Fancm Cas9-CKO Strategy

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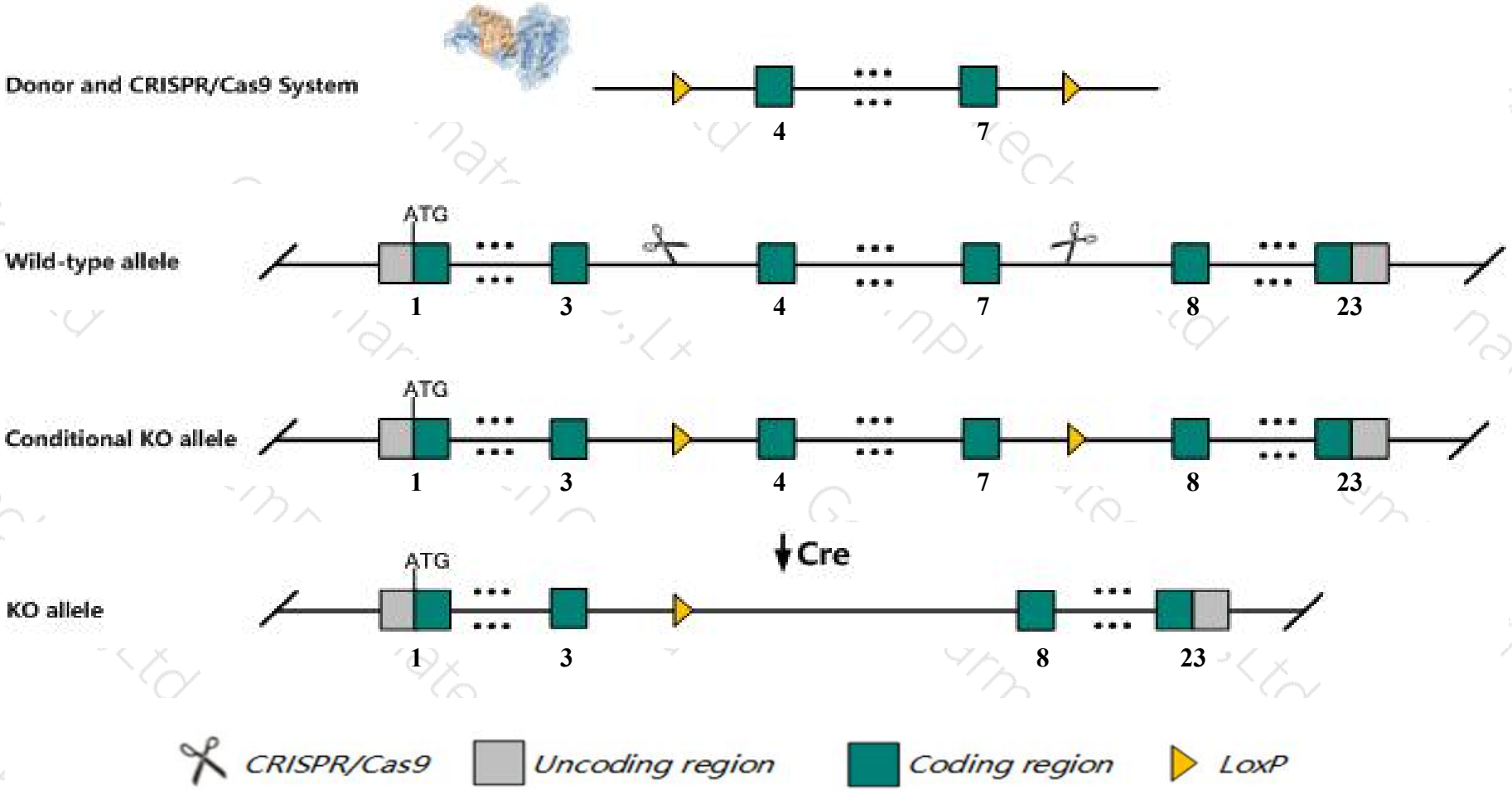
Project Overview

Project Name	***Fancm***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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



Technical routes

- The *Fancm* gene has 9 transcripts. According to the structure of *Fancm* gene, exon4-exon7 of *Fancm-201* (ENSMUST00000058889.4) transcript is recommended as the knockout region.The region contains 541bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Fancm* 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,Mice homozygous for a knock-out allele exhibit reduced female transmission, hypogonadism, premature death, and increased incidence of tumors.

