

Fanc1 Cas9-CKO Strategy

Designer: Shuang Zhang

Reviewer: Yun Li

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Project Overview

Project Name

Fancl

Project type

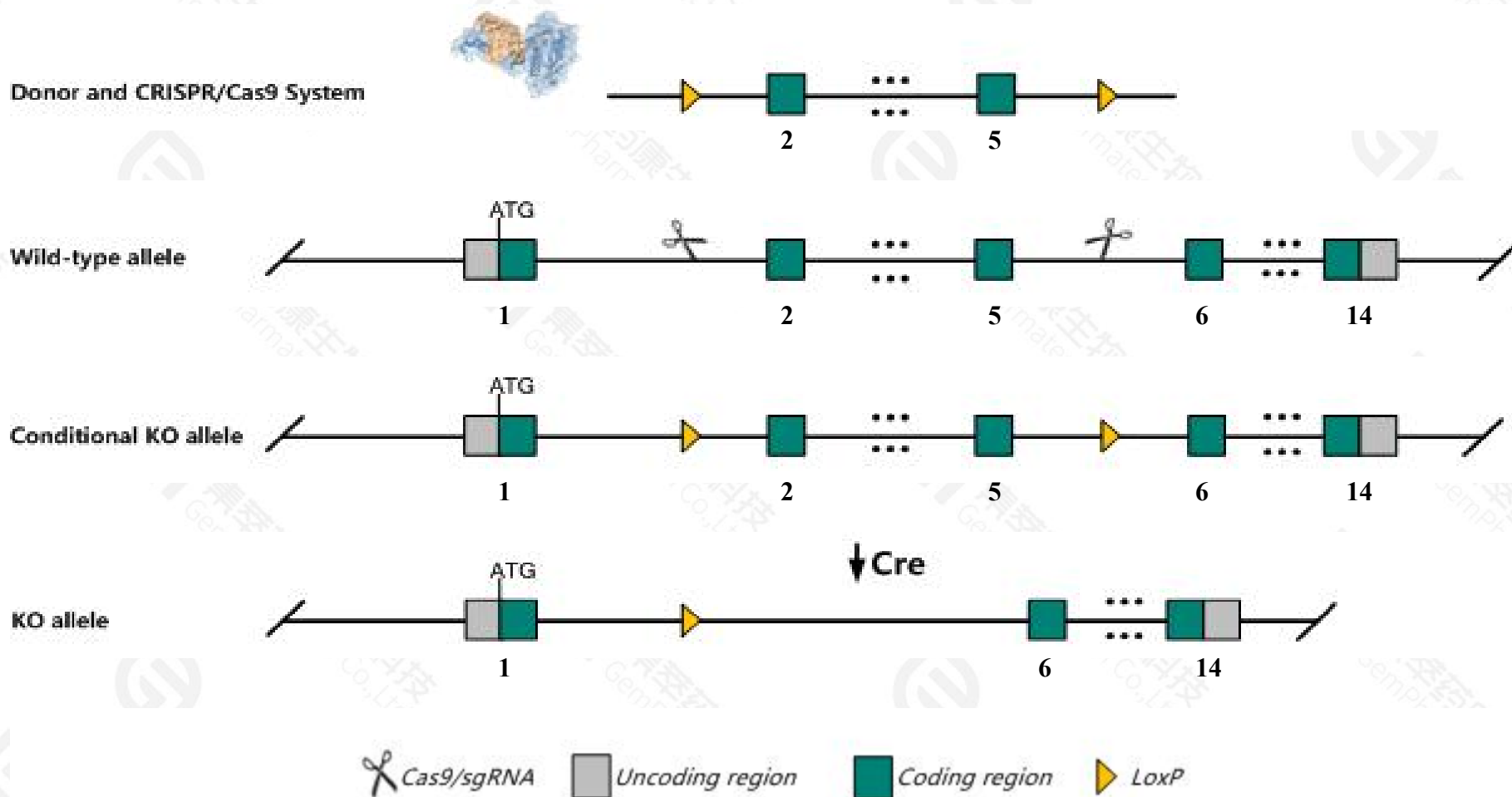
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Fancl* gene has 5 transcripts. According to the structure of *Fancl* gene, exon2-exon5 of *Fancl*-201(ENSMUST00000004120.9) transcript is recommended as the knockout region. The region contains 278bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Fancl* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor was constructed. Cas9, sgRNA 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 was 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.

