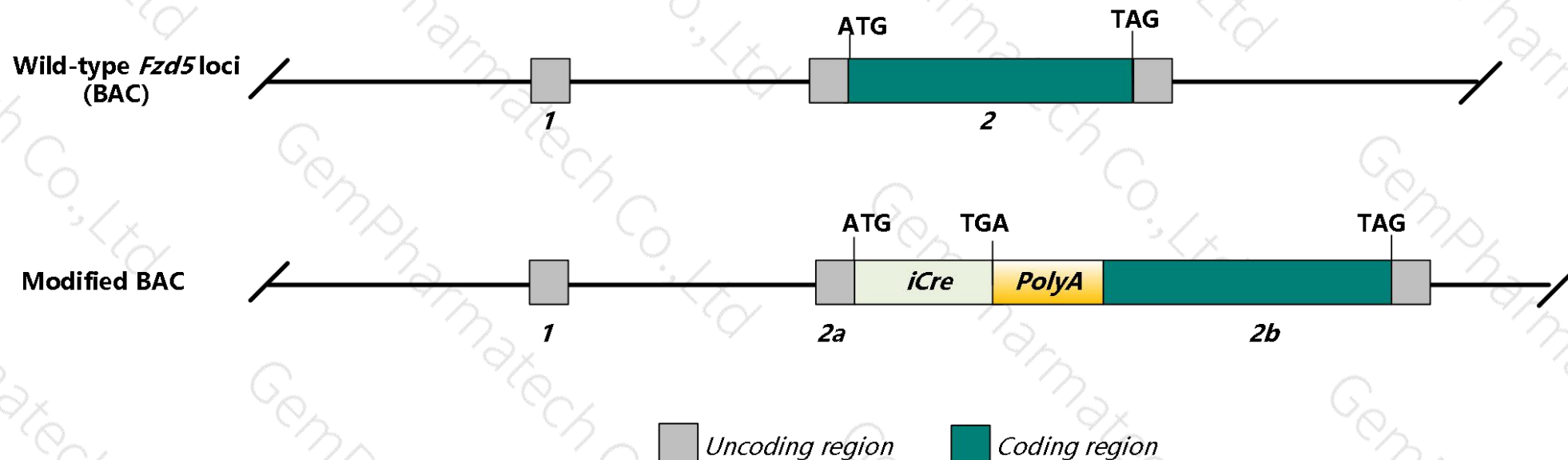
BAC-TG

Strain background

C57BL/6J

Knockin strategy

This model will use pronucleus injection technology to obtain the *Fzd5-iCre* model. The schematic diagram is as follows:



- Transcript *Fzd5*-201(ENSMUST00000063982.6) is selected for presentation of the recommended strategy.
- *Fzd5*-201 gene has 2 exons, with the ATG start codon at exon 2 and TAG stop codon at exon 2.
- In this study, RP23-304D20 (~207kb) or RP23-153G19 (~191kb) of C57BL/6J mouse bacterial artificial chromosome (BAC) containing the entire *Fzd5* locus (and other genes), was modified by targeting iCre-polyA sequence to the exon2 near the translation start codon of the *Fzd5* locus, ensuring iCre is expressed from the endogenous promoter/enhancer elements of *Fzd5*. transgenic fragments containing *Fzd5-iCre-polyA* were micro-injected into the fertilized eggs of C57BL/6J mice, and obtained positive F0 generation (i.e., founder) mice.

- According to the MGI, Mice homozygous for disruption of this gene die as embryos. Extra embryonic vascular development is abnormal.
- Cre-mediated recombination is mainly expressed in the retina, which can be detected in embryonic E16.5.
- While expressing iCre, other genes coexisting on BAC will also be expressed together.
- Transgene fragment will be injected into the fertilized eggs, and randomly integrated into the genome, by the influence of insertion site and copy number, expression level of the transgenic mice may be different.
- This strategy is designed based on genetic information in existing databases. Due to the complexity of gene transcription and translation processes, all risks cannot be predicted under existing information.