- Transcript *Fancm-202,203* may not be affected.

- The *Fancm* gene is located on the Chr12. 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）

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Fancm Fanconi anemia, complementation group M [Mus musculus (house mouse)]

Gene ID: 104806, updated on 19-Mar-2019

Summary

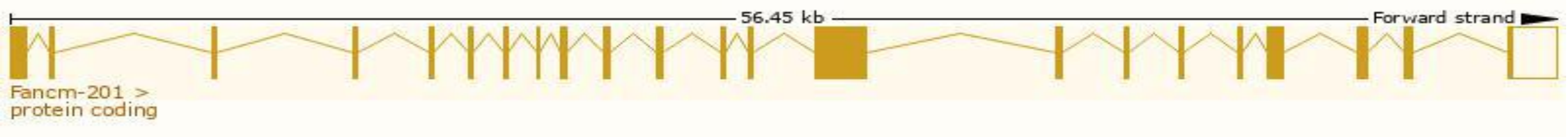
Official Symbol	Fancm provided by MGI
Official Full Name	Fanconi anemia, complementation group M provided by MGI
Primary source	MGI:MGI:2442306
See related	Ensembl:ENSMUSG00000055884
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI427100, C730036B14Rik, D12Ertd364e
Expression	Ubiquitous expression in CNS E11.5 (RPKM 3.1), CNS E14 (RPKM 2.0) and 25 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

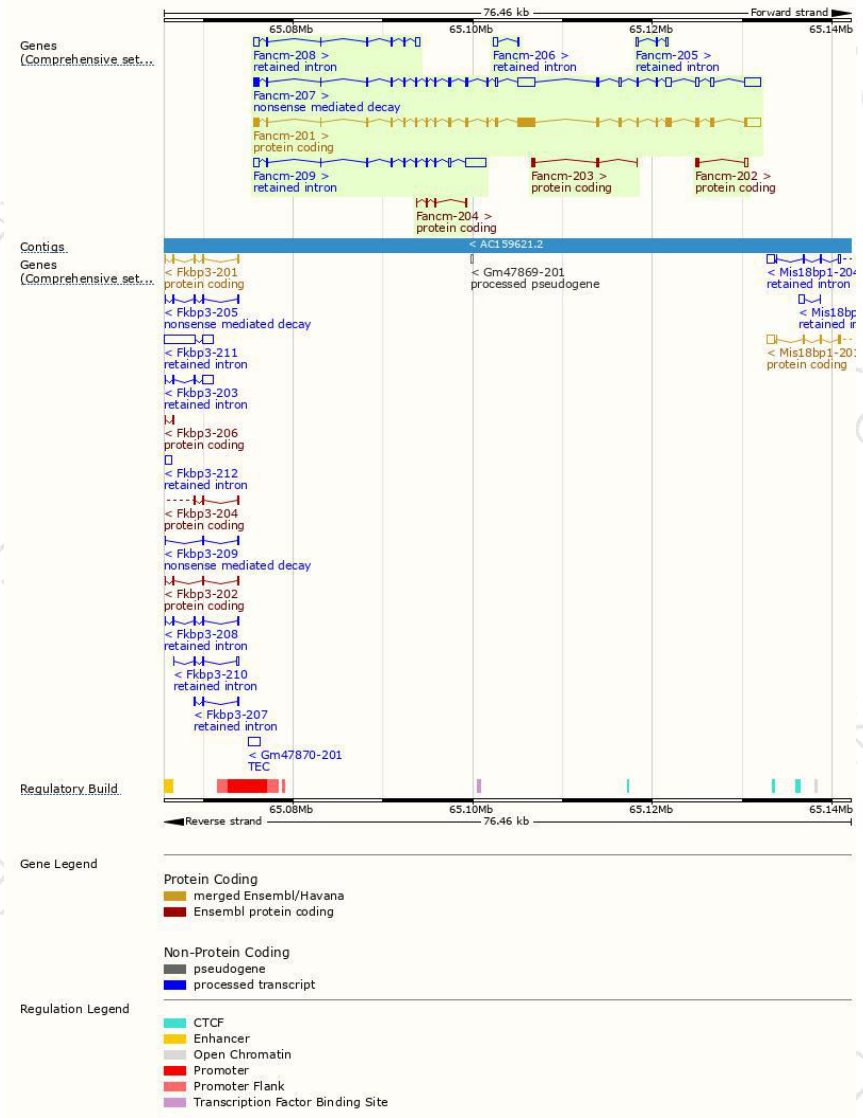
The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Fancm-201	ENSMUST00000058889.4	7775	2021aa	Protein coding	CCDS25941	Q8BGE5	TSL:1 GENCODE basic APPRIS P1
Fancm-202	ENSMUST00000221175.1	621	121aa	Protein coding	-	A0A1Y7VKY9	CDS 5' incomplete TSL:5
Fancm-203	ENSMUST00000221838.1	502	167aa	Protein coding	-	A0A1Y7VLB8	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3
Fancm-204	ENSMUST00000221937.1	325	109aa	Protein coding	-	A0A1Y7VMA3	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
Fancm-207	ENSMUST00000222540.1	7816	664aa	Nonsense mediated decay	-	A0A1Y7VKF2	TSL:1
Fancm-209	ENSMUST00000223401.1	4050	No protein	Retained intron	-	-	TSL:1
Fancm-208	ENSMUST00000223051.1	1682	No protein	Retained intron	-	-	TSL:1
Fancm-205	ENSMUST00000222086.1	563	No protein	Retained intron	-	-	TSL:3
Fancm-206	ENSMUST00000222521.1	542	No protein	Retained intron	-	-	TSL:2