- According to the existing MGI data, homozygosity for mutations that inactivate the allele results in male and female infertility due to defects in primordial germ cell proliferation. Homozygosity is embryonic lethal on some backgrounds.
- Transcript *Fancl-203* may not be affected.
- The *Fancl* gene is located on the Chr11. 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)

Fancl Fanconi anemia, complementation group L [Mus musculus (house mouse)]

Gene ID: 67030, updated on 17-Nov-2020

Summary

Official Symbol Fancl provided by [MGI](#)

Official Full Name Fanconi anemia, complementation group L provided by [MGI](#)

Primary source [MGI:MGI:1914280](#)

See related [Ensembl:ENSMUSG00000004018](#)

Gene type protein coding

RefSeq status REVIEWED

Organism [Mus musculus](#)

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

Also known as 2010322C19Rik, AW554273, B230118H11Rik, P, Phf, Phf9, Pog, gcd

Summary This gene encodes the complementation group L subunit of the multimeric Fanconi anemia (FA) nuclear complex composed of proteins encoded by over ten Fanconi anemia complementation (FANC) group genes. The FA complex is necessary for protection against DNA damage. This gene product, an E3 ubiquitin ligase, catalyzes and is required for the monoubiquitination of the protein encoded by the Fanconi anemia, complementation group D2 gene, a critical step in the FA pathway (PMID: 12973351, 21229326). In mouse, mutations of this E3 ubiquitin ligase gene can lead to infertility in adult males and females, and a deletion of this gene can cause embryonic lethality in some genetic backgrounds. A pseudogene of this gene has been identified on chromosome 1. Alternative splicing results in multiple transcript variants. [provided by RefSeq, Mar 2013]

Expression Ubiquitous expression in liver E14 (RPKM 7.9), CNS E11.5 (RPKM 7.5) and 26 other tissues [See more](#)

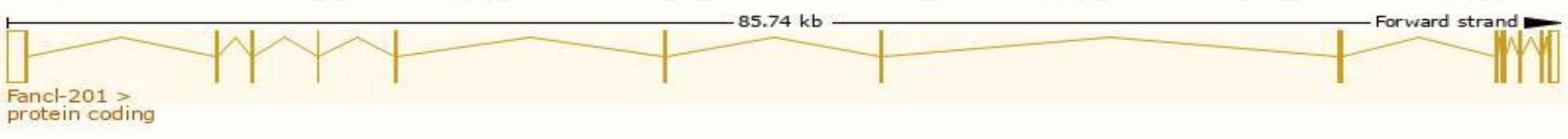
Orthologs [human](#) [all](#)

Transcript information (Ensembl)

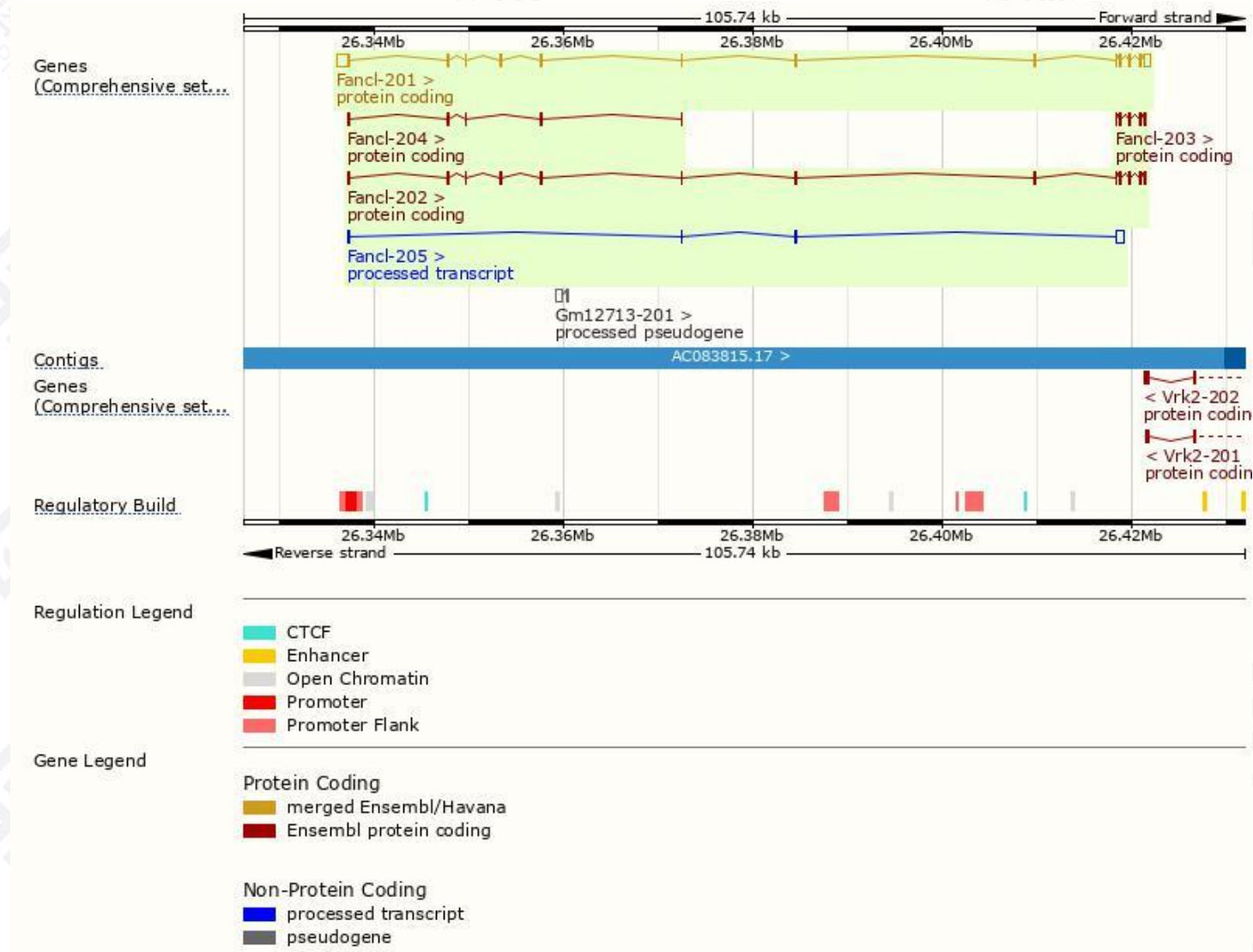
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Fancl-201	ENSMUST00000004120.9	2724	375aa	Protein coding	CCDS24485		TSL:1 , GENCODE basic , APPRIS P1 ,
Fancl-202	ENSMUST00000109509.8	1187	370aa	Protein coding	CCDS70152		TSL:1 , GENCODE basic ,
Fancl-203	ENSMUST00000134445.2	577	145aa	Protein coding	-		CDS 5' incomplete , TSL:3 ,
Fancl-204	ENSMUST00000136830.2	383	123aa	Protein coding	-		CDS 3' incomplete , TSL:3 ,
Fancl-205	ENSMUST00000143471.2	1065	No protein	Processed transcript	-		TSL:1 ,