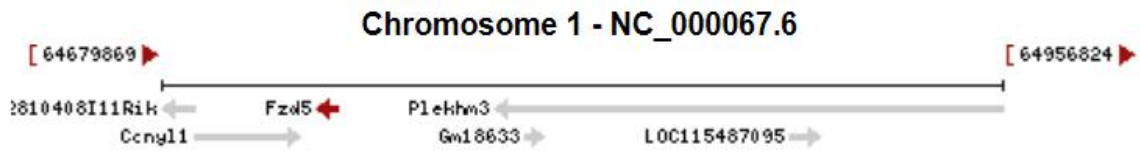
Gene information (NCBI)

Fzd5 frizzled class receptor 5 [*Mus musculus* (house mouse)]

Gene ID: 14367, updated on 21-Aug-2019

Summary

Official Symbol	Fzd5 provided by MGI
Official Full Name	frizzled class receptor 5 provided by MGI
Primary source	MGI:MGI:108571
See related	Ensembl:ENSMUSG00000045005
Gene type	protein coding
RefSeq status	VALIDATED
Organism	<i>Mus musculus</i>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Fz5; Fz-5; mFz5; AI427138; 5330434N09Rik
Expression	Broad expression in colon adult (RPKM 30.4), large intestine adult (RPKM 20.0) and 15 other tissues See more
Orthologs	human all

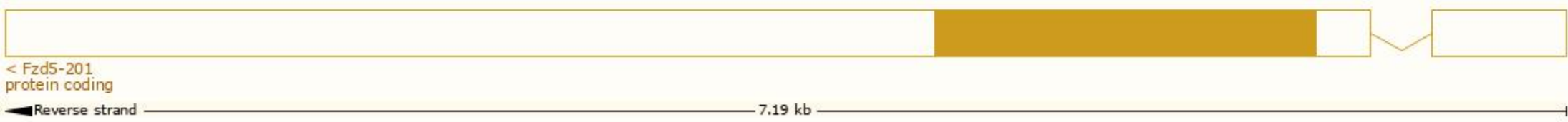


Transcript information (Ensembl)

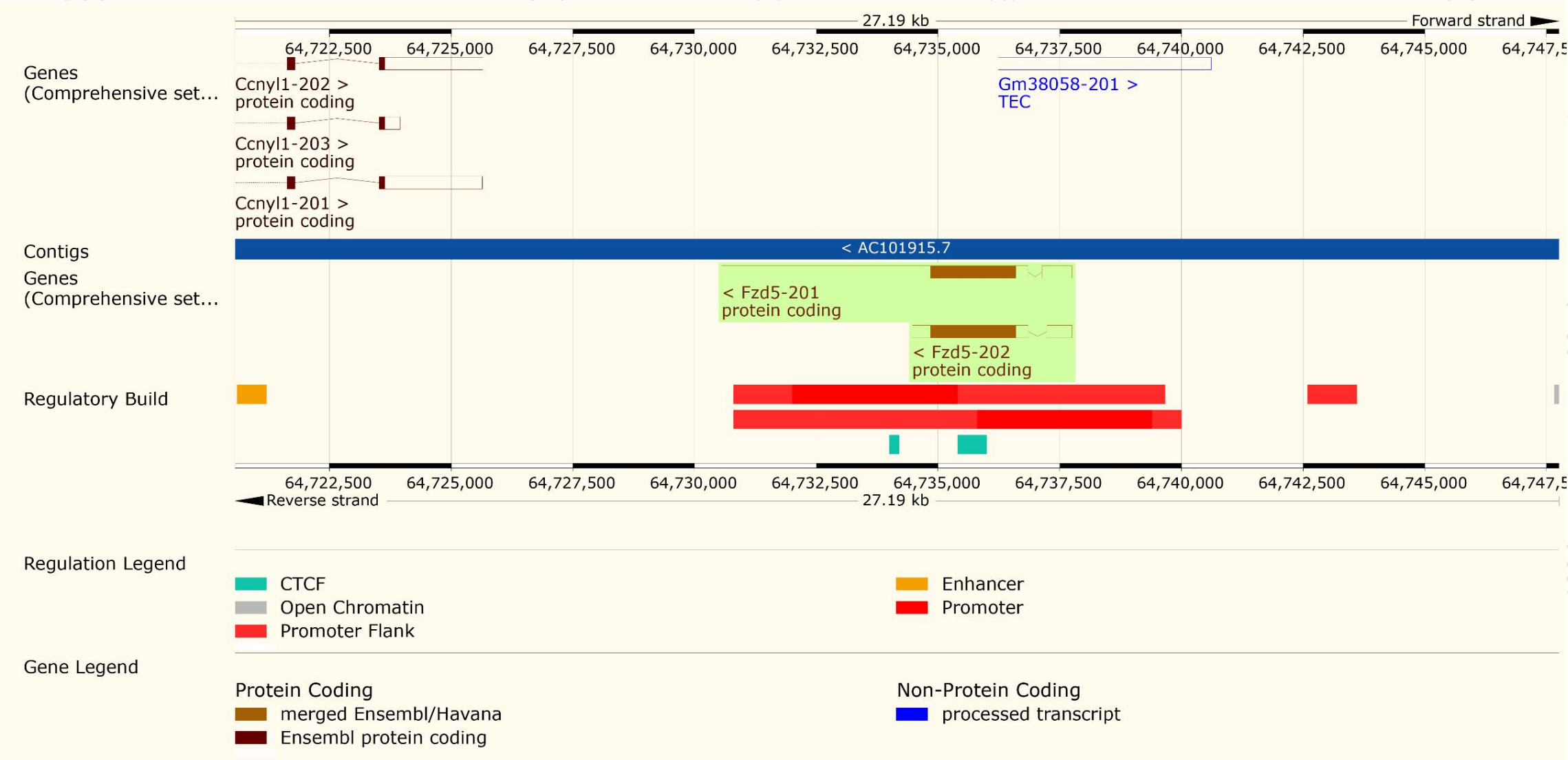
The gene has 2 transcripts, and all transcripts are shown below:

Fzd5-201	ENSMUST00000063982.6	6915	585aa	ENSMUSP00000067783.5	Protein coding	CCDS15008	Q9EQD0	TSL:1	GENCODE basic	APPRIS P1
Fzd5-202	ENSMUST00000116133.3	2895	585aa	ENSMUSP00000111828.2	Protein coding	CCDS15008	Q9EQD0	TSL:1	GENCODE basic	APPRIS P1

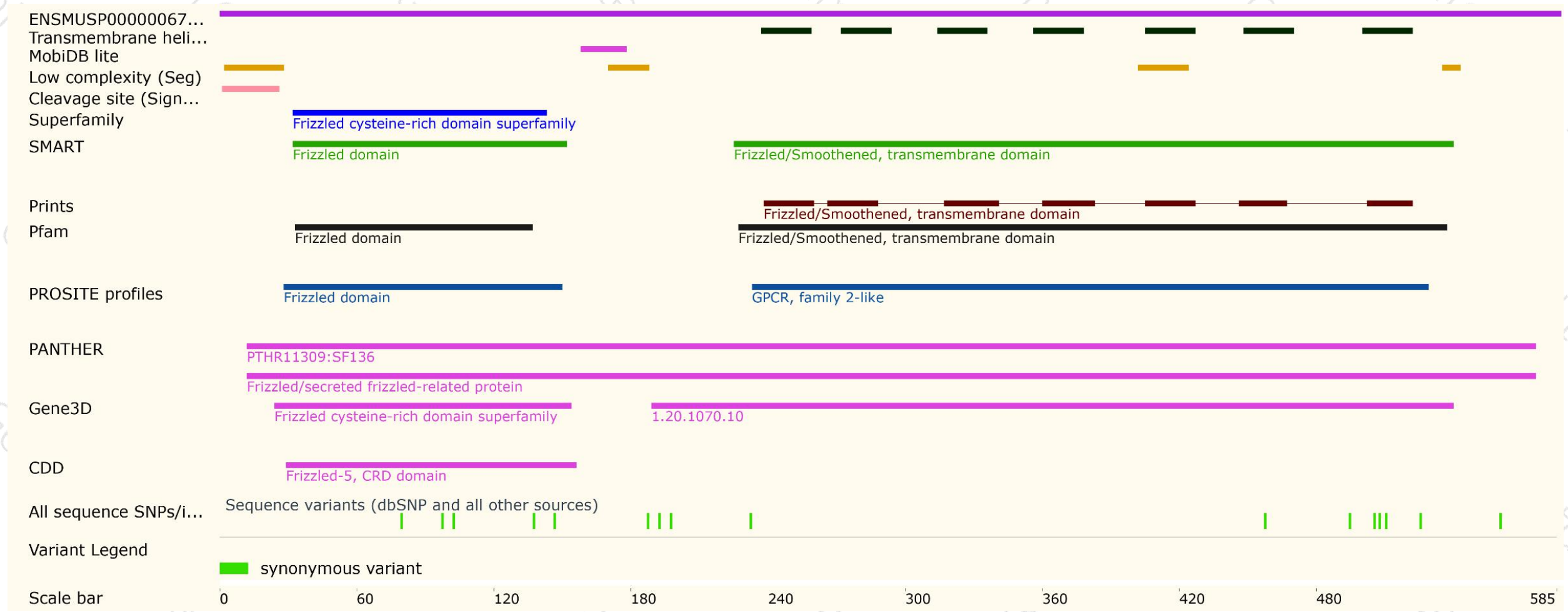
The strategy is based on the design of *Fzd5-201* transcript, the transcription is shown below:



Genomic location distribution

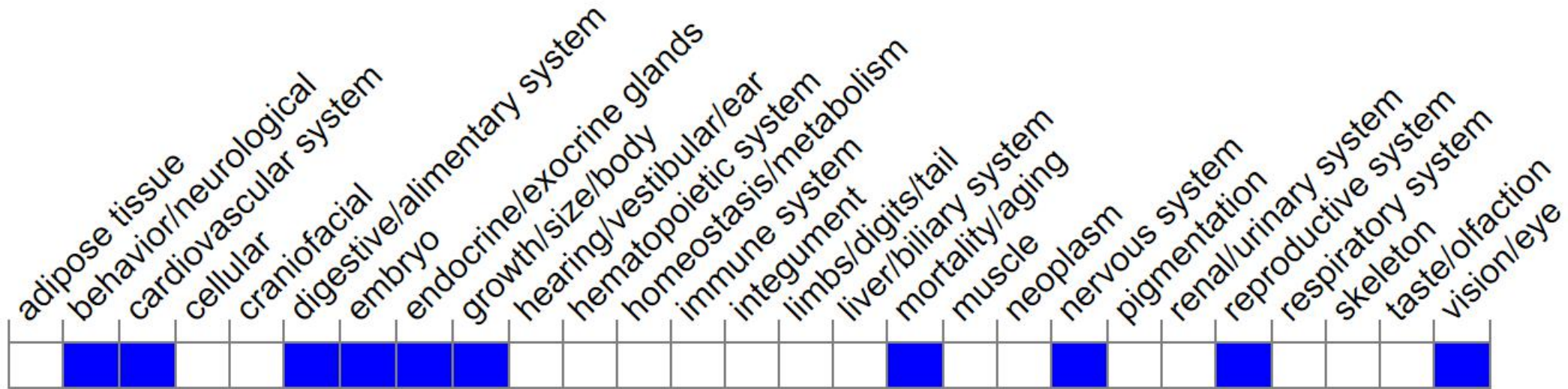


Protein domain



Mouse phenotype description(MGI)

Phenotype Overview ?



Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(<http://www.informatics.jax.org/marker/MGI:108571>) .

According to the existing MGI data, Mice homozygous for disruption of this gene die as embryos. Extra embryonic vascular development is abnormal.

Coding Sequence of Codon-Optimized Cre Gene^[1]



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ATGGTGCCCAAGAAGAAGAGGAAAGTCTCCAACCTGCTGACTGTGCACCAAAACCTGCCTGCCCTCCCTGTGGATGCCACCTCTGATGAAGTCAGGAAGA
ACCTGATGGACATGTTTCAGGGACAGGCAGGCCTTCTCTGAACACACCTGGAAGATGCTCCTGTCTGTGTGCAGATCCTGGGCTGCCTGGTGCAAGCTGAA
CAACAGGAAATGGTTCCCTGCTGAACCTGAGGATGTGAGGGACTACCTCCTGTACCTGCAAGCCAGAGGCCTGGCTGTGAAGACCATCCAACAGCACCTG
GGCCAGCTCAACATGCTGCACAGGAGATCTGGCCTGCCTCGCCCTTCTGACTCCAATGCTGTGTCCCTGGTGATGAGGAGAATCAGAAAGGAGAATGTGG
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GAACCTGGCCTTCCTGGGCATTGCCTACAACACCCTGCTGCGCATTGCCGAAATTGCCAGAATCAGAGTGAAGGACATCTCCCGCACCGATGGTGGGAGA
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GGCCCTGGAAGGGATCTTTGAGGCCACCCACCGCCTGATCTATGGTGCCAAGGATGACTCTGGGCAGAGATACCTGGCCTGGTCTGGCCACTCTGCCAGA
GTGGGTGCTGCCAGGGACATGGCCAGGGCTGGTGTGTCCATCCCTGAAATCATGCAGGCTGGTGGCTGGACCAATGTGAACATTGTGATGAACTACATCA
GAAACCTGGACTCTGAGACTGGGGCCATGGTGAGGCTGCTCGAGGATGGGGACTGA

References

[1] Shimshek DR, Kim J, Hübner MR, Spergel DJ. Codon-improved Cre recombinase (iCre) expression in the mouse.

GeAlbis.2002 Jan.32(1):19-26.

If you have any questions, you are welcome to inquire.
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