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

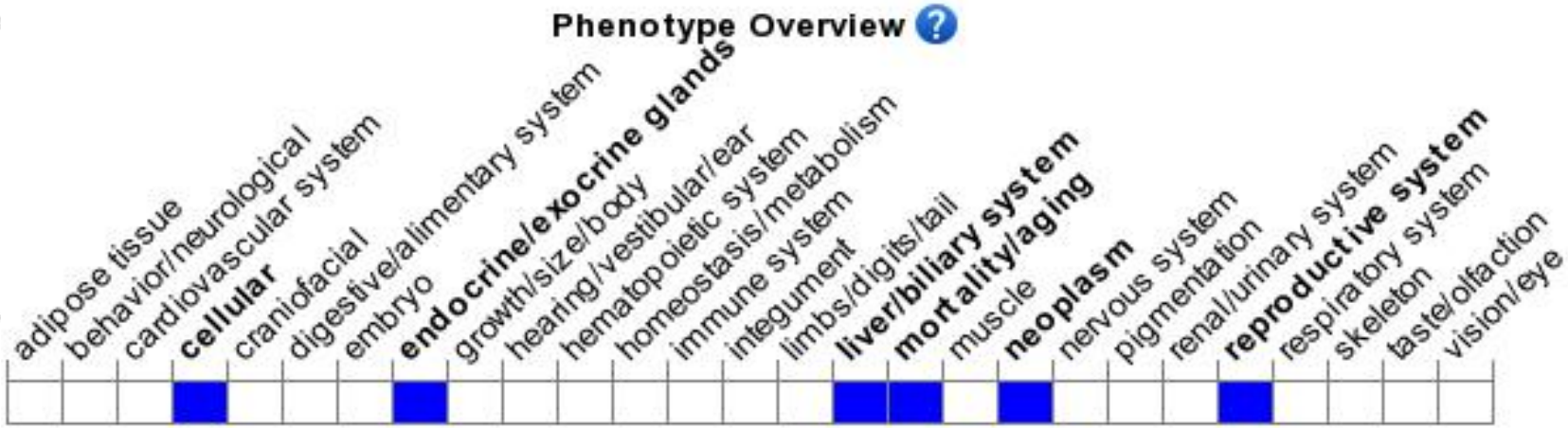


Genomic location distribution



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,Mice homozygous for a knock-out allele exhibit reduced female transmission, hypogonadism, premature death, and increased incidence of tumors.

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

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