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



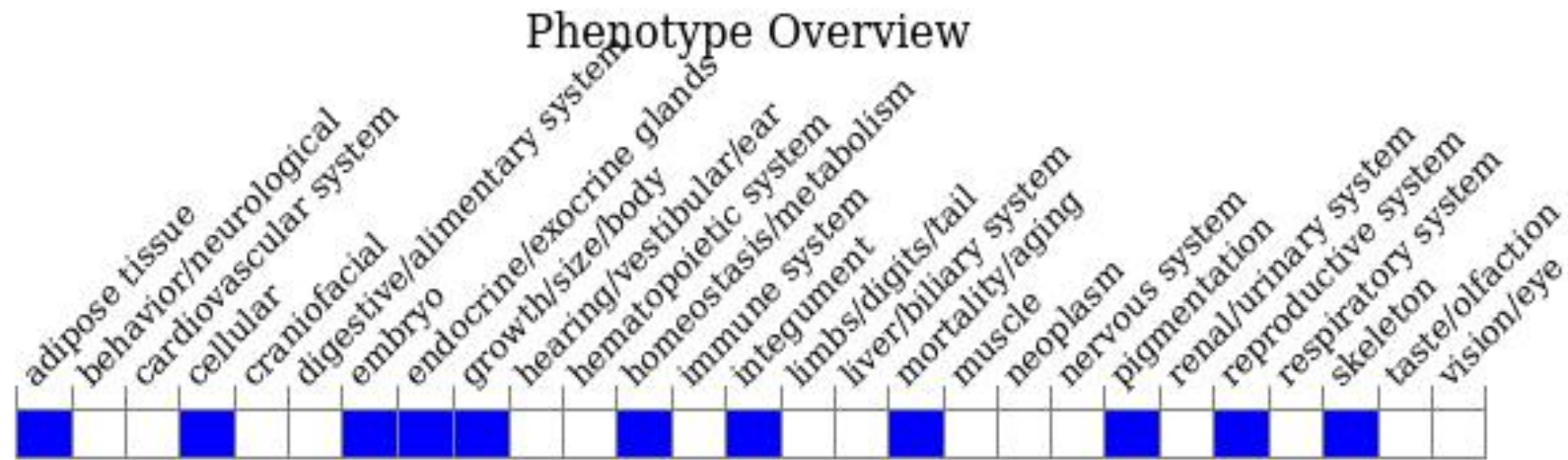
Genomic location distribution



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, homozygosity for mutations that inactivate the allele results in male and female infertility due to defects in primordial germ cell proliferation. Homozygosity is embryonic lethal on some backgrounds.

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

Tel: 025-5864 1